Relating nutrient uptake with transient storage in forested mountain streams

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Abstract

Streams control the timing and delivery of fluvial nutrient export from watersheds, and hydraulic processes such as transient storage may affect nutrient uptake and transformation. Although we expect that hydraulic processes that retain water will increase nutrient uptake, the relationship between transient storage and nutrient uptake is not clear. To examine this relationship, we injected a conservative tracer and nutrients (ammonium and phosphate) into 13 streams for a total of 37 injections at Hubbard Brook Experimental Forest (HBEF), New Hampshire. Transient storage was estimated by fitting conservative solute data to a one-dimensional advection, dispersion, transient storage model. To correct for variation in depth and velocity among streams, we considered nutrient uptake as a mass-transfer coefficient (V_f), which estimates benthic demand for nutrients relative to supply. Transient storage decreased with increasing specific discharge (discharge per unit stream width). Transient storage explained only 14% of variation in ammonium V_f during the entire year and 35% of variation during summer months. Phosphate uptake was not related to transient storage, presumably because P uptake is predominantly by chemical sorption at HBEF. At HBEF, surface water pools can store water but were not modeled as such by use of the transient storage model. These pools were probably not important areas of nutrient uptake; further variation in the relationship between nutrient uptake and transient storage may be explained by biological demand.

Streams are important landscape features because they provide an avenue for nutrient loss from the terrestrial landscape and subsequent delivery to downstream ecosystems. Streams are not simply conduits, however, because they alter the form and amounts of nutrients through uptake and transformation of dissolved and particulate forms (Burns 1998; Fisher et al. 1998; Alexander et al. 2000). The pattern of element loss observed from forests (e.g., Likens and Bormann 1995) may, in part, be a function of in-stream processes (Hall et al. 2001). One way to examine how in-stream processes affect nutrient transport is to measure nutrient uptake length, which is the average distance downstream traveled by a nutrient atom before being removed from the water. This measure indicates the degree of retentiveness for a given element (Newbold et al. 1981, 1983). Essentially, a small amount of nutrient or isotopic tracer is added to a stream to estimate uptake relative to a nonreactive hydrologic tracer. This approach has been used to describe stream nutrient dynamics and has effectively demonstrated the importance of streams in processing nutrients (Newbold et al. 1981; Munn and Meyer 1990; Mulholland et al. 1997).

To understand the role of in-stream processes in determining watershed nutrient export, we need to estimate what factors control uptake length in streams. Uptake length varies among streams, and this variation may be caused by hydrologic, geomorphological, and biological processes. For example, streams with greater depth and velocity will have longer uptake lengths, and indeed most of the variation in uptake length can be a function of these two components of discharge (Valett et al. 1996; Butterini and Sabater 1998). Nonetheless, many studies use uptake length as a measure of nutrient uptake, but because uptake length is sensitive to stream discharge, it is difficult to compare lengths among different size streams. One way to correct for this effect is to calculate a mass-transfer coefficient of uptake (Stream Solute Workshop 1990; Davis and Minshall 1999), which represents demand for nutrients relative to supply in the water column.
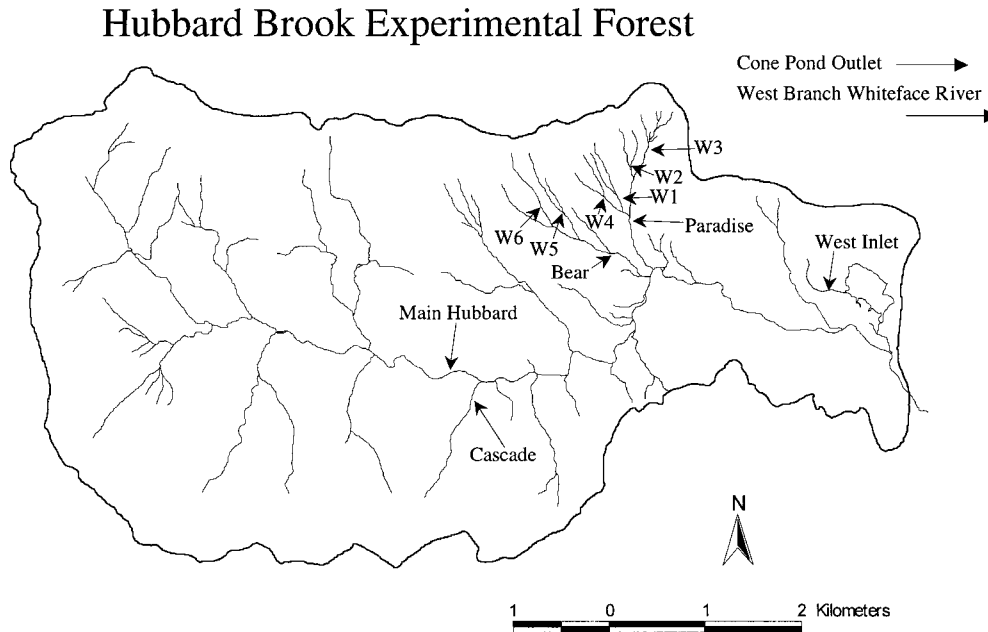
Hubbard Brook Experimental Forest

Fig. 1.   Map of HBEF and the study streams. The Cone Pond outlet and West Branch of White-face river are located east of HBEF and are not shown on this map.

Beyond discharge, stream geomorphology and hydraulics may also influence nutrient uptake, and transient storage in particular has received much recent attention (Valett et al. 1996; Mulholland et al. 1997; Butterini and Sabater 1999). Transient storage is water that is moving more slowly than water in the channel, and these storage zones can be hypor-heic or surface areas (Harvey et al. 1996). Transient storage is measured by adding a conservative tracer that integrates flows into transient storage zones; the effect of these flows will modify the change in solute concentration with time at some point downstream of the tracer addition. By fitting the conservative solute time series to an advection-dispersion model that accounts for storage of water in transient storage, the size of transient storage can be estimated for an entire reach (Bencala and Walters 1983).

Because transient storage and nutrient uptake are reach-scale measurements, they can be easily compared to examine whether water exchange with transient storage affects nutrient uptake. Modeling suggests that transient storage may increase nutrient uptake (De Angelis et al. 1995; Mulholland and DeAngelis 2000), but empirical studies are equivocal. Using six addition experiments, Valett et al. (1996) showed that uptake length and transient storage were related, but both covaried with discharge, so it was difficult to infer cause. Mulholland et al. (1997) suggested that high transient storage lowered nutrient uptake length, but they compared only one measurement from each of two streams. In contrast, size of the transient storage zone appeared to have no effect on nutrient uptake length in Sycamore Creek, Arizona, despite large changes in transient storage after recovery from a flood (Martí et al. 1997).

In the present article, we examine the relationship between transient storage and ammonium and phosphate uptake in forested headwater streams in New Hampshire. Because other studies have not shown a conclusive relationship between transient storage and nutrient uptake, we modified the approach in two ways. One is that we focused on the mass-transfer coefficient of nutrient uptake (Davis and Minshall 1999). Most studies that have examined relationships between transient storage and nutrient uptake have focused solely on uptake length, which often is simply a function of discharge. In order to compare nutrient uptake among different size streams, it is necessary to correct for the overriding effect of discharge on nutrient uptake length. Calculating the mass-transfer coefficient allows us to standardize nutrient uptake across a range of stream depths and velocities. The second modification was to use a large number of streams in a similar location so that there would not be large differences in geology and/or nutrient regimes that could influence differences in nutrient uptake. We performed many more injections than has been previously attempted ($n = 37$), to improve the resolution between uptake and causal factors. High replication allowed us to examine closely relationships between transient storage and nutrient, which in our case was important because there was high variation in this relationship.

Materials and methods

Study area—We used 13 streams within and near the Hubbard Brook Experimental Forest (HBEF) to measure transient storage and nutrient uptake (Fig. 1, Table 1). The HBEF is located in the White Mountains of New Hampshire (43°56′N, 75°45′W) and has a cool, continental climate with about one third of annual precipitation occurring as snow. The basin is forested primarily by American beech (*Fagus grandifolia*), sugar maple (*Acer saccharum*), and yellow birch (*Betula alleghaniensis*). Streams at HBEF are dominated by cobble and boulder substrates and drain steep, for-

Table 1. Summary data from all injection experiments.

Stream	Date	Physical data					Transient storage parameters						Nutrient uptake			
		Q (L s⁻¹)	Q/w (m² min⁻¹)	Velocity (m min⁻¹)	Width (m)	Depth (m)	k_1 (min⁻¹)	k_2 (min⁻¹)	$1/k_1$ (min)	$1/k_2$ (min)	k_1/k_2	DaI	NH_4 uptake length (m)	$NH_4\ V_f$ (mm min⁻¹)	PO_4 uptake length (m)	$PO_4\ V_f$ (mm min⁻¹)
Bear Brook	10 Jul 1998	14.2	0.310	3.80	2.74	0.082	0.013	0.062	74.6	16.1	0.22	1.59	151	2.05	44	7.07
Bear Brook	26 Oct 1998	16.4	0.381	2.90	2.58	0.132	0.012	0.045	80.6	22.5	0.28	1.96	73	5.22	54	7.07
Bear Brook	23 Apr 1999	13.0	0.292	4.73	2.67	0.062	0.016	0.071	62.9	14.0	0.22	1.84	118	2.48	49	5.95
Bear Brook	19 Jun 1999	4.2	0.097	1.68	2.62	0.058	0.009	0.024	116.1	42.0	0.36	1.74	60	1.62	—	—
West Branch, Bowl	23 Oct 1998	77.9	0.862	12.56	5.42	0.069	0.022	0.091	45.7	10.9	0.24	1.80	237	3.64	—	—
Cascade Brook	23 Jun 1999	2.0	0.030	0.54	3.96	0.057	0.009	0.022	117.4	46.3	0.39	2.25	19	1.62	—	—
Cone Pond Outlet	17 Jul 1998	2.3	0.085	1.20	1.62	0.071	0.019	0.043	52.1	23.5	0.45	1.18	64	1.33	8	11.2
Cone Pond Outlet	22 Oct 1998	6.5	0.209	3.10	1.87	0.067	0.031	0.066	31.9	15.2	0.47	1.26	29	7.29	—	—
Hubbard Brook	22 Jul 1998	86.6	0.509	5.75	10.20	0.089	0.029	0.170	34.2	5.9	0.17	6.25	271	1.88	85	5.99
Hubbard Brook	3 Nov 1998	151.0	0.888	7.97	10.20	0.111	0.008	0.033	129.5	29.9	0.23	0.77	—	—	—	—
Hubbard Brook	25 Jun 1999	29.3	0.192	3.12	9.17	0.061	0.015	0.052	65.4	19.1	0.29	2.60	214	0.90	—	—
Paradise Brook	16 Jul 1998	6.7	0.181	1.58	2.22	0.115	0.025	0.058	39.8	17.3	0.43	2.10	104	1.74	29	6.20
Paradise Brook	13 Jun 1999	3.7	0.085	1.10	2.59	0.078	0.013	0.044	77.5	22.7	0.29	3.11	105	0.81	—	—
W1 stream	18 Jun 1999	1.5	0.065	1.11	1.40	0.058	0.017	0.033	59.9	30.8	0.51	1.33	14	4.72	—	—
W2 stream	13 Jul 1998	0.8	0.030	0.96	1.64	0.032	0.017	0.029	58.8	34.5	0.59	1.87	9	3.53	5	6.47
W2 stream	21 Oct 1998	0.5	0.023	0.81	1.25	0.028	0.015	0.027	68.0	37.0	0.54	2.05	9	2.54	5	4.59
W2 stream	21 Apr 1999	2.1	0.078	1.12	1.65	0.069	0.016	0.024	62.9	42.2	0.67	1.06	—	—	8	9.67
W3 stream	29 Jul 1998	1.1	0.034	1.14	2.00	0.029	0.022	0.062	45.5	16.1	0.35	1.70	24	1.42	4	8.68
W3 stream	20 Oct 1998	3.6	0.104	3.41	2.05	0.030	0.084	0.196	11.9	5.1	0.43	4.19	33	3.16	18	5.87
W3 stream	20 Apr 1999	7.7	0.211	3.91	2.19	0.054	0.064	0.208	15.6	4.8	0.31	3.48	—	—	32	6.53
W3 stream	17 Jun 1999	5.6	0.184	1.58	1.84	0.116	0.015	0.063	67.6	15.9	0.24	2.42	61	3.01	—	—
W3 stream	6 Jul 1999	1.7	0.052	1.37	1.91	0.038	0.012	0.037	82.0	26.8	0.33	1.78	61	0.85	—	—
W3 stream	30 Jun 1998	15.2	0.382	5.40	2.39	0.071	0.025	0.157	40.0	6.4	0.16	1.75	270	1.41	33	11.5
W4 stream	28 Jul 1998	0.6	0.026	0.78	1.47	0.033	0.016	0.024	62.5	41.0	0.66	1.55	11	2.44	5	5.73
W4 stream	1 Oct 1998	5.5	0.134	0.96	2.46	0.139	0.008	0.038	125.5	26.2	0.21	1.62	26	5.12	12	11.6
W4 stream	21 Apr 1999	6.6	0.182	2.77	2.17	0.066	0.021	0.063	46.7	15.8	0.34	1.53	—	—	25	7.38
W5 stream	21 Jul 1998	1.6	0.061	1.67	1.57	0.036	0.030	0.063	33.3	15.9	0.48	1.67	19	3.18	12	5.19
W5 stream	21 Oct 1998	0.8	0.022	0.96	2.16	0.023	0.014	0.044	73.0	22.7	0.31	1.54	8	2.72	11	1.93
W5 stream	28 Apr 1999	2.4	0.080	1.55	1.82	0.052	0.015	0.036	66.7	27.7	0.42	1.57	10	8.29	35	2.30
W6 stream	30 Jul 1998	0.4	0.018	0.81	1.39	0.022	0.054	0.087	18.6	11.5	0.62	3.48	6	2.76	2	8.85
W6 stream	19 Oct 1998	2.0	0.046	1.66	2.55	0.028	0.013	0.039	79.4	25.6	0.32	1.55	18	2.55	24	1.96
W6 stream	22 Apr 1999	2.7	0.089	1.58	1.82	0.048	0.015	0.032	67.6	31.0	0.46	1.34	36	2.44	18	4.93
W6 stream	29 Jun 1998	5.7	0.188	3.90	1.82	0.048	0.031	0.091	31.9	11.0	0.34	1.32	48	3.91	18	10.6
W6 stream	21 Jun 1999	1.4	0.061	1.09	1.40	0.056	0.017	0.026	60.2	39.2	0.65	1.51	18	3.33	—	—
West Inlet to Mirror Lake	14 Oct 1998	0.8	0.048	0.96	1.03	0.050	0.024	0.045	41.0	22.1	0.54	1.82	—	—	—	—
West Inlet to Mirror Lake	19 Apr 1999	1.0	0.045	1.60	1.34	0.028	0.029	0.043	35.0	23.1	0.66	0.81	—	—	12	3.70
West Inlet to Mirror Lake	16 Jun 1999	1.1	0.054	0.84	1.22	0.064	0.022	0.031	45.9	32.5	0.71	1.28	5	10.81	—	—

ested watersheds. The only exception is Hubbard Brook, which is low gradient and has an open canopy. These streams have a shallow hyporheic zone because of bedrock close to the surface. First-order streams have many debris dams, whereas larger streams such as Bear Brook, Paradise Brook, and Main Hubbard Brook have few to no debris dams (Bilby and Likens 1980).

Specific streams studied were those draining the gauged south-facing watersheds (W1–W6). Study reaches on W2, W4, and W5 were upstream of the gauging weirs, and those on W1, W3, and W6 reaches were downstream of the weirs. We also used reaches of Paradise Brook and Bear Brook downstream of the Forest Service road; Hubbard Brook, just upstream of the Forest Service road; Cascade Brook, 100 m upstream of where it enters Hubbard Brook; and the west inlet of Mirror Lake 50 m downslope of the meteorological station (Fig. 1). Two streams outside of HBEF were also studied: the outlet of Cone Pond (Bailey et al. 1995) and the stream (West Branch Whiteface River) draining the Bowl Research Natural Area, an old-growth forest in North Sandwich, New Hampshire (Martin and Bailey 1999). We used the same reaches on each stream for multiple measurements.

Injection procedures—We performed 37 short-term nutrient addition experiments using chloride as a conservative tracer to measure nutrient uptake and transient storage in 13 streams from summer 1998–summer 1999 (Stream Solute Workshop 1990; Webster and Ehrman 1996). During summer, we performed these injections early in the morning, to avoid daily discharge decreases from evapotranspiration; in small watersheds where this effect was most pronounced, we measured stream flow at gauges or culverts before and after each injection. At each stream, we chose a reach long enough to give ~0.5–1 h of transport time. Reach length ranged from 20 m on West Inlet to 300 m at the Bowl Natural Area. Nutrients and sodium chloride were dissolved in stream water and injected simultaneously by use of a Watson-Marlow peristaltic pump. Target concentrations in the streams were 15–20 μg NH_4-N L^{-1}, 15 μg PO_4-P L^{-1}, and 5–7 mg Cl L^{-1}. Background concentrations were 1–4 μg NH_4-N L^{-1} and 1–2 μg PO_4-P L^{-1}. Using an electrical conductivity meter, we measured change in relative salt concentration with time during the injections at a spot near the thalweg of each stream. During summer 1999, we measured conductivity in one well and several side pools for some of the additions using recording Hydrolab multiprobes. We sampled nutrient and Cl concentrations at eight locations along the study reach after the salt concentration reached a plateau (1–3 h for small streams). During summer 1998, NH_4 and PO_4 were added at the same time. During winter and spring seasons, PO_4 was added immediately after the NH_4 injection, with use of Br as a conservative tracer. No PO_4 was added during summer 1999.

During summer additions, unfiltered ammonium concentrations were measured the same day by use of the phenol-hypochlorite method (Solorzano 1969) and a 10-cm cell in the spectrophotometer to maximize sensitivity. For autumn and spring additions, ammonium samples were filtered immediately (Gelman A/E glass-fiber filter) and preserved with 1% chloroform and analyzed colorimetrically by use of an
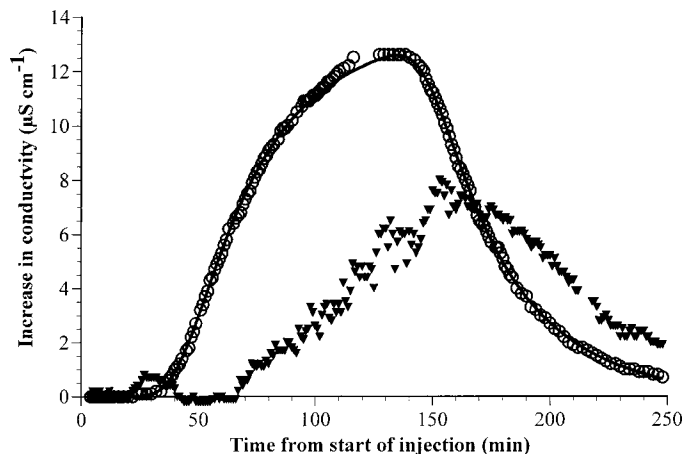

Fig. 2. Solute concentration curve for the 13 June 1999 injection into Paradise Brook. Hollow circles are stream channel conductivity measurements and the line is the one-dimensional, advection, dispersion, and transient storage model fitted to the conductivity data (see text for details). Triangles are conductivity data from a side pool near the stream channel measurement site.

Alpkem autoanalyzer. Because we were interested in the relative decrease of NH_4 with distance downstream (see below), using a variety of methods should not affect our measurements of uptake length, because it is estimated by the relative decline in NH_4 concentrations. The detection limit was 2–4 μg N L^{-1}.

Phosphate, NO_3, Cl, and Br were measured by use of ion chromatography on a Dionex DX 500 with an AS4A column. Detection limits for NO_3, and those for PO_4 were 2 μg N or P L^{-1}. All samples were filtered with Gelman A/E filters in the field and preserved with chloroform. For PO_4 we used a 500-μl injection loop; for all other anions we used a 100-μl loop.

Data analysis—Transient storage was estimated for each injection by estimating parameters of a one-dimensional advection, dispersion, transient storage model (Bencala and Walters 1983). This model is commonly used in other studies that have examined transient storage in streams (D'Angelo et al. 1993; Valett et al. 1996; Hart et al. 1999). The parameters estimated were velocity (v m min^{-1}), exchange rate of water between the main channel and transient storage (k_1 min^{-1}), and exchange rate of water between transient storage and the main channel (k_2 min^{-1}). Relative transient storage zone size (referred herein as transient storage) is k_1/k_2 and is equivalent to A_s/A, used in other studies (Bencala and Walters 1983; D'Angelo et al. 1993; Morrice et al. 1997), and can be conceptually considered as the residence time of water in transient storage relative to the residence time of water in the channel. We estimated these parameters by solving the model iteratively and finding a least-squares best fit of the model output relative to the solute concentration curve measured as conductivity in the field (Hart 1995) (Fig. 2). To estimate whether the reach length chosen was appropriate for measuring transient storage parameters, we calculated the Damkohler number (*DaI*) for each reach, following the method of Wagner and Harvey (1997) by

$$DaI = [(k_1 + k_2) \times L]/v \qquad (1)$$

where L is the length (m) of stream reach. If the reach length is too short ($DaI < 0.5$), then not enough water has entered transient storage to properly estimate parameters. If the reach length is too long ($DaI > 5$), then dispersion dominates the shape of the solute concentration curve (Wagner and Harvey 1997).

In order to understand the role of side pools and hyporheic zones as transient storage areas, we compared solute loss from these areas with that of the main channel after the injection. Stream margins at HBEF contain water that is clearly moving more slowly that that of the main channel. Some of these areas are diffusely distributed along stream margins, whereas others are discrete pools that lie within the wetted channels and have a small area of surface water exchange with the main channel. We used these discrete pools for the present study. After the main pulse of conservative solute in the main channel passes, the rate of decline of solute in the water column is roughly equivalent to k_2 (Hart 1995); therefore, we estimated turnover time of solute in side pools after the main pulse of solute passed by in the channel. We defined the time that the main pulse passed in the channel by when modeled concentration of solute in the channel was 10% of its highest value. By taking the slope of ln concentration of solute in the side pool versus time, we calculated turnover time in that side pool.

Discharge (Q m³ min⁻¹) was estimated on the basis of mass balance of chloride in stream water. Because depth is difficult to measure in these uneven-substratum (boulder, cobble, and organic debris) streams, we calculated an effective depth using discharge, velocity, and width (w m) measured at 10 spots along each reach.

We calculated uptake length of nutrients for each injection using a linear form of an exponential uptake model:

$$\ln C_x = \ln C_0 - ax \qquad (2)$$

where C_x and C_0 are nutrient concentrations at x m downstream from the addition site (0 m) and a is the per meter uptake rate (Newbold et al. 1981). Uptake length S (m) is equal to a^{-1} and is the average distance traveled by a nutrient ion before uptake. We corrected nutrient concentrations for dilution from groundwater inputs using concentrations from the conservative tracer additions (Webster and Ehrman 1996) and used linear regression to estimate parameters for Eq. 2 from field data. Because the primary determinant of uptake length in these streams was depth × velocity (equivalent to Q/w, which is specific discharge), we calculated a mass-transfer coefficient:

$$V_f \text{ (m min}^{-1}) = Q/(wS) \qquad (3)$$

The mass-transfer coefficient (Stream Solute Workshop 1990) corrects for the effects of depth and velocity on nutrient uptake. It can be considered as the velocity at which a nutrient moves through the water column toward the sediments and thus represents demand for nutrients relative to concentration in the water column. Because ambient NH_4 and PO_4 concentrations were at or below detection limits, we were unable to calculate area-specific uptake rates for those ions. For six injection experiments, we could not calculate uptake length because NH_4 concentrations did not significantly decline with distance downstream.

We used simple and multiple linear regression to estimate relationships between predictor variables (k_1/k_2 and NO_3 concentration) and nutrient V_f. The relationship between Q/w and transient storage was highly curvilinear; because we did not have an a priori nonlinear model that relates Q/w with transient storage values, we used a Spearman rank correlation to test whether a relationship exists between the two variables. To compare transient storage among streams, we corrected for the effect of specific discharge by calculating residuals from the regression of ln Q/w versus k_1/k_2. We then performed an ANOVA on these residuals using different streams as treatments. For that test, we only used the subset of streams for which we had three or more measurements of transient storage ($n = 30$). We used t tests to compare seasonal differences in nutrient mass-transfer coefficients, and we used paired t tests to compare NH_4 and PO_4 V_f. We considered spring and fall as one season because the forest canopy was not leafed out at this time and background concentrations of NO_3 were higher. Nitrate concentrations were ln-transformed because they were strongly skewed to high values. All statistics were performed with JMP, version 2.0.5.

Results

Variation in transient storage—Transient storage (k_1/k_2) in HBEF streams ranged from 0.16 to 0.71 (Table 1). The median Damkohler number was 1.7 (Table 1), and these values were not related in any way to Q/w, which suggests that parameter estimation was not affected by change in Q. Combining transient storage measurements from all injections and all seasons gave a curvilinear negative relationship between specific discharge (Q/w) and relative transient storage zone size (Fig. 3); these variables were significantly related to each other (Spearman rank correlation $r_s = -0.69$, $P < 0.005$). There was no relationship with specific discharge and residence time of water in the channel (k_1^{-1}) ($r_s = -0.05$, $P > 0.1$); however, the residence time of water in storage (k_2^{-1}) tended to be lower at higher specific discharges (Fig. 3) ($r_s = -0.52$, $P < 0.005$). Using the subset of streams for which we had three or more measurements, there was a significant linear relationship between k_1/k_2 and ln Q/w ($k_1/k_2 = -0.117$ (ln Q/w) $- 0.129$, $r^2 = 0.51$ and $P < 0.0001$), however, ANOVA of the residuals from this regression was not significant ($P = 0.12$), which shows that most of the among-stream differences were explained by variation in specific discharge.

Water in side pools turned over more slowly than the estimated time that water spends in transient storage in nearly all cases (Table 2, Fig. 2). In W3, conductivity in the side pool was actually increasing after conductivity in the main channel was decreasing, which suggests that this side pool was acting as a storage zone. We have only one measurement from a hyporheic well, and it also turned over more slowly than modeled transient storage (Table 2).

Nutrient uptake—Uptake length of NH_4 ranged from 5 to 277 m (Table 1), on the basis of 31 injections. Phosphorus uptake lengths were shorter, ranging from 2 to 54 m, al-
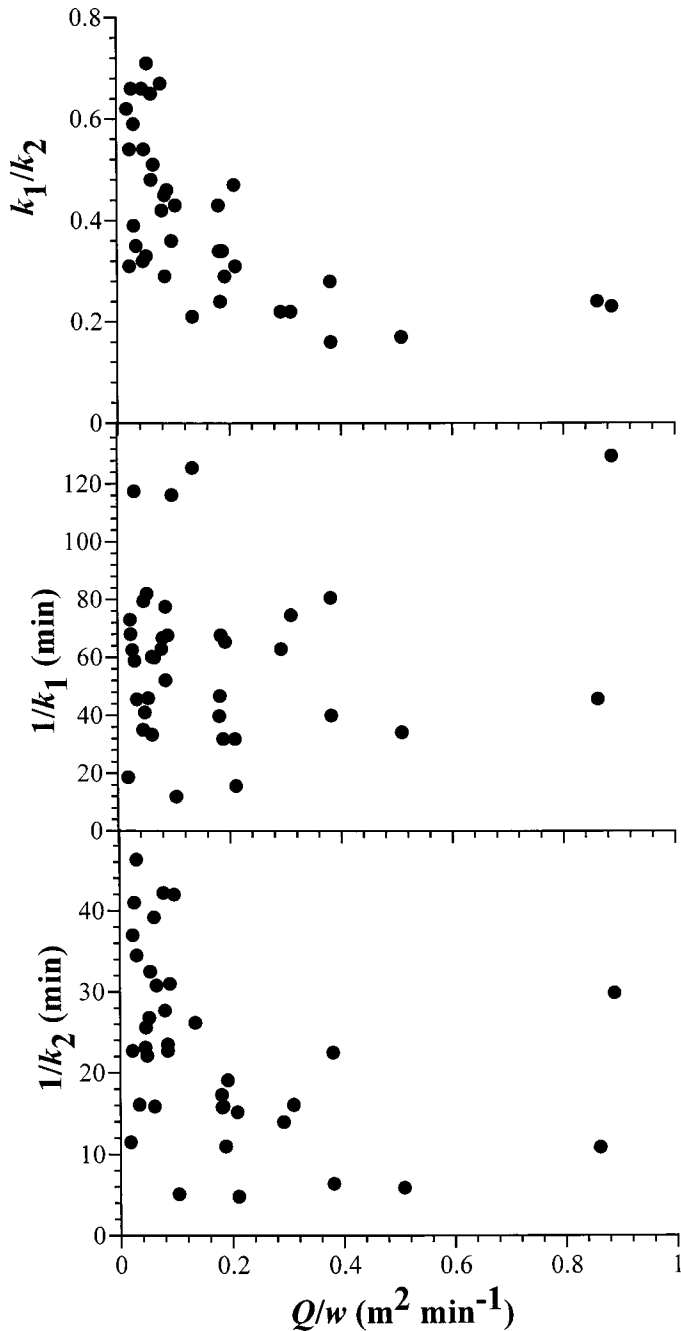
Fig. 3. Relationships between specific discharge (Q/w m² min⁻¹) and transient storage parameters for all of the injection experiments.

Fig. 4. Relationships between specific discharge (Q/w m² min⁻¹) and uptake lengths (S). Equations for the linear regressions are (NH₄) $S = 353(Q/w) + 13.0$, $r^2 = 0.64$, $P < 0.0001$; and (PO₄)$S = 131(Q/w) + 3.5$, $r^2 = 0.81$ and $P < 0.0001$.

though we have P uptake data from fewer streams (Table 1). Residence times of these nutrients in the water column were short, ranging from 4.5 to 70 min for NH_4 and 2.5 to 23 min for PO_4. Uptake lengths for both nutrients were strongly related to specific discharge, with 64% of variation in NH_4 uptake explained by Q/w and 81% of PO_4 uptake (Fig. 4). Because most variation in uptake length was a function of specific discharge, we converted to V_f. Mass-transfer coefficients for PO_4 were, on average, threefold higher than those for NH_4 (paired t test $P < 0.0001$) (Table 3); however, V_f
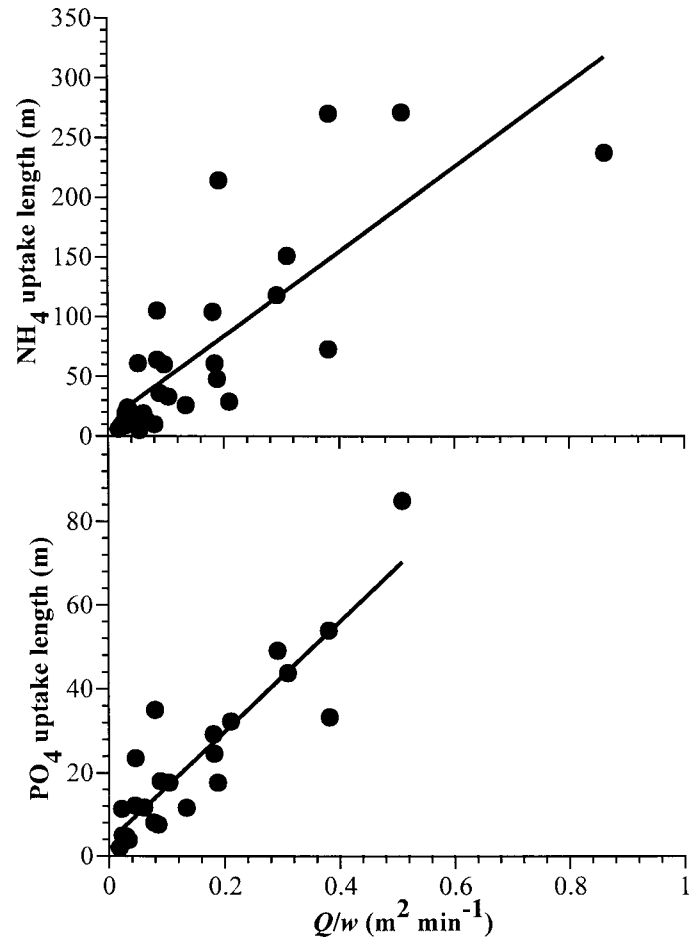
for NH_4 was more variable (coefficient of variation [CV] = 70%) than V_f for PO_4 (CV = 43%). Ammonium V_f was 1.6 times higher during autumn and spring injections (4.13 mm min⁻¹) than summer (2.66 mm min⁻¹), although this difference is not strongly significant (t test $P = 0.082$). For PO_4, this effect was more pronounced; autumn and spring V_f (7.65 mm min⁻¹) was higher than summer (5.95 mm min⁻¹) (t test, $P = 0.046$). Although there was high variation in NH_4 V_f, there were no statistically significant differences among streams when those streams that had three or more experiments (ANOVA $P = 0.33$) were used; thus, variation among streams was not higher than variation within streams.

Ammonium V_f was weakly but significantly positively related to transient storage ($r^2 = 0.14$, $P = 0.04$) (Fig. 5) when all seasons were considered. However, when only data from summer experiments were used, this relationship improved ($r^2 = 0.35$, $P = 0.008$). When only spring and autumn experiments were used, there was no relation between transient storage and NH_4 V_f. During summer experiments, NH_4 V_f was negatively related to NO_3 concentration (Fig 5); this pattern was not observed when data from all seasons or autumn and spring alone were used. Nitrate concentrations

Table 2. Comparison of turnover time of water in side pool with whole-stream transient storage turnover time (k_2^{-1}).

Stream	Date	Location	Pool/hyporheic turnover time (min)	Whole-stream transient storage turnover time (k_2^{-1}) (min)
W3	17 Jun 1999	Side pool	NA*	15.9
W3	17 Jun 1999	Hyporheic	32.6	15.9
W6	21 Jun 1999	Side pool	40.3	39.2
W6	21 Jun 1999	Side pool	48.1	39.2
West Inlet	16 Jun 1999	Side pool	54.0	32.5
West Inlet	16 Jun 1999	Side pool	45.6	32.5
Paradise Brook	13 Jun 1999	Side pool	58.1	22.7
Cascade Brook	23 Jun 1999	Side pool	84.0	46.3
Cascade Brook	23 Jun 1999	Side pool	82.6	46.3
Bear Brook	19 Jun 1999	Side pool	69.9	42.0
Bear Brook	19 Jun 1999	Side pool	59.8	42.0
Hubbard Brook	25 Jun 1999	Side pool	43.6	19.1

* Turnover time was not calculated because the solute concentration inside pool was still increasing during solute decline in the channel.

were higher in autumn and spring (geometric mean 136 μg NO$_3$-N L^{-1}) than in summer (geometric mean 28 μg NO$_3$-N L^{-1}) (*t* test $P = 0.001$). Together, NO$_3$ concentration and transient storage explained 46% of variation in ammonium V_f from summer experiments (Table 4). Phosphate V_f was unrelated to transient storage, NO$_3$ concentration, or NH$_4$ V_f (Fig. 6).

Discussion

Transient storage—Transient storage varied from 0.16 to 0.71 and was within the range of many other observations, although not as high as some small streams, where transient storage is >1 (e.g., Valett et al. 1996; Martí et al. 1997;

Butterini and Sabater 1999). Relative to other forest streams in the eastern United States, transient storage values at HBEF were generally lower that those for Coweeta Hydrologic Laboratory, North Carolina, but higher than Walker Branch, Tennessee (D'Angelo et al. 1993; Hart et al. 1999). Variation in transient storage at HBEF was largely a function of velocity and depth of the streams, which has been observed at HBEF (Paul and Hall unpubl. data); Coweeta, North Carolina (D'Angelo et al. 1993); Sycamore Creek, Arizona (Martí et al. 1997); and Gallina Creek, New Mexico (Valett et al. 1996). Specific discharge did not relate to the exchange of water from the channel to transient storage (k_1^{-1}), but rather it related better to the exchange of water from transient storage to the channel (k_2^{-1}). This finding is

Table 3. Summarized mass-transfer coefficient (V_f) for NH$_4$ and PO$_4$ uptake experiments. Only data from nonisotopic measurements are shown.

Nutrient, location	Reference	Mean V_f (mm min^{-1})	SD	Range (mm min^{-1})
NH$_4$				
HBEF	This study	3.2	2.2	0.18–10.8
Walker Branch, TN	Mulholland et al. (2001)	4.1		4.1
La Solana, Spain Riera Major, Spain	Martí and Sabater (1996)	6.9*		
Riera Major, Spain	Martí and Sabater (1996)	13.0*		
Hugh White Creek,	Sabater et al. (2000)			1.6–7.6
NC	Hall et al. (1998)	3.6		3.6
PO$_4$				
HBEF	This study	67	2.9	1.9–11.5
Idaho	Davis and Minshall (1999)	7.0		6.8–7.3
La Solana, Spain Riera Major, Spain	Martí and Sabater (1996)	5.7*		
Riera Major, Spain	Martí and Sabater (1996)	9.9*		
Hugh White Creek,	Sabater et al. (2000)			0.7–4.7
NC	Munn and Meyer (1990)	3.2	1.6	1.17–4.5
WS2, OR	Munn and Meyer (1990)	0.11	0.07	0.04–0.17

* Mean of bedrock and sand-cobble reaches. Calculated as grand mean of uptake (μg m^{-2} min^{-1}) for all experiments divided by mean concentration.
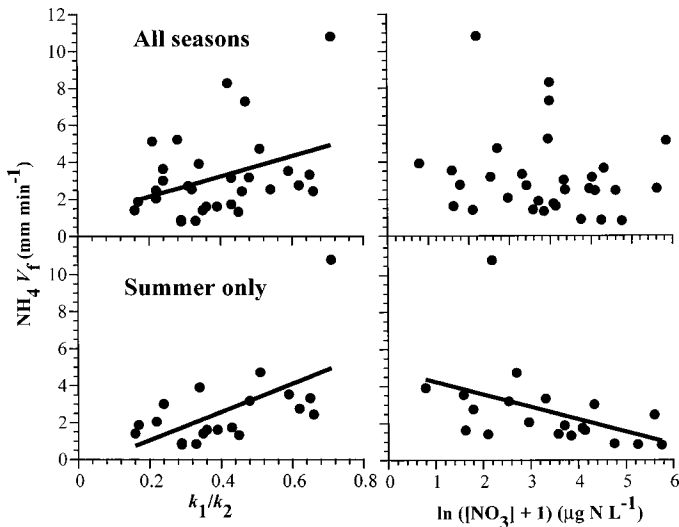
Fig. 5. Relationships between transient storage (k_1/k_2) and nitrate concentration and mass-transfer coefficient (V_f) of NH_4. Top panels are for all seasons, and bottom panels are for only summer injection experiments. Lines are statistically significant linear regressions and are (NH_4 V_f) = 5.46(k_1/k_2) + 1.05, r^2 = 0.14, P = 0.042 (for all injections), (NH_4 V_f) = 7.64(k_1/k_2) − 0.48, r^2 = 0.34, P = 0.0072 (for summer injections), and (NH_4 V_f) = −0.673(ln[NO_3 + 1]) + 4.91, r^2 = 0.19, P = 0.056. The relationship between summer NH_4 V_f and transient storage is still significant (P = 0.035) without the outlying point.

Table 4. Results of multiple regression relating nitrate and transient storage with NH_4 mass-transfer coefficient for summer injection experiments. Nitrate concentrations were transformed to ln (NO_3 + 1). R^2 for this model is 0.46 and n = 20.

Parameter	Value	F ratio	P
k_1/k_2	1.60	7.99	0.012
NO_3 concentration	−0.523	3.36	0.084
Intercept	6.82		

were in one stream (Walker Branch, Tennessee), and variation in transient storage was low, from 0.075 to 0.17, whereas transient storage in HBEF streams varied more than fourfold.

Perhaps because of our choice of streams—those with similar geology, watershed vegetation, and hydrologic regime—among-stream variation was not higher than within-stream variation once corrected for the effect of specific discharge. Of course, variation in stream size (as expressed by discharge) is associated with varying stream geomorphology, so the relationship between Q/w and transient storage may indeed be a function of geomorphic changes along a gradient of stream size. Because of the high within-stream variability in transient storage and low replication within individual streams, we cannot separate variation caused by discharge with geomorphic variation associated with stream size.

An important consideration examining transient storage is to understand where it is found in a stream, because we might expect that its relationship with nutrient uptake will differ if, e.g., transient storage lies in surface pools or if it is truly hyporheic (see below) (Harvey et al. 1996). The observed decline in k_2 with Q/w may be a function of surface transient storage; water may flow faster through surface transient storage zones in high-discharge streams. We suggest that side pools do indeed retain solute (and thus water) but that they turn over more slowly than what we are modeling as transient storage. Harvey et al. (1996) showed that deep alluvium had much slower turnover than modeled storage; thus, the transient storage model did not correctly account for storage provided by this deep alluvium. Side pools at HBEF streams may represent a important storage area analogous to this deep alluvium; they store water but are not properly considered when the transient storage model is used.
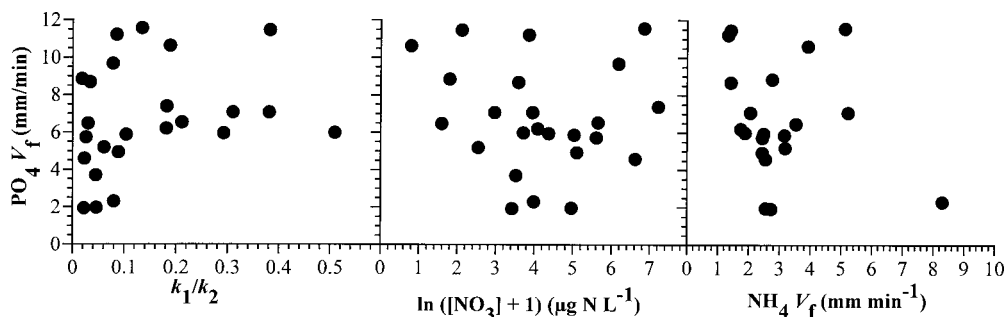
opposite of that found by Hart et al. (1999), who found that the rate of water exchange from the channel to transient storage increased with increasing discharge. Our finding that residence time of water in transient storage decreases with increasing specific discharge suggests that, at higher discharge, water moves through transient storage more quickly than at low discharges. However, we must interpret this result with caution, because at higher flows this tracer method may be most sensitive to fast-turnover transient storage in the reach (Wagner and Harvey 2000). Although our estimation of transient storage parameters was robust at high discharges (because *DaI* was not related to Q/w), this tracer method may be measuring different components of surface transient storage at high Q, whereas actual hyporheic flow paths remain the same. Hart et al. (1999) found no relationship between Q and k_1/k_2; however, all of their injections

Fig. 6. Relationships between transient storage (k_1/k_2), NO_3 concentration, and NH_4 mass-transfer coefficient with mass-transfer coefficient (V_f) of PO_4. None was significantly significant.

Nutrient uptake—Our estimates of nutrient mass-transfer coefficients are within the range of others (Table 3). Interestingly, there is low variation in V_f for NH_4 and PO_4 among regions, and the range at HBEF nearly encompasses mean values or single measurements from other streams. An exception is WS 2 at H. J. Andrews Experimental Forest in Oregon, which has low PO_4 V_f, possibly because of higher soluble reactive phosphorus there (5 μg L^{-1}), relative to Hugh White Creek, North Carolina (1 μg L^{-1}) (Munn and Meyer 1990) or HBEF (<2 μg L^{-1}). We recognize that we probably have underestimated true mass-transfer coefficients, because the method of adding nutrients to streams can saturate microbial uptake mechanisms (Mulholland et al. 1990, 2001), so we compared our data only with those studies that used the same method.

Mass-transfer coefficients of NH_4 were weakly related with transient storage, which explains 14% of variation throughout the year and 35% of the variation during summer. The decrease in r^2 when all the data were included was the result of including autumn and spring experiments, in which we saw no relationship between NH_4 uptake and transient storage. There are several explanations why transient storage only weakly explains variation in nutrient V_f. Measurement error of uptake or transient storage certainly explains some of this variation; however, parameter estimates for transient storage should be robust because we chose appropriate reach lengths for all but one injection (i.e., Damkohler numbers between 0.5 and 5; Harvey and Wagner 2000).

Another possibility is the *nature* of transient storage at HBEF: what exactly are we measuring when we calculate transient storage, and how does the type of transient storage influence nutrient uptake? If transient storage were predominantly hyporheic, then the mechanism would be fairly clear; water flowing into sediments would more likely encounter nutrient-hungry biofilms (Mulholland et al. 1997; Harvey and Wagner 2000). In this situation, high transient storage values will indicate longer hydraulic residence times in the hyporheic zone and should be associated with higher rates of biogeochemical processes (Findlay 1995). Model simulations predict that increasing the surface water exchange rate (k_1) decreases nutrient uptake length and that increasing k_1/k_2 will also decrease nutrient uptake length (Mulholland and DeAngelis 2000). These predictions are only partially supported by our data. There was no relationship between k_1 and either V_f or uptake length of NH_4. However, ammonium V_f did positively relate with k_1/k_2. If transient storage were primarily in surface pools, then we would expect to see a much weaker relationship between transient storage and V_f. Water held up in side pools would not be exposed to biofilms to the degree that water flowing through sediments would encounter biofilms; hence, we would expect lower nutrient uptake rate. In a side pool, microbially colonized surface area relative to the volume of water above it would be lower than that for water flowing through the hyporheic zone. On the other hand, pools are often depositional areas that have high stocks of organic matter that may have high nutrient demand relative to riffle areas.

One aspect of ammonium uptake that we did not test was differences in biological demand among streams at HBEF. Streams with high organic matter and thus high microbial
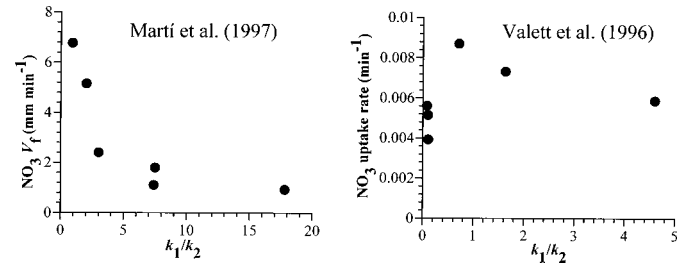

Fig. 7. Relationships between transient storage (k_1/k_2) and NO_3 uptake for two published studies, Martí et al. (1997) and Valett et al. (1996).

secondary production should have higher demand for NH_4 (but not PO_4, see below). An experimental leaf-litter exclusion in a Coweeta stream decreased nutrient uptake, showing the importance of biological processes (Webster et al. 2001). We have indirect evidence, by the negative relationship between NH_4 V_f and NO_3 concentration found during summer, that biological demand may be important. We suggest that high NO_3 concentration alleviates microbial demand for NH_4, thereby lowering NH_4 V_f (Bernhardt et al. unpubl. data). Additional evidence for the role of biological demand is from an experimental acetate addition on W6 stream that stimulated bacterial productivity and increased NH_4 V_f (Bernhardt and Likens in press).

Phosphate uptake did not significantly relate to any of our measured physical variables nor to NH_4 V_f (Fig. 6). We might a priori expect that PO_4 V_f would relate to transient storage if NH_4 relates to transient storage; however, we did not see this relation. Phosphate uptake at HBEF is controlled by chemical sorption to sediments; biological uptake is relatively unimportant (Meyer 1979). Variation in PO_4 uptake is driven by particle size of the sediments, with smaller sediment particles having higher PO_4 demand and lower equilibrium PO_4 concentrations (Meyer 1979). It is possible that all the variation in PO_4 uptake that we observe is solely caused by differences in sediment properties at each site. In addition, hyporheic transient storage may be expected to relate inversely with PO_4 uptake, because fine sediment particles will have low hydraulic conductivity and thus low transient storage (Morrice et al. 1997) but will have high P demand (Meyer 1979). If PO_4 uptake were biotic, we might have expected it to have varied inversely with NH_4 V_f or NO_3 concentration, depending in the N/P limitation status of each stream, but this is not the case. Although we know that PO_4 uptake is controlled by sediments at HBEF, we were not able to provide any further insight as to what controls P uptake (beyond the effect of velocity and depth of water) at the reach scale at HBEF.

Other studies have suggested that increasing the size of transient storage zones promotes nutrient uptake in streams (Valett et al. 1996; Mulholland et al. 1997); however, none has converted uptake length to a mass-transfer coefficient. Although we found a significant positive relationship between k_1/k_2 and NH_4 V_f, reanalysis of data from two other studies does not support this relationship. We reanalyzed data from Valett et al. (1996) and Martí et al. (1997), who reported multiple nutrient uptake measurements coupled

with measurements of the size of the transient storage zone. Using data from Martí et al. (1997), we converted nutrient uptake length to a mass-transfer coefficient with Eq. (3), which showed a negative relationship between transient storage and nutrient V_f (Fig. 7). This result is perplexing because Martí et al. (1997) hypothesized that high transient storage in Sycamore Creek may be caused by thick algal mats, which should have high nutrient uptake. Valett et al. (1996) did not report width or depth; therefore, we calculated a per unit time uptake rate by dividing water velocity by nutrient uptake length. If depth data were available, then V_f would be calculated as depth (mm) $\times$ uptake rate (min^{-1}). There was no relationship between NO_3 uptake rate and k_1/k_2 (Fig. 7) (Valett et al. 1996). Valett et al. (1996) did report a positive relationship between uptake length and k_1/k_2, but because both of these parameters covaried with discharge, it is difficult to interpret whether differences in either discharge or transient storage drove variation in nutrient uptake length in their streams. On the basis of this reanalysis, we suggest that in their streams, variation in nutrient uptake rate is caused by something other than transient storage. Mulholland et al. (1997) showed higher PO_4 V_f for a stream with higher k_1/k_2, but with only two streams in their study, this relationship cannot be tested statistically. These three studies taken together do not show a strong role for transient storage in controlling variation in nutrient uptake among streams.

Our study showed that streams can remove inorganic nutrients relatively rapidly from the water column and that transient storage can explain a small fraction of this variance. However, we hesitate to suggest that transient storage, as measured and modeled in our study, will generally be a strong predictor of nutrient uptake. It is possible that transient storage may control nutrient uptake at other sites, although reanalysis of two of such sites did not show a positive effect of transient storage on nutrient uptake; indeed, at Sycamore Creek, transient storage was negatively correlated with nutrient uptake. To improve understanding of how transient storage might affect nutrient uptake, we suggest further investigation into the nature and location of transient storage and the degree to which it indicates hyporheic or surface storage, as indicated by Harvey et al. (1996). Examining the nature of transient storage (e.g., its location) and the hydrologic basis of hyporheic flow (Bencala 2000) will improve our mechanistic understanding of how transient storage does or does not enhance nutrient uptake in streams; this approach may require empirical investigations into the location of transient storage and modifications of models to consider surface versus subsurface transient storage. Given that streams can rapidly transform and remove nitrogen (Alexander et al. 2000; Peterson et al. 2001), we suggest that better understanding of physical basis of transient storage in streams will improve predictions and mechanistic understanding of this process.

References

ALEXANDER, R. B., R. A. SMITH, AND G. E. SCHWARZ. 2000. Effect of stream channel size on the delivery of nitrogen to the Gulf of Mexico. Nature **403:** 758–761.

BAILEY, S. W., C. T. DRISCOLL, AND J. W. HORNBECK. 1995. Acid-base chemistry and aluminum transport in an acidic watershed and pond in New Hampshire. Biogeochemistry **28:** 69–91.

BENCALA, K. E. 2000. Hyporheic zone hydrological processes. Hydrol. Proc. **14:** 2797–2798.

———, AND R. A. WALTERS. 1983. Simulation of solute transport in a mountain pool-and-riffle stream-a transient storage model. Water Resour. Res. **19:** 718–724.

BERNHARDT, E. S., AND G. E. LIKENS. In press. Effects of DOC enrichment on stream nitrogen dynamics at the Hubbard Brook Experimental Forest. Ecology.

BILBY, R. E., AND G. E. LIKENS. 1980. Importance of organic debris dams in the structure and function of stream ecosystems. Ecology **61:** 1107–1113.

BURNS, D. A. 1998. Retention of NO_3^- in an upland stream environment: A mass balance approach. Biogeochemistry **40:** 73–96.

BUTTERINI, A., AND F. SABATER. 1998. Ammonium and phosphate retention in a Mediterranean stream: Hydrological vs. temperature control. Can. J. Fish. Aquat. Sci. **55:** 1938–1945.

———, AND F. SABATER. 1999. Importance of transient storage for ammonium and phosphate retention in a sandy-bottom Mediterranean stream. Freshw. Biol. **41:** 593–603.

D'ANGELO, D. J., J. R. WEBSTER, S. V. GREGORY, AND J. L. MEYER. 1993. Transient storage in Appalachian and Cascade mountain streams as related to hydraulic characteristics. J. N. Am. Benthol. Soc. **12:** 223–235.

DAVIS, J. C., AND G. W. MINSHALL. 1999. Nitrogen and phosphorus uptake in two Idaho (USA) headwater wilderness streams. Oecologia **19:** 247–255.

DeANGELIS, D. L., M. LOREAU, D. NEERGAARD, P. J. MULHOLLAND, AND E. R. MARZOLF. 1995. Modelling nutrient-periphyton dynamics in streams: The importance of transient storage zones. Ecol. Model. **80:** 149–160.

FINDLAY, S. 1995. Importance of surface-subsurface exchange in stream ecosystems. Limnol. Oceanogr. **40:** 159–164.

FISHER, S. G., N. B. GRIMM, E. MARTÍ, R. M. HOLMES, AND J. J. JONES. 1998. Material spiraling in stream corridors: A telescoping ecosystem model. Ecosystems **1:** 19–34.

HALL, R. O., K. H. MACNEALE, E. S. BERNHARDT, M. FIELD, AND G. E. LIKENS. 2001. Biogeochemical response of two forest streams to a two-month calcium addition. Freshw. Biol. **46:** 291–302.

———, B. J. PETERSON, AND J. L. MEYER. 1998. Testing a nitrogen cycling model of a forest stream by using a nitrogen-15 tracer addition. Ecosystems **1:** 283–298.

HART, D. R. 1995. Parameter estimation and stochastic interpretation of the transient storage model for solute transport in streams. Water Resour. Res. **31:** 323–328.

———, P. J. MULHOLLAND, E. R. MARZOLF, D. L. DeANGELIS, AND S. P. HENDRICKS. 1999. Relationships between hydraulic parameters in a small stream under varying flow and seasonal conditions. Hydrol. Proc. **13:** 1497–1510.

HARVEY, J. W., AND B. J. WAGNER. 2000. Quantifying hydrologic interaction between streams and their subsurface hyporheic zones, p. 3–44. *In* J. B. Jones and P. J. Mulholland [eds.], Streams and ground waters. Academic.

———, ———, AND K. E. BENCALA. 1996. Evaluating the reliability of the stream tracer approach to characterize stream-subsurface water exchange. Water Resour. Res. **32:** 2441–2451.

LIKENS G. E., AND F. H. BORMANN. 1995. Biogeochemistry of a forested ecosystem, 2nd ed. Springer-Verlag.

MARTÍ, E., N. B. GRIMM, AND S. G. FISHER. 1997. Pre- and post-flood retention efficiency of nitrogen in a Sonoran Desert stream. J. N. Am. Benthol. Soc. **16:** 805–819.

———, AND F. SABATER. 1996. High variability in temporal and

spatial nutrient retention in Mediterranean streams. Ecology **77:** 854–869.

MARTIN, C. W., AND A. S. BAILEY. 1999. Twenty years of change in a northern hardwood forest. Forest Ecol. Manage. **123:** 253–260.

MEYER, J. L. 1979. The role of sediments and bryophytes in phosphorus dynamics in a headwater stream ecosystem. Limnol. Oceanogr. **24:** 365–375.

MORRICE, J. A., H. M. VALETT, C. N. DAHM, AND N. E. CAMPANA. 1997. Alluvial characteristics, groundwater-surface water exchange, and hydrologic retention in headwater streams. Hydrol. Proc. **11:** 253–267.

MULHOLLAND, P. J., AND D. L. DEANGELIS. 2000. Surface-subsurface exchange and nutrient spiraling, p. 149–166. *In* J. B. Jones and P. J. Mulholland, [eds.], Streams and ground waters. Academic.

———, E. R. MARZOLF, J. R. WEBSTER, D. R. HART, AND S. P. HENDRICKS. 1997. Evidence that hyporheic zones increase heterotrophic metabolism and phosphorus uptake in forest streams. Limnol. Oceanogr. **42:** 443–451.

———, A. D. STEINMAN, AND J. W. ELWOOD. 1990. Measurements of phosphorus uptake length in streams: Comparison of radiotracer and stable PO_4 releases. Can. J. Fish. Aquat. Sci. **47:** 2351–2357.

———, J. L. TANK, D. M. SANZONE, J. R. WEBSTER, W. WOLLHEIM, B. J. PETERSON, AND J. L. MEYER. 2001. Ammonium and nitrate uptake lengths in a small forested stream determined by ^{15}N tracer and short-term nutrient addition experiments. Int. Verh. Verein. Limnol. **27:** 1320–1325.

MUNN, N. L., AND J. L. MEYER. 1990. Habitat-specific solute retention in two small streams: An intersite comparison. Ecology **71:** 2069–2082.

NEWBOLD, J. D., J. W. ELWOOD, R. V. O'NEILL, AND W. VAN-

WINKLE. 1981. Measuring nutrient spiraling in streams. Can. J. Fish. Aquat. Sci. **38:** 860–863.

———, ———, ———, AND A. L. SHELDON. 1983. Phosphorus dynamics in a woodland stream ecosystem: A study of nutrient spiraling. Ecology **64:** 1249–1265.

PETERSON, B. J., AND OTHERS. 2001. Control of nitrogen export from watersheds by headwater streams. Science **292:** 86–90.

SABATER, F., A. BUTTERINI, E. MARTÍ, I. MUÑOZ, A. ROMANÍ, J. WRAY, AND S. SABATER. 2000. Effects of riparian vegetation removal on nutrient retention in a Mediterranean stream. J. N. Am. Benthol. Soc. **19:** 609–620.

SOLORZANO, L. 1969. Determination of ammonia in natural waters by phenolhypochlorite method. Limnol. Oceanogr. **14:** 799–801.

STREAM SOLUTE WORKSHOP. 1990. Concepts and methods for assessing solute dynamics in stream ecosystems. J. N. Am. Benthol. Soc. **9:** 95–119.

VALETT, H. M., J. A. MORRICE, C. M. DAHM, AND M. E. CAMPANA. 1996. Parent lithology, surface-groundwater exchange, and nitrate retention in headwater streams. Limnol. Oceanogr. **41:** 333–345.

WAGNER, B. J., AND J. W. HARVEY. 1997. Experimental design for estimating parameters of rate-limited mass transfer: Analysis of stream transfer studies. Water Resour. Res. **33:** 1731–1741.

WEBSTER, J. R., AND T. P. EHRMAN. 1996. Solute dynamics. *In* F. R. Hauer and G. A. Lamberti [eds.], Methods in stream ecology. Academic.

———, AND OTHERS. 2001. Effects of litter exclusion and wood removal on phosphorus and nitrogen retention in a forest stream. Int. Verh. Verein. Limnol. **27:** 1337–1340.