

Nitrification in the hyporheic zone of a desert stream ecosystem

JEREMY B. JONES, JR.¹, STUART G. FISHER,
AND NANCY B. GRIMM

Department of Zoology, Arizona State University, Tempe, Arizona 85287 USA

Abstract. Nitrification in the hyporheic zone of Sycamore Creek, a Sonoran Desert stream, was examined, focusing on the association between respiration and nitrate production. Subsurface respiration in Sycamore Creek is highest in regions of hydrologic downwelling where organic matter derived from the stream surface is transported into the hyporheic zone. Similarly, nitrification was closely related to hydrologic exchange between the surface and hyporheic zone. Nitrification in downwelling regions averaged $13.1 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ compared with $1.7 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ in upwelling regions. Hyporheic respiration also varies temporally as a result of flash floods which scour and remove algae from the stream and thus reduce the pool of organic matter to support subsurface metabolism. Nitrification was also significantly affected by flooding; nitrification increased from an average of only $3.0 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ immediately following floods to $38.5 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ late in succession. Nitrification was significantly correlated with hyporheic respiration, supporting the hypothesis that nitrification is fueled by mineralization of organic nitrogen to ammonium. The coupling between subsurface respiration and nitrification is one step in a cyclic interaction between surface and hyporheic zones and serves to transform nitrogen from an organic to inorganic form.

Key words: nutrient dynamics, nitrate, nitrification, desert streams, hyporheic zone, hydrologic exchange.

In nutrient-limited ecosystems, factors governing import and transformation of nutrients can have strong effects on production. Streams are composed of a mosaic of interacting subsystems (e.g., surface-stream, riparian, hyporheic and parafluvial zones) that can function as sources or sinks for nutrients (e.g., Peterjohn and Correll 1984, Ford and Naiman 1989, Triska et al. 1989a, Valett et al. 1994). Nutrient demand in particular subsystems, such as that caused by photoautotrophic production in surface channels (Elwood et al. 1981, Newbold et al. 1983, Triska et al. 1989b), and denitrification in riparian zones (Peterjohn and Correll 1984, Lowrance et al. 1985, Duff and Triska 1990), may reduce nutrient concentration. Nutrient transformation or release from other subsystems may elevate nutrient concentration, for example nitrogen fixation in surface streams (Howarth et al. 1988) and groundwater input (Rutherford and Hynes 1987, Ford and Naiman 1989, Hendricks and White 1991). Elucidation of processes occurring within subsystems, as well as inter-

actions between them, is critical to understanding stream ecosystem functioning.

Primary production in streams of the Sonoran Desert is nitrogen limited (Grimm and Fisher 1986a, 1986b). An important source of nitrogen for photoautotrophic production is hydrologic linkage with the nutrient-rich hyporheic zone (Valett et al. 1994). As water flows from the surface through hyporheic and parafluvial zones, nitrate concentration is elevated through nitrification (Grimm et al. 1991, Holmes et al. 1994). This nitrate-rich subsurface water eventually reenters the surface through hydrologic exchange, and alleviates local nutrient limitation (Valett et al. 1994). In fact, nitrate production in hyporheic sediments may be great enough to account for 20-100% of algal nitrogen demand (Grimm 1992).

The objectives of our study were to determine factors regulating nitrification in hyporheic sediments and the contribution of nitrification as a source of inorganic nitrogen for photoautotrophic production. Grimm (1992) and Holmes et al. (1994) speculated that nitrification was fueled by mineralization of organic nitrogen to ammonium during decomposition. This paper specifically focuses on the association between hyporheic respiration and nitrification. Nitrifi-

¹ Present address: Environmental Sciences Division, Oak Ridge National Laboratory, P.O. Box 2008, Building 1000, Oak Ridge, Tennessee 37831-6335 USA.

fication also is examined in relation to the direction of hydrologic exchange with the surface-stream (i.e., downwelling and upwelling zones) and disturbance by flash floods.

Study Site

Sycamore Creek is an intermittent Sonoran Desert stream 32 km northeast of Phoenix, Arizona. The stream drains a 505-km² mountainous drainage basin varying in elevation from 427 to 2164 m. The catchment is composed of igneous and metamorphic rocks with shallow overlying soils and unconsolidated sediments (Thomsen and Schumann 1968). The predominant vegetation is ponderosa pine and piñon-juniper woodlands at higher elevations and Sonoran desert scrub at lower elevations. Precipitation is bimodal with peaks in winter and summer and annual means of 58.4 and 33.9 cm at higher and lower elevations, respectively (Thomsen and Schumann 1968). Runoff from storms frequently produce flash floods which scour the stream and severely reduce the biota (Fisher et al. 1982).

In mid-elevation reaches (600 to 760 m) of Sycamore Creek, where this study was performed, mean stream depth is 5 cm and average wetted channel width is 5–6 m. The wetted channel is bounded by an active channel of alluvium that is over 20 m wide (Fisher et al. 1982). Stream substrata consist primarily of sand and fine gravel with a mean depth to bedrock of 62 cm (Valett et al. 1990). Interstitial flow rate through hyporheic sediments is rapid, averaging 2 m/h (Valett et al. 1990, Holmes et al. 1994). Riparian cover is sparse; consequently the stream receives full sunlight most of the day and has potential for high algal production. In-stream gross primary production and algal biomass are lowest immediately after floods (2.7 g O₂ · m⁻² · d⁻¹ and 7.1 mg Chl *a*/m², respectively) and increase during succession to as high as 12 g O₂ · m⁻² · d⁻¹ and 350 mg Chl *a*/m², respectively (Grimm 1987).

Three study reaches were established in sandy runs at ~ 650 m elevation. Reaches I, II, and III were 295, 115, and 104 m long, respectively, and were separated by pools and riffles.

Methods

Nitrification was measured in one upwelling and one downwelling zone in each of three

study reaches ($n = 3$ upwelling, $n = 3$ downwelling sites) approximately monthly from April 1992 through July 1993, encompassing four flash floods and three subsequent successional sequences. Upwelling and downwelling zones were at the upstream and downstream ends, respectively, of each of the three study runs (Valett et al. 1994). Vertical hydraulic gradient at each end of each run was measured using mini-piezometers (Lee and Cherry 1978) inserted to a depth of 25 cm (Valett et al. 1994, Jones et al. 1995).

Net nitrification was measured as change in nitrate over time in chambers that contained hyporheic sediments and hyporheic water (Holmes et al. 1994). Chambers were constructed from clear plastic pipe (32 cm long, 4.4 cm inside diameter) that were sealed on both ends with rubber stoppers. Chambers were filled with hyporheic sediments that were collected from a depth of 2–17 cm below the sediment surface. Sediments to fill chambers were collected by scraping away benthic sediments and algae (top 2 cm of sediments) then inserting chambers to a depth of 17 cm and withdrawing the plastic pipe filled with 15 cm of sediments. Chambers were filled with water pumped from the hyporheic zone, gently inverted three times to allow any air within sediments to escape and sealed on the ends with rubber stoppers. Sealed chambers were then buried in hyporheic sediments at a depth of 10 cm and incubated there for 2 to 4 h.

Three metabolism chambers of replicated sediment samples from each upwelling and downwelling zone were incubated, for a total of 18 incubations per sampling date. Replicated incubations within a site were started immediately after filling chambers with sediments and water. Incubations at different sites were started ~ 0.5 h apart (lag between sites due to transit and set-up time) and alternated between downwelling and upwelling zones.

Water to fill chambers was collected from the same piezometer used for vertical hydraulic gradient measurements. Water was drawn with a peristaltic pump and initially pumped into a 4-L polyethylene container, from which the three chambers, three acid-washed polyethylene bottles, and three dissolved oxygen bottles were filled for each sample site ($n = 3$ samples from each upwelling and downwelling site, $n = 18$ subsurface samples total per sampling date). Initial chemical composition of water in the

three chambers for a given site was determined as the average of the three water samples for that site. Final water samples were collected by inverting chambers three times (to mix water in chambers) and filling a polyethylene bottle and dissolved oxygen bottle from a continuous column of water drawn from the chamber. The volume of sediments and water in a chamber, and sediment porosity (water volume per sediment volume) were determined; sediments were saved for later analysis of sediment particles-size composition and particulate organic carbon (POC). Triplicate samples were also collected of surface stream water on each sampling date.

Water chemistry samples were stored at 4°C, filtered upon return to the laboratory (Whatman GF/F glass fiber filters), and analyzed for ammonium nitrogen ($\text{NH}_4\text{-N}$), nitrate nitrogen ($\text{NO}_3\text{-N}$), total dissolved nitrogen (TDN), soluble reactive phosphorus (SRP), and dissolved organic carbon (DOC). Ammonium was measured with the phenolphthalein method (Solorzano 1969). Nitrate was analyzed colorimetrically following reduction to nitrite in cadmium-copper columns (Wood et al. 1967); nitrite nitrogen has never exceeded 1–2 $\mu\text{gNO}_2\text{-N/L}$ in Sycamore Creek waters (unpublished data). Dissolved organic nitrogen (DON) was determined by difference between TDN and dissolved inorganic nitrogen ($\text{NH}_4\text{-N} + \text{NO}_3\text{-N}$); total dissolved nitrogen was analyzed as nitrate following 4 h of ultraviolet oxidation (Manny et al. 1971). Molybdate-antimony analysis was used to measure SRP (Murphy and Riley 1962). Dissolved organic carbon was analyzed using persulfate oxidation on an Oceanography International Model 700 Total Carbon Analyzer (Menzel and Vaccaro 1964). Dissolved oxygen samples were analyzed by the Winkler method.

Sediment samples were stored on ice for transport to the laboratory after which samples were frozen until analyzed. Sediment particle-size class (> 4.75 mm, 2–4 mm, 1–2 mm, < 1 mm) was determined by drying sediments for 48 h (60°C), sieving, and weighing. POC was analyzed by sealing sediments (< 1 mm size class) in glass ampules, digesting with persulfate, and measuring resulting CO_2 on a total organic carbon analyzer (Menzel and Vaccaro 1964).

Annual net nitrification rate was calculated as a weighted mean; nitrification rate on each sampling date was weighted by the time inter-

val between the previous and successive sampling dates. Effects of run location (upstream end versus downstream end) and days-post-flood on water and sediment chemistry, and nitrogen transformations (e.g., net nitrification) were assessed using a repeated measures ANOVA (Wilkinson 1990). The relationships of respiration rate (determined from uptake of dissolved oxygen in chambers; respiration data from Jones et al. 1995), $\text{NH}_4\text{-N}$, DON, dissolved oxygen, temperature, and vertical hydraulic gradient with nitrification rate were analyzed by step-wise multiple linear regression (Wilkinson 1990). Data were log transformed prior to analysis, if necessary to meet assumptions of normality and equal variances.

Results

Hyporheic zone chemistry

Total dissolved nitrogen increased from an average of 229 $\mu\text{gN/L}$ in surface stream water to 323 $\mu\text{gN/L}$ in interstitial water of downwelling zones ($p < 0.001$), but increased little between interstitial waters of downwelling and upwelling zones ($p \geq 0.05$). Much of the increase was in the form of $\text{NO}_3\text{-N}$ (73%), which was almost two-fold greater in hyporheic (170 $\mu\text{gNO}_3\text{-N/L}$) than surface water (99 $\mu\text{gNO}_3\text{-N/L}$; $p < 0.01$; Table 1). The remaining 27% difference in nitrogen between hyporheic and surface water was in the forms of $\text{NH}_4\text{-N}$ (19%) and DON (8%). Neither $\text{NH}_4\text{-N}$ or DON, however, were significantly different between the surface and hyporheic zone ($p \geq 0.05$) averaging 26 $\mu\text{gNH}_4\text{-N/L}$ and 122 $\mu\text{gDON/L}$ across subsystems.

Concentration of $\text{NO}_3\text{-N}$ in stream water significantly decreased over time following flash floods ($p < 0.001$; Fig. 1). Nitrate content of surface water averaged 295 $\mu\text{gNO}_3\text{-N/L}$ immediately after floods and decreased 93% to a mean of only 20 $\mu\text{gNO}_3\text{-N/L}$ late in succession. Similarly, interstitial $\text{NO}_3\text{-N}$ concentration declined on average from 278 $\mu\text{gNO}_3\text{-N/L}$ to 69 $\mu\text{gNO}_3\text{-N/L}$ with time following floods.

Dissolved oxygen was also variable in water of different subsystems. Concentration of dissolved oxygen was nearly equal in surface stream water and interstitial water of downwelling zones ($p \geq 0.05$), but nearly 50% lower in interstitial water of upwelling zones ($p < 0.05$; Table 1) where mean oxygen concentration was

TABLE 1. Mean (± 1 SE) values for chemical and physical characteristics of surface water, hyporheic water in downwelling and upwelling zones, and hyporheic sediments in downwelling and upwelling zones of Sycamore Creek, April 1992 through June 1993 (values are means of three locations sampled). Values within a row having different letter designation have means distinguishable by Tukey's multiple comparison ($\alpha = 0.05$). NA = not applicable.

	Stream location		
	Surface	Downwelling zone	Upwelling zone
NO ₃ ($\mu\text{gN/L}$)*	99 ^a (2)	170 ^b (9)	173 ^b (18)
NH ₄ ($\mu\text{gN/L}$)*	13 (4)	26 (8)	38 (12)
DON ($\mu\text{gN/L}$)*	117 (13)	127 (10)	122 (14)
DOC (mgC/L)*	2.97 (0.13)	3.40 (0.19)	3.15 (0.16)
POC (mgC/L sediment)†	NA	35.7 (3.8)	27.6 (4.0)
SRP ($\mu\text{gP/L}$)*	47 (2)	57 (4)	73 (10)
Dissolved oxygen (mgO ₂ /L)*	7.70 ^a (0.13)	7.62 ^a (0.33)	3.89 ^b (0.38)
Vertical hydraulic gradient (cm/cm)†‡	NA	-0.059 ^a (0.008)	0.013 ^b (0.004)
Porosity (%)	NA	32.4 (1.6)	33.6 (1.5)

* Degrees of freedom for between-subject effects are: location, 2; and error, 4; degrees of freedom for within-subject effects are: time, 11; location \times time, 22; and error, 44.

† Degrees of freedom for between-subject effects are: location, 1; and error, 4; degrees of freedom for within-subject effects are: time, 11; location \times time, 11; and error, 44.

‡ Positive vertical hydraulic gradient indicates hyporheic water upwelling into stream surface; negative vertical hydraulic gradient indicates surface water downwelling into hyporheic zone.

3.89 mg O₂/L. Dissolved oxygen also varied significantly over time ($p < 0.001$); however, the concentration in all subsystems was never lower than 1.30 mg O₂/L and typically greater than 2.00 mg O₂/L.

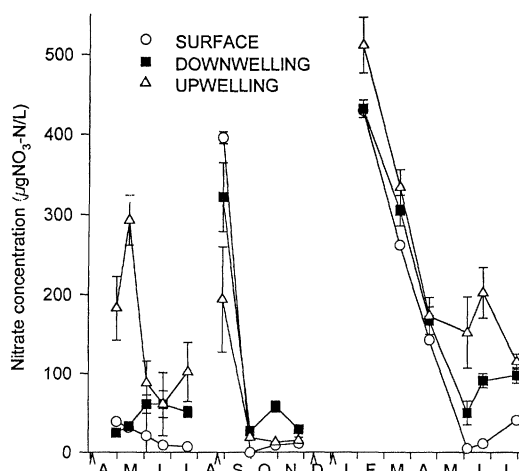


FIG. 1. Mean nitrate concentration (± 1 SE) in surface stream water and interstitial water in downwelling and upwelling zones of Sycamore Creek, Arizona, during three successional sequences following flash floods, April 1992 through July 1993. Arrow heads on abscissa denote time of floods.

In contrast to the spatial variation observed in nitrate and dissolved oxygen, DOC and SRP were not significantly different between surface stream water and interstitial waters of downwelling and upwelling zones ($p \geq 0.05$; Table 1). POC content and porosity of sediments did not vary significantly ($p \geq 0.05$) between sediments in downwelling and upwelling zones with mean values of 31.7 mgC/L sediment and 33%, respectively (Table 1). Sediment in downwelling and upwelling zones were similar, being (by mass) 25% > 4.75 mm, 37% 2-4 mm, 23% 1-2 mm, and 15% < 1 mm, and averaging 1.57 kg sediment/L sediment.

Net nitrification

Nitrification rate was closely related to the direction of hydrologic exchange between the surface and hyporheic zone. In chambers filled with sediments from downwelling zones, nitrification was 13.1 $\mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ compared with only 1.7 $\mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ in chambers from upwelling zones ($p < 0.001$; Table 2). In fact, sediment-associated nitrification in upwelling zones was not significantly different from zero ($p \geq 0.05$), therefore further discussion of nitrification is restricted to downwelling zones.

TABLE 2. Weighted mean (± 1 SE) annual rates of nitrate, ammonium, dissolved organic nitrogen production, and respiration in chambers filled with sediments from downwelling and upwelling zones of Sycamore Creek, April 1992 through June 1993 (values are means of three locations sampled). Values within a row having different letter designation have means distinguishable by Tukey's multiple comparison ($\alpha = 0.05$).

		Hyporheic zone location	
		Downwelling zone	Upwelling zone
Nitrification	($\mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$)	13.1 ^a (4.3)	1.7 ^b (3.0)
Ammonium production	($\mu\text{gNH}_4\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$)	8.7 (4.7)	9.9 (6.9)
DON production	($\mu\text{gDON}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$)	14.8 (8.3)	13.8 (8.9)
Respiration*	($\text{mgO}_2\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$)	1.12 ^a (0.17)	0.46 ^b (0.11)

* From Jones et al. (1995).

Nitrification in downwelling zones was strongly affected by floods, increasing an average of 11-fold over three successional sequences ($p < 0.001$; Fig. 2). Nitrification on average increased from only $3.0 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ immediately following floods to $38.5 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ late in succession. During the two spring-summer successional sequences, when surface and interstitial water temperature ranged from 21 to 25°C, nitrification late in succession was 54.0 (1992) and 45.5 (1993) $\mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$. During the autumn-winter successional sequences, when water temperature ranged from 14 to 23°C, nitrification did not exceed $16.9 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$.

Respiration in the hyporheic zone of Sycamore Creek, like nitrification, was also affected by flooding, increasing from an average of $0.4 \text{ mg O}_2\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ immediately following floods to $1.8 \text{ mg O}_2\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ late in succession (respiration data from Jones et al. 1995) and explaining 76% of the variance in nitrification ($p < 0.001$; Fig. 3). For individual successional sequences, 40 to 96% of the variance was explained by a total of four independent variables (Table 3). During the spring-summer successional sequences, when water temperature was higher and sediments were metabolically active (Jones et al. 1995), nitrification was closely associated with sediment respiration (0.94 and 0.83 partial correlation coefficients for 1992 and 1993, respectively). Furthermore, a total of 96% of the variance in nitrification during the spring-summer successional sequence of 1992 was explained by including interstitial $\text{NH}_4\text{-N}$ concentration; 83% of the 1993 spring-summer nitrification variance was explained with addition of vertical

hydraulic gradient to the model (Table 3). In contrast, during the autumn-winter successional sequence, when hyporheic decomposition rate was reduced (Jones et al. 1995), nitrification was not related to respiration. Only 40% of the variance in nitrification rate was attributable to interstitial water temperature ($p < 0.05$; Table 3).

In addition to nitrate production in sediments, $\text{NH}_4\text{-N}$ and DON were also produced during decomposition in the hyporheic zone (Table 2). Unlike the distinct spatial confinement of nitrification to downwelling zones, $\text{NH}_4\text{-N}$ and DON production were equal between downwelling and upwelling zones ($p \geq$

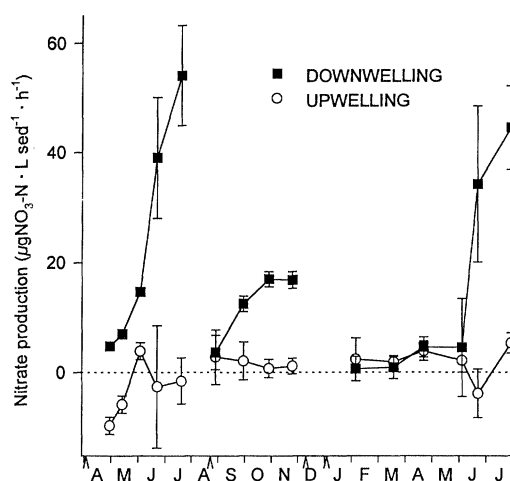


FIG. 2. Hyporheic net nitrification rate in chambers filled with sediments from downwelling and upwelling zones in Sycamore Creek, Arizona, during three successional sequences following flash floods, April 1992 through July 1993. Nitrification values are mean of three sites ± 1 SE. Arrow heads on abscissa denote timing of flash floods.

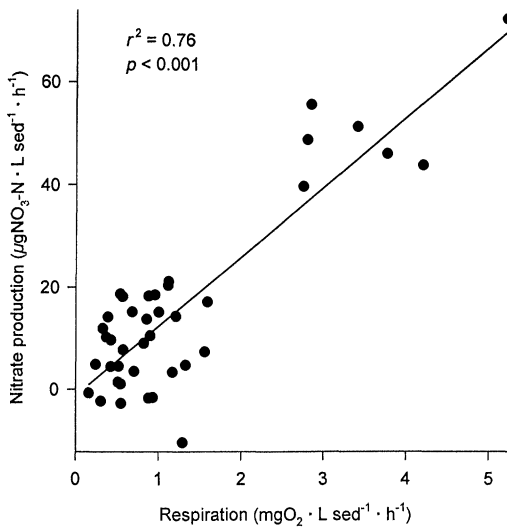


FIG. 3. Hyporheic net nitrification rate versus respiration rate in downwelling zones in Sycamore Creek, Arizona.

0.05), averaging $9.3 \mu\text{gNH}_4\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ and $14.3 \mu\text{gDON}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$.

Discussion

Grimm et al. (1991) hypothesized that elevated hyporheic nitrate concentration is due to either biotic production of nitrate or storage of nitrate-rich floodwater. Although the two hypotheses are not mutually exclusive, the present study supports the nitrification hypothesis.

During respiration, organic nitrogen is mineralized to ammonium, which is then transformed to nitrate. The net nitrification rate of $13.1 \mu\text{gNO}_3\text{-N}\cdot\text{L sediments}^{-1}\cdot\text{h}^{-1}$ would produce the observed increase of nitrate between surface and hyporheic water in only 1.8 h (assuming a sediment porosity of 0.33; Table 1). Similar patterns further supporting the nitrification hypothesis also occur in parafluvial gravel bars of Sycamore Creek. Holmes et al. (1994) reported increased nitrate due to nitrification along parafluvial subsurface flowpaths and pointed out that if nitrate-rich floodwater were the only source of nitrate to parafluvial zones, then nitrate should be constant along subsurface flowpaths.

Storage of nitrate-rich floodwater may also serve as a source of nitrate to the hyporheic zone. Nitrate concentration in floodwater of Sonoran Desert streams is on average six-fold greater than at baseflow (Fisher and Grimm 1985, Grimm and Fisher 1986b), and as Grimm et al. (1991) observed, channel alluvium of Sycamore Creek is expansive and as a result can store a large volume of flood water. Thus, both nitrification and floodwater are probably sources of nitrate to the hyporheic zone.

Model of surface-hyporheic interactions

The dependence of hyporheic nitrification on mineralization of organic nitrogen is one step in a chain of interactions between the surface

TABLE 3. Results of multiple regression analysis (partial correlations) of hyporheic nitrate production rate in chambers versus six independent variables in downwelling zones for three successional sequences. Variables for which partial correlations are not listed were excluded from the model because of low *F*-values.

Independent variables	Successional sequence		
	I ^a	II ^b	IV ^c
Respiration rate	0.94	—	0.83
Ammonium concentration	0.27	—	—
DON concentration	—	—	—
Dissolved oxygen concentration	—	—	—
Temperature	—	-0.67	—
Vertical hydraulic gradient	—	—	-0.29
Multiple <i>r</i> ²	0.96	0.40	0.83
Sample size	10	12	15
<i>F</i> -ratio	106.8	8.3	34.2
<i>p</i> -value	<0.001	<0.05	<0.001

^a April 1992 through July 1992.

^b August 1992 through November 1992.

^c February 1993 through July 1993.

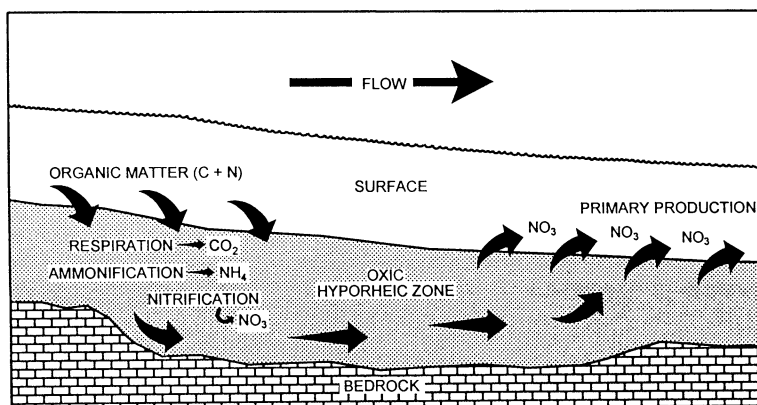


FIG. 4. Conceptual model of surface stream and hyporheic linkage in Sycamore Creek, Arizona. Labile organic matter is transported advectively from the surface stream to the hyporheic zone and serves as a source of organic carbon and nitrogen. Through decomposition, organic carbon is mineralized to carbon dioxide and organic nitrogen to ammonium. In the presence of oxygen ammonium can be further oxidized to nitrate. Nitrogen is then transported back to the surface stream as nitrate by hydrologic upwelling. In the surface stream, local nutrient limitation of photoautotrophs is alleviated in upwelling zones by the flux of nitrate; algal production is stimulated, carbon and nitrogen are reduced, and more organic matter is generated that is potentially transported into hyporheic sediments.

and hyporheic zones. To illustrate these couplings we present a simple model (Fig. 4) describing the relationship between algal production, hyporheic respiration, and hyporheic nitrification. Conclusions are drawn from the present study and previous research in Sycamore Creek.

Organic matter derived from the surface, presumably from algal production, is transported into hyporheic sediments through hydrologic downwelling. Biota respond to this flux of labile organic matter in downwelling zones with high respiration (Jones et al. 1995), a product of which is mineralization of organic nitrogen to NH_4 . Nitrifying organisms oxidize the ammonium and elevate nitrate concentration in the hyporheic zone (Fig. 1 and 3; Holmes et al. 1994). Nitrate-rich water flows through sediments to upwelling zones where it is transported to the surface (Grimm et al. 1991). Algal production at the surface, which is nitrogen limited (Grimm and Fisher 1986a), rapidly assimilates inorganic nitrogen (Valett et al. 1994) into organic matter. The cycle begins again when organic matter from algal production is transported to the hyporheic zone. Nitrogen is mineralized from an organic form (at the surface) to ammonium, then rapidly oxidized to nitrate (in the subsurface environment). Only nitrogen fixation and de-

nitrification represent true sources or sinks during periods between floods (baseflow) when terrestrial-aquatic interactions are minimal (Grimm 1992).

Mineralization of organic nitrogen, and hence flux of ammonium, is a consequence of organic matter decomposition (McGill and Cole 1981, Qualls and Haines 1992), and the stoichiometry of organic matter (C:N) imposes a constraint on the linkage between decomposition and nitrification (Bowden 1986). As long as organic matter is "enriched" in nitrogen relative to carbon, microbial production is presumably not limited by nitrogen and, consequently, excess nitrogen is mineralized to ammonium (i.e., organic nitrogen is not assimilated by microorganisms). Further, the rate of hyporheic respiration is dependent on import of labile organic matter from the surface to hyporheic zone as a source of carbon for heterotrophic respiration (Jones et al. 1995). Respiration is highest in downwelling zones and largely supported by organic matter derived from algae (Jones et al. 1995). Algae in Sycamore Creek have an atomic ratio of carbon to nitrogen of 9–12 (Grimm 1987), a ratio generally thought to indicate carbon limitation (Triska et al. 1975). Consequently, organic nitrogen is in excess and mineralized to ammonium.

Hyporheic decomposition of organic matter and subsequent nitrification feed back on surface algal production through transport of nitrate from hyporheic sediments to the stream surface by hydrologic exchange (Fig. 4; Grimm et al. 1991). Algae, which are nitrogen limited in Sycamore Creek (Grimm and Fisher 1986a), respond to the flux of nitrate in upwelling water with high biomass and a community composition dominated by chlorophytes (Valett et al. 1994). In contrast, downwelling zones have lower algal biomass and are frequently colonized by cyanobacteria (Valett et al. 1994).

Given high algal production at the surface (Busch and Fisher 1981) and dependence of production on hyporheic nitrogen-rich water, an important question is: how much of surface algal production and nitrogen demand is potentially supplied from hyporheic nitrification? Weighting nitrification rate by areal extent of downwelling and upwelling zones (downwelling zones 35% and upwelling zones 65% of stream area; Jones et al., 1994) and sediment depth (62 cm; Valett et al. 1990) derives a mean daily hyporheic nitrification rate over a 1-y period of $84 \text{ mgNO}_3\text{-N}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$ (range -67 to $280 \text{ mgNO}_3\text{-N}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$). Surface algal nitrogen demand averages $94 \text{ mgN}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$ (range -94 to $620 \text{ mgN}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$; Grimm and Fisher 1986a, Grimm 1992), and thus 89% of nutrient demand is potentially accounted for by hyporheic mineralization of ammonium and subsequent nitrification.

Other potential inputs of inorganic nitrogen may be derived from sources such as nitrogen fixation, advective import from upstream and deep groundwater input (i.e., stored floodwater). Grimm (1987) reported advective import of dissolved inorganic nitrogen into two $\sim 100\text{-m}$ stream reaches of Sycamore Creek to average $470 \text{ mgN}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$. Assuming hyporheic nitrification and upstream import of dissolved inorganic nitrogen are the two dominant inputs, subsurface nitrification accounts for 18% of the total dissolved inorganic nitrogen flux into the surface stream.

Surface-hyporheic linkages and ecosystem functioning

Interactions between the surface and hyporheic zone are not static, but vary temporally in response to flooding, drying, and seasons. Flash

floods in Sycamore Creek scour algae from the stream and reduce the pool of organic matter to support subsurface heterotrophic decomposition and subsequent nitrification. As algal biomass increases over succession following floods the flux of labile organic matter into the hyporheic zone rises resulting in higher respiration and nitrification rates (Jones et al. 1995; Figs. 2, 3). Hydrologic exchange between the surface and hyporheic zone also increases during succession as discharge declines (Valett et al. 1994, Jones et al. 1995); consequently, the potential for transport of materials between subsystems increases with time following storms. Seasonality also imposes an important constraint on surface-hyporheic interactions. Successional sequences following floods during spring-summer are markedly different from those occurring during autumn-winter (Grimm and Fisher 1989), potentially because of temperature constraints on microorganisms (Focht and Verstraete 1977). Thus, surface-hyporheic interactions are dependent on not only the time since flood, but also time of year.

Viewing ecosystems as a mosaic of interacting subsystems has great utility for our understanding of nutrient dynamics in streams. As nutrients spiral through streams, discrete subsystems are responsible for nutrient retention whereas others function to release nutrients. An interdependence between surface and hyporheic zones is not restricted to desert streams, but occurs in others. As an example, Triska et al. (1989a, 1989b, 1990, 1993) reported similar processes in Little Lost Man Creek, in northwestern California, where nitrate is elevated in hyporheic water by nitrification, the hyporheic zone serves as a source of inorganic nitrogen for the surface stream, and nitrate is assimilated in the surface stream by microorganisms. Whether a subsystem serves as a source or sink for nitrogen is likely determined by the origin of organic matter. In the hyporheic zone of Sycamore Creek, organic matter is predominantly derived from algae and consequently enriched in nitrogen (i.e., low atomic ratio of carbon to nitrogen). In contrast, subsystems in which most organic matter is derived from terrestrial vegetation and thus enriched in carbon (i.e., high atomic ratio of carbon to nitrogen) probably function as sinks for nitrogen. A complete understanding of nutrient dynamics requires expanding definitions of streams to incorporate not only the hypor-

heic but also the parafluvial zone (Holmes et al. 1994) and riparian zone (Peterjohn and Correll 1984, Lowrance et al. 1985, Cooper 1990, Hill 1990).

Acknowledgements

Thanks to M. Mallett, D. Greene, and A. Wiebel for help in the laboratory and field. Thanks also to R. M. Holmes for comments and discussion throughout this project, and to Drs. C. N. Dahm, J. J. Elser, W. L. Minckley, R. J. Mackay, A. D. Steinman, and two anonymous reviewers for constructive comments on the manuscript. In addition, we thank Mr. John Whitney for access to Dos S Ranch at Sycamore Creek. This research was supported by National Science Foundation's Ecosystem Studies Program (grants DEB-9224059 to JBJ and SGF, and BSR-8818612, DEB-9108362, and DEB-9306909 to SGF and NBG).

Literature Cited

- BOWDEN, W. B. 1986. Nitrification, nitrate reduction, and nitrogen immobilization in a tidal freshwater marsh sediment. *Ecology* 67:88-99.
- BUSCH, D. E., AND S. G. FISHER. 1981. Metabolism of a desert stream. *Freshwater Biology* 11:301-307.
- COOPER, A. B. 1990. Nitrate depletion in the riparian zone and stream channel of a small headwater catchment. *Hydrobiologia* 202:13-26.
- DUFF, J. H., AND F. J. TRISKA. 1990. Denitrification in sediments from the hyporheic zone adjacent to a small forested stream. *Canadian Journal of Fisheries and Aquatic Sciences* 47:1140-1147.
- ELWOOD, J. W., J. D. NEWBOLD, A. F. TRIMBLE, AND R. W. STARK. 1981. The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary production. *Ecology* 62:146-158.
- FISHER, S. G., L. J. GRAY, N. B. GRIMM, AND D. E. BUSCH. 1982. Temporal succession in a desert stream ecosystem following flash flooding. *Ecological Monographs* 52:92-110.
- FISHER, S. G., AND N. B. GRIMM. 1985. Hydrologic and material budgets for a small Sonoran Desert watershed during three successive cloudburst floods. *Journal of Arid Environments* 9:105-118.
- FOCHT, D. D., AND W. VERSTRAETE. 1977. Biochemical ecology of nitrification and denitrification. *Advances in Microbial Ecology* 1:135-214.
- FORD, T. E., AND R. J. NAIMAN. 1989. Groundwater-surface water relationships in boreal forest watersheds: dissolved organic carbon and inorganic nutrient dynamics. *Canadian Journal of Fisheries and Aquatic Sciences* 46:41-49.
- GRIMM, N. B. 1987. Nitrogen dynamics during succession in a desert stream. *Ecology* 68:1157-1170.
- GRIMM, N. B. 1992. Biogeochemistry of nitrogen in Sonoran Desert streams. *Journal of the Arizona-Nevada Academy of Science* 26:139-155.
- GRIMM, N. B., AND S. G. FISHER. 1986a. Nitrogen limitation in a Sonoran Desert stream. *Journal of the North American Benthological Society* 5:2-15.
- GRIMM, N. B., AND S. G. FISHER. 1986b. Nitrogen limitation potential of Arizona streams and rivers. *Journal of the Arizona-Nevada Academy of Science* 21:31-43.
- GRIMM, N. B., AND S. G. FISHER. 1989. Stability of periphyton and macroinvertebrates to disturbance by flash floods in a desert stream. *Journal of the North American Benthological Society* 8:293-307.
- GRIMM, N. B., H. M. VALETT, E. H. STANLEY, AND S. G. FISHER. 1991. Contribution of the hyporheic zone to stability of an arid-land stream. *Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte Limnologie* 24:1595-1599.
- HENDRICKS, S. P., AND D. S. WHITE. 1991. Physicochemical patterns within a hyporheic zone of a northern Michigan river, with comments on surface water patterns. *Canadian Journal of Fisheries and Aquatic Sciences* 48:1645-1654.
- HILL, A. R. 1990. Ground water flow paths in relation to nitrogen chemistry in the near-stream zone. *Hydrobiologia* 206:39-52.
- HOLMES, R. M., S. G. FISHER, AND N. B. GRIMM. 1994. Parafluvial nitrogen dynamics in a desert stream ecosystem. *Journal of the North American Benthological Society* 13:468-478.
- HOWARTH, R. W., R. MARINO, AND J. J. COLE. 1988. Nitrogen fixation in freshwater, estuarine, and marine ecosystems. 2. Biogeochemical controls. *Limnology and Oceanography* 33:688-701.
- JONES, J. B., S. G. FISHER, AND N. B. GRIMM. 1995. Vertical hydrologic exchange and ecosystem metabolism in a Sonoran Desert stream. *Ecology* 76:942-952.
- JONES, J. B., R. M. HOLMES, S. G. FISHER, AND N. B. GRIMM. 1994. Chemoautotrophic production and respiration in the hyporheic zone of a Sonoran Desert stream. Pages 329-338 in J. A. Stanford and H. M. Valett (editors). *Proceedings of the Second International Conference on Ground Water Ecology*. American Water Resources Association, Herndon, Virginia.
- LEE, D. R., AND J. A. CHERRY. 1978. A field exercise on groundwater flow using seepage meters and mini-piezometers. *Journal of Geological Education* 27:6-10.

- LOWRANCE, R. R., R. A. LEONARD, AND L. E. ASMUSSEN. 1985. Nutrient budgets for agricultural watersheds in the southeastern coastal plain. *Ecology* 66:287-296.
- MANNY, B. A., M. C. MILLER, AND R. G. WETZEL. 1971. Ultraviolet combustion of dissolved organic nitrogen compounds in lake waters. *Limnology and Oceanography* 16:71-85.
- MCGILL, W. B., AND C. V. COLE. 1981. Comparative aspects of cycling of organic C, N, S, and P through soil organic matter. *Geoderma* 26:267-286.
- MENZEL, D. W., AND F. R. VACCARO. 1964. The measurement of dissolved and particulate organic carbon in seawater. *Limnology and Oceanography* 9:138-142.
- MURPHY, J., AND J. P. RILEY. 1962. Determination of phosphate in natural water. *Annals Chemica Acta* 27:31-36.
- NEWBOLD, J. D., J. W. ELWOOD, R. V. O'NEILL, AND A. L. SHELDON. 1983. Phosphorus dynamics in a woodland stream ecosystem: a study of nutrient spiralling. *Ecology* 64:1249-1265.
- PETERJOHN, W. T., AND D. L. CORRELL. 1984. Nutrient dynamics in an agricultural watershed: observations on the role of a riparian forest. *Ecology* 65:1466-1475.
- QUALLS, R. G., AND B. L. HAINES. 1992. Biodegradability of dissolved organic matter in forest throughfall, soil solution, and stream water. *Soil Science Society of America Journal* 56:578-586.
- RUTHERFORD, J. E., AND H. B. N. HYNES. 1987. Dissolved organic carbon in streams and groundwater. *Hydrobiologia* 154:33-48.
- SOLORZANO, L. 1969. Determination of ammonium in natural water by the phenolhypochlorite method. *Limnology and Oceanography* 14:799-801.
- THOMSEN, B. W., AND H. H. SCHUMANN. 1968. Water resources of the Sycamore Creek watershed, Maricopa County, Arizona. U.S. Geological Survey Water-Supply Paper 1861.
- TRISKA, F. J., J. H. DUFF, AND R. J. AVANZINO. 1990. Influence of exchange flow between the channel and hyporheic zone on nitrate production in a small mountain stream. *Canadian Journal of Fisheries and Aquatic Sciences* 47:2099-2111.
- TRISKA, F. J., J. H. DUFF, AND R. J. AVANZINO. 1993. Patterns of hydrologic exchange and nutrient transformation in the hyporheic zone of a gravel-bottom stream: examining terrestrial-aquatic linkages. *Freshwater Biology* 29:259-274.
- TRISKA, F. J., V. C. KENNEDY, R. J. AVANZINO, G. W. ZELLWEGER, AND K. E. BENCALA. 1989a. Retention and transport of nutrient in a third-order stream in northwestern California: hyporheic processes. *Ecology* 70:1893-1905.
- TRISKA, F. J., V. C. KENNEDY, R. J. AVANZINO, G. W. ZELLWEGER, AND K. E. BENCALA. 1989b. Retention and transport of nutrients in a third-order stream: channel processes. *Ecology* 70:1877-1892.
- TRISKA, F. J., J. R. SEDELL, AND B. BUCKLEY. 1975. The processing of conifer and hardwood leaves in two coniferous forest streams. II. biochemical and nutrient changes. *Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte Limnologie* 19:1628-1639.
- VALETT, H. M., S. G. FISHER, N. B. GRIMM, AND P. CAMILL. 1994. Vertical hydrologic exchange and ecological stability of a desert stream ecosystem. *Ecology* 75:548-560.
- VALETT, H. M., S. G. FISHER, AND E. H. STANLEY. 1990. Physical and chemical characteristics of the hyporheic zone of a Sonoran Desert stream. *Journal of the North American Benthological Society* 9:201-215.
- WILKINSON, L. 1990. SYSTAT: the system for statistics. SYSTAT, Inc., Evanston, Illinois, USA.
- WOOD, E. D., F. A. J. ARMSTRONG, AND F. A. RICHARDS. 1967. Determination of nitrate in seawater by cadmium-copper reduction to nitrate. *Journal of the Marine Biology Association of the United Kingdom* 47:23-31.

Received: 12 August 1994

Accepted: 11 December 1994

LINKED CITATIONS

- Page 1 of 4 -



You have printed the following article:

Nitrification in the Hyporheic Zone of a Desert Stream Ecosystem

Jeremy B. Jones, Jr.; Stuart G. Fisher; Nancy B. Grimm

Journal of the North American Benthological Society, Vol. 14, No. 2. (Jun., 1995), pp. 249-258.

Stable URL:

<http://links.jstor.org/sici?sici=0887-3593%28199506%2914%3A2%3C249%3ANITHZO%3E2.0.CO%3B2-A>

This article references the following linked citations. If you are trying to access articles from an off-campus location, you may be required to first logon via your library web site to access JSTOR. Please visit your library's website or contact a librarian to learn about options for remote access to JSTOR.

Literature Cited

Nitrification, Nitrate Reduction, and Nitrogen Immobilization in a Tidal Freshwater Marsh Sediment

William B. Bowden

Ecology, Vol. 67, No. 1. (Feb., 1986), pp. 88-99.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198602%2967%3A1%3C88%3ANNRANI%3E2.0.CO%3B2-4>

The Limiting Role of Phosphorus in a Woodland Stream Ecosystem: Effects of P Enrichment on Leaf Decomposition and Primary Producers

Jerry W. Elwood; J. Denis Newbold; Ann F. Trimble; Robert W. Stark

Ecology, Vol. 62, No. 1. (Feb., 1981), pp. 146-158.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198102%2962%3A1%3C146%3ATLROPI%3E2.0.CO%3B2-1>

Temporal Succession in a Desert Stream Ecosystem Following Flash Flooding

Stuart G. Fisher; Lawrence J. Gray; Nancy B. Grimm; David E. Busch

Ecological Monographs, Vol. 52, No. 1. (Mar., 1982), pp. 93-110.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9615%28198203%2952%3A1%3C93%3ATSIADS%3E2.0.CO%3B2-6>

LINKED CITATIONS

- Page 2 of 4 -



Nitrogen Dynamics During Succession in a Desert Stream

Nancy B. Grimm

Ecology, Vol. 68, No. 5. (Oct., 1987), pp. 1157-1170.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198710%2968%3A5%3C1157%3ANDDSIA%3E2.0.CO%3B2-C>

Nitrogen Limitation in a Sonoran Desert Stream

Nancy B. Grimm; Stuart G. Fisher

Journal of the North American Benthological Society, Vol. 5, No. 1. (Mar., 1986), pp. 2-15.

Stable URL:

<http://links.jstor.org/sici?sici=0887-3593%28198603%295%3A1%3C2%3ANLIASD%3E2.0.CO%3B2-Z>

Stability of Periphyton and Macroinvertebrates to Disturbance by Flash Floods in a Desert Stream

Nancy B. Grimm; Stuart G. Fisher

Journal of the North American Benthological Society, Vol. 8, No. 4. (Dec., 1989), pp. 293-307.

Stable URL:

<http://links.jstor.org/sici?sici=0887-3593%28198912%298%3A4%3C293%3ASOPAMT%3E2.0.CO%3B2-5>

Parafluvial Nitrogen Dynamics in a Desert Stream Ecosystem

Robert M. Holmes; Stuart G. Fisher; Nancy B. Grimm

Journal of the North American Benthological Society, Vol. 13, No. 4. (Dec., 1994), pp. 468-478.

Stable URL:

<http://links.jstor.org/sici?sici=0887-3593%28199412%2913%3A4%3C468%3APNDIAD%3E2.0.CO%3B2-X>

Nitrogen Fixation in Freshwater, Estuarine, and Marine Ecosystems. 2. Biogeochemical Controls

Robert W. Howarth; Roxanne Marino; Jonathan J. Cole

Limnology and Oceanography, Vol. 33, No. 4, Part 2: Comparative Ecology of Freshwater and Marine Ecosystems. (Jul., 1988), pp. 688-701.

Stable URL:

<http://links.jstor.org/sici?sici=0024-3590%28198807%2933%3A4%3C688%3ANFIFEAE%3E2.0.CO%3B2-Y>

Vertical Hydrologic Exchange and Ecosystem Metabolism in a Sonoran Desert Stream

Jeremy B. Jones, Jr.; Stuart G. Fisher; Nancy B. Grimm

Ecology, Vol. 76, No. 3. (Apr., 1995), pp. 942-952.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28199504%2976%3A3%3C942%3AVHEAEM%3E2.0.CO%3B2-D>

LINKED CITATIONS

- Page 3 of 4 -



Nutrient Budgets for Agricultural Watersheds in the Southeastern Coastal Plain

R. Richard Lowrance; Ralph A. Leonard; Loris E. Asmussen; Robert L. Todd

Ecology, Vol. 66, No. 1. (Feb., 1985), pp. 287-296.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198502%2966%3A1%3C287%3ANBFAWI%3E2.0.CO%3B2-Y>

Ultraviolet Combustion of Dissolved Organic Nitrogen Compounds in Lake Waters

B. A. Manny; M. C. Miller; R. G. Wetzel

Limnology and Oceanography, Vol. 16, No. 1. (Jan., 1971), pp. 71-85.

Stable URL:

<http://links.jstor.org/sici?sici=0024-3590%28197101%2916%3A1%3C71%3AUCODON%3E2.0.CO%3B2-M>

The Measurement of Dissolved Organic and Particulate Carbon in Seawater

David W. Menzel; Ralph F. Vaccaro

Limnology and Oceanography, Vol. 9, No. 1. (Jan., 1964), pp. 138-142.

Stable URL:

<http://links.jstor.org/sici?sici=0024-3590%28196401%299%3A1%3C138%3ATMODOA%3E2.0.CO%3B2-F>

Phosphorus Dynamics in a Woodland Stream Ecosystem: A Study of Nutrient Spiralling

J. D. Newbold; J. W. Elwood; R. V. O'Neill; A. L. Sheldon

Ecology, Vol. 64, No. 5. (Oct., 1983), pp. 1249-1265.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198310%2964%3A5%3C1249%3APDIAWS%3E2.0.CO%3B2-7>

Nutrient Dynamics in an Agricultural Watershed: Observations on the Role of A Riparian Forest

William T. Peterjohn; David L. Correll

Ecology, Vol. 65, No. 5. (Oct., 1984), pp. 1466-1475.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198410%2965%3A5%3C1466%3ANDIAAW%3E2.0.CO%3B2-A>

Determination of Ammonia in Natural Waters by the Phenolhypochlorite Method

Lucia Solorzano

Limnology and Oceanography, Vol. 14, No. 5. (Sep., 1969), pp. 799-801.

Stable URL:

<http://links.jstor.org/sici?sici=0024-3590%28196909%2914%3A5%3C799%3ADOAINW%3E2.0.CO%3B2-7>

LINKED CITATIONS

- Page 4 of 4 -



Retention and Transport of Nutrients in a Third-Order Stream in Northwestern California: Hyporheic Processes

Frank J. Triska; Vance C. Kennedy; Ronald J. Avanzino; Gary W. Zellweger; Kenneth E. Bencala
Ecology, Vol. 70, No. 6. (Dec., 1989), pp. 1893-1905.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198912%2970%3A6%3C1893%3ARATONI%3E2.0.CO%3B2-M>

Retention and Transport of Nutrients in a Third-Order Stream: Channel Processes

Frank J. Triska; Vance C. Kennedy; Ronald J. Avanzino; Gary W. Zellweger; Kenneth E. Bencala
Ecology, Vol. 70, No. 6. (Dec., 1989), pp. 1877-1892.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28198912%2970%3A6%3C1877%3ARATONI%3E2.0.CO%3B2-O>

Vertical Hydrologic Exchange and Ecological Stability of a Desert Stream Ecosystem

H. Maurice Valett; Stuart G. Fisher; Nancy B. Grimm; Philip Camill
Ecology, Vol. 75, No. 2. (Mar., 1994), pp. 548-560.

Stable URL:

<http://links.jstor.org/sici?sici=0012-9658%28199403%2975%3A2%3C548%3AVHEAES%3E2.0.CO%3B2-1>

Physical and Chemical Characteristics of the Hyporheic Zone of a Sonoran Desert Stream

H. Maurice Valett; Stuart G. Fisher; Emily H. Stanley
Journal of the North American Benthological Society, Vol. 9, No. 3. (Sep., 1990), pp. 201-215.

Stable URL:

<http://links.jstor.org/sici?sici=0887-3593%28199009%299%3A3%3C201%3APACCOT%3E2.0.CO%3B2-C>