Verh. Internat. Verein. Limnol.
2009, vol. 30, Part 5, p. 809–812, Stuttgart, January 2009


# Quantifying riverine sediment denitrification rates *in situ* with a novel, portable hyporheic chamber

Tara M. Higgins, James H. McCutchan, Jr. and William M. Lewis, Jr.

## Introduction

As global concerns over nitrogen loading to freshwaters have grown, so too has interest in understanding the biogeochemical processes comprising the nitrogen cycle. Of particular interest is denitrification, which represents the only permanent loss of nitrogen in the aquatic nitrogen cycle, through the microbial reduction of nitrate to inert $N_2$ gas (plus <1 % $N_2O$). Running-water ecosystems can support high rates of denitrification (Piña-Ochoa & Álvarez-Cobelas 2006), but the factors controlling rates of denitrification *in situ* are not thoroughly understood (Davidson & Seitzinger 2006).

A complete understanding of the controls on denitrification in running waters has been hampered by high spatial variability in rates of denitrification and limitations of available methods for quantifying rates (Groffman et al. 2006). Whole-reach methods (McCutchan et al. 2003, Mulholland et al. 2004, Pribyl et al. 2005) integrate rates of denitrification in spatially heterogeneous environments and can provide high precision with modest effort, but these methods may not be well suited to the development of a mechanistic understanding of denitrification. The alternative approach, using sediment incubations in laboratory chambers (Pfenning & McMahon 1996, Bernot et al. 2003), can be problematic due to the numerous artifacts and errors associated with simulating natural conditions.

This paper describes a novel hyporheic chamber for quantifying denitrification rates *in situ*. Rates are estimated in a flow-through chamber from changes in mass balance for nitrogen along a simulated hyporheic flow-path. The hyporheic chamber described here can be used to study spatial heterogeneity in rates of denitrification in river sediments at a fine scale, but also can be used to quantify the factors that control rates of denitrification in sediments through experimental manipulations in a field setting. By measuring denitrification rates *in situ* in riverine sediments, the hyporheic chamber avoids many of the difficulties associated with replicating natural biogeochemical conditions in a laboratory.

**Key words:** chamber, denitrification, hyporheic zone, nitrate, sediment core, South Platte River

## Materials and methods

The hyporheic chamber consists of a 6-m long, 48.6-L PVC pipe (10.16 cm i.d.; Fig. 1a). These dimensions provide a good balance between sustaining the necessary biogeochemical conditions for denitrification to proceed and portability of the chamber. Preliminary trials demonstrated that shorter chambers developed reducing conditions only at extremely low flow velocities, while larger chambers proved difficult to maneuver. If necessary, 2 or more 6-m chambers can be connected end-to-end to simulate a longer hyporheic flow-path. The chamber was filled on site with hyporheic sediments from the South Platte River and suspended horizontally in the river in the direction of flow. The device was deployed in the South Platte River near Fort Lupton (40°10'N; 104°50'W), 53 km downstream of Denver, Colorado, USA, during October and November 2006 (detailed site description given by Pribyl et al. 2005). Care was taken to ensure that sediments within the chamber were packed to avoid the formation of flow channels along the sides of the chamber. Ports were installed at 1-m intervals to allow sampling of sediment pore water for dissolved oxygen and nutrient concentrations.

Flow velocity through the chamber was determined by a salt tracer test (Fig. 1b); an average velocity of 0.0093 cm/s (18-h travel time) was maintained by use of a rotating disk in the upstream cap will adjustable inflow holes and a valve at the outflow (Fig. 1a). The conductance profile from the salt tracer test indicated that water passed though the chamber steadily as a single parcel, with only moderate longitudinal dispersion (Fig. 1b).

Concentration measurements of dissolved oxygen (DO), nitrate, ammonium, total dissolved phosphorus (TDP), and dissolved organic carbon (DOC) were made on 13 October, 25 October, 8 November, 20 November, and 22 November at the point of inflow (0 m), at 2 m and at 4 m along the chamber, and at the outflow (6 m) from the chamber. Porewater samples were extracted from the chamber via the sampling ports with a hand-operated peristaltic pump; at 6 m water was collected from a bag attached to the outflow valve (Fig. 1a). All water samples for analysis of dissolved nutrients were filtered through glass-fiber (GF/C) filters within 6 h of collec-

0368-0770/09/0809 $ 1.00

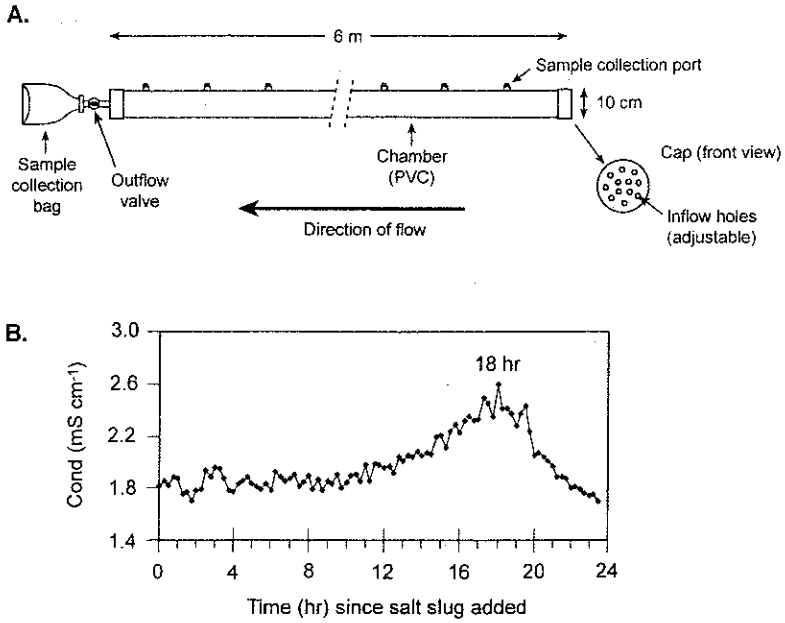


**Fig. 1.** (a) Schematic diagram of the hyporheic chamber, and (b) results of a conductivity tracer test used to measure travel time through the hyporheic chamber.

tion. Nitrate was analyzed with an ion chromatograph. Ammonium and TDP were analysed colorimetrically (Murphy & Riley 1962, Grashoff 1976) and DOC was analysed by carbon analyzer. On each sampling occasion, measurements of pH, specific conductance, temperature, and dissolved oxygen in the river channel also were taken with the multi-parameter meter. Grain size, organic content, and porosity of the hyporheic sediments were determined by standard methods (APHA 1998).

## Results

During the trials, nitrate concentrations decreased by an average of 43 % (maximum, 66 %) along the length of the chamber (Fig. 2a), from 6.56 to 3.83 mg $NO_3^-$-N $L^{-1}$. Thus, the 6-m chamber was successful in simulating conditions in the hyporheic zone that support microbial denitrification, as measured previously by mass-balance methods (Pribyl et al. 2005). As expected, decreases in nitrate were very strongly correlated with reductions in dissolved oxygen ($r^2$ = 0.81, P < 0.0001, n = 16) along the length of the chamber, demonstrating the importance of reducing conditions for denitrification to proceed (Cey et al. 1999). The decline in nitrate began before oxygen in the chamber was completely exhausted. While the majority of water passed through the chamber in 18 h, travel times ranged from <12 h to >20 h (Fig. 1b). Decreases in nitrate were not significantly correlated with changes in TDP ($r^2$ = 0.05, P = 0.45), DOC ($r^2$ = 0.23, P = 0.08) or ammonium ($r^2$ = 0.05, P = 0.34; all n = 16) along the length of the chamber (Fig. 2c-e).

Based on the assumption that denitrification was limited to the upper 50 cm of the sediments in the South Platte River, the mean reduction in nitrate recorded during this study corresponds to an areal denitrification rate of 0.41 g N $m^{-2}$ $d^{-1}$ for October–November 2006. This value is similar to published estimates of late-fall early-winter denitrification rates for this region of the South Platte (Pribyl el al. 2005).

## Discussion

The South Platte River downstream of Denver has high rates of denitrification (Pribyl et al. 2005). These high rates are driven by high concentrations of nitrate and labile organic carbon and coarse alluvial sediments that favor hyporheic metabolism (Table 1). These characteristics make the South Platte River an ideal location in which to test the application of our hyporheic chamber design for quantifying denitrification.

Rates of denitrification reported here were calculated from the disappearance of nitrate along the hyporheic flow path in the chamber. While assimilatory nitrate reduction might have resulted in the disappearance of nitrate in the absence of denitrification, the high ambient nitrate concentrations within the chamber (Fig. 2c) make it unlikely that processes other than denitrification were responsible for the observed nitrate decrease. Rates of denitrification also could be calculated from the production of $N_2$ in the chambers as measured with membrane-inlet mass spectrometry (MIMS) or by a combination of approaches.

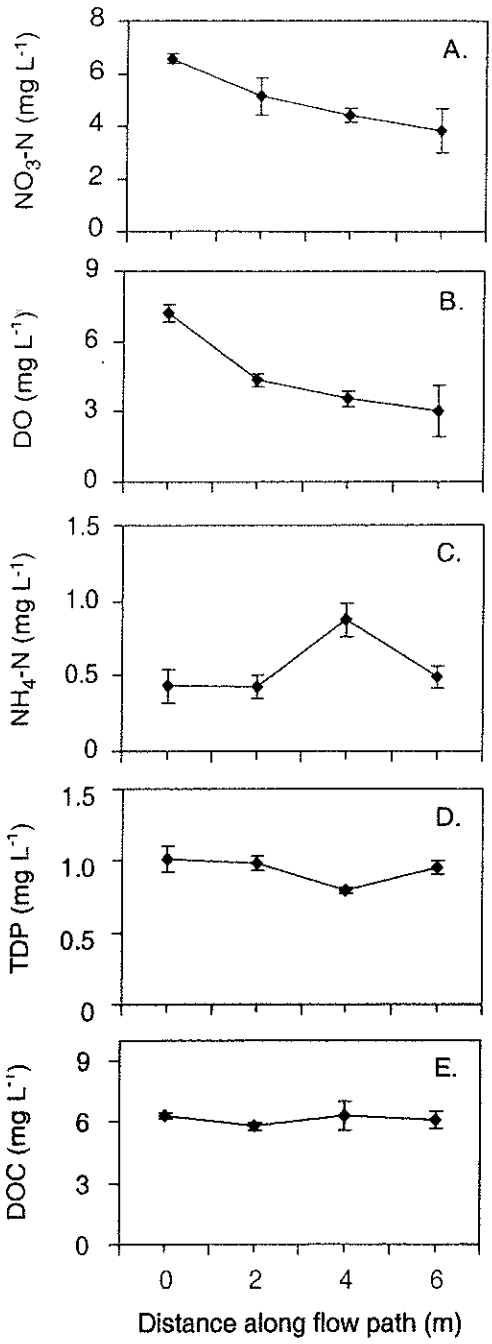


**Fig. 2.** Changes in concentrations of (a) nitrate, (b) dissolved oxygen, (c) ammonia, (d) total dissolved phosphorus, and (e) total organic carbon with distance along the 6-m hyporheic chamber. Values shown are means of 5 measurements taken in October and November 2006; error bars show standard errors. (Fig. 1)

These initial trials demonstrate the potential of the hyporheic chamber as a portable, economical, and accurate method for quantifying denitrification rates *in situ* in riverine sediments. Such an approach avoids many of the artefacts associated with replicating natural biogeochemical conditions in a laboratory. Its low cost and simple design would allow multiple chambers to be installed within a single study site to account for the high spatial heterogeneity in denitrification rates (Saunders & Lewis 2001). While beyond the scope of the current investigation, the design of the hyporheic chamber also facilitates easy manipulation of a range of environmental variables (travel time, quantity and quality of organic carbon, nitrate concentration), allowing the factors controlling denitrification to be studied both singly and in combination.

**Table 1.** Characteristics of the South Platte River at Fort Lupton, Colorado, during October-November 2006. Values shown are means ± standard errors (n = 5).

| | |
|---|---|
| pH | 7.94 ± 0.1 |
| Temperature (°C) | 12.5 ± 1.3 |
| Dissolved oxygen (mg $L^{-1}$) | 7.21 ± 0.3 |
| Conductance (µS $cm^{-1}$) | 1119 ± 40 |
| DIN (mg $L^{-1}$) | 7.1 ± 0.17 |
| TDP (mg $L^{-1}$) | 1.01 ± 0.09 |
| DOC (mg $L^{-1}$) | 6.29 ± 0.1 |
| Sediment porosity | 27.3 % |
| Sediment organic content | 0.43 % |
| Sediment particle sizes: | |
| Gravel (> 2.36 mm) | 34 % |
| Sand (0.25–2.36 mm) | 60 % |
| Silt/clay (< 0.25 mm) | 6 % |

## Acknowledgements

This work was supported by a Fulbright scholarship sponsored by Teagasc, the Irish Agriculture and Food Development Authority.

## References

[APHA] American Public Health Association. 1998. Standard Methods for the Examination of Water and Wastewater, 20th ed. American Water Work Association, Water Environment Federation.

Bernot, M.J., W.K. Dodds, W.S. Gardner, M.J. McCarthy, D. Sobolev & J.L. Tank. 2003. Comparing denitrification estimates for a Texas estuary by using acetylene inhibition and membrane inlet mass spectrometry. Appl. Environ. Microbiol. **69:** 5950–5956.

Cey, E.E., D.L. Rudolph, G.W. Parkin & R. Aravena. 1999. Quantifying groundwater discharge to a small perennial stream in southern Ontario. Can. J. Hydrol. **210:** 21–37.

Davidson, E.A. & A. Seitzinger. 2006. The enigma of progress in denitrification research. Ecol. Appl. **16:** 2057–2063.

Groffman, P.M., M.A. Altabet, J.K. Bohlke, K. Butterbach-Bahl, M.B. David, M.K. Firestone, A.E. Giblin, T.M. Kana, L.P. Nielson & M.A. Voytek. 2006. Methods for measuring denitrification: diverse approaches to a difficult problem. Ecol. Appl. **16:** 2091–2122.

Grashoff, K. 1976. Determination of dissolved oxygen and nutrients, p. 126–137. *In* K. Grashoff, M. Ehrhardt & K. Kremling [eds], Methods of Seawater Analysis. Verlag Chemie.

McCutchan, J.H., J.F.I. Saunders, A.L. Pribyl & W.M. Lewis. 2003. Open-channel estimation of denitrification. Limnol. Oceanogr: Methods **1:** 74–81.

Mulholland, P.J., H.M. Valett, J.R. Webster, S.A. Thomas, L.W. Cooper, S.K. Hamilton & B.J. Peterson. 2004. Stream denitrification and total nitrate uptake rates measured using a field $^{15}N$ tracer addition approach. Limnol. Oceanogr. **49:** 809–820.

Murphy, J. & J.P. Riley. 1962. A modified single solution method for the determination of phosphate in natural waters. Anal. Chim. Acta **27:** 31–36.

Pfenning, K.S. & P.B. McMahon. 1996. Effect of nitrate, organic carbon and temperature on potential denitrification rates in nitrate-rich riverbed sediments. J. Hydrol. **187:** 283–295.

Piña-Ochoa, E. & M. Álvarez-Cobelas. 2006. Denitrification in aquatic environments: a cross-system analysis. Biogeochemistry **81:** 111–130.

Pribyl, A.L., J.H. McCutchan, W.M. Lewis & J.F. Saunders. 2005. Whole-system estimation of denitrification in a plains river: a comparison of two methods. Biogeochemistry **73:** 439–455.

Saunders, J.F. & W.M. Lewis. 2001. Denitrification rates in the sediments of Lake Memphemagog, Canada-USA. Water Res. **35:** 1879–1904.

**Authors' addresses:** T.M. Higgins, Center for Limnology, Cooperative Institute for Research in Environmental Sciences, University of Colorado, Boulder, Colorado 80309-0216, USA.
Current address: Department of Zoology, National University of Ireland, Galway, Ireland. Email: tara.higgins@nuigalway.ie
J.H. McCutchan, Jr., W.M. Lewis, Jr., Center for Limnology, Cooperative Institute for Research in Environmental Sciences, University of Colorado, Boulder, Colorado 80309-0216, USA