Verh. Internat. Verein. Limnol.
2008, vol. **30**, Part **2**, p. 000–000, Stuttgart, April 2008

Spatial and temporal patterns of denitrification in an effluent-dominated plains river

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

Introduction

Denitrification, the microbial reduction of nitrate to gaseous forms (primarily N_2 but also N_2O), is an important mechanism for the removal of fixed N from aquatic systems. Although denitrification rates tend to be higher in rivers than in other aquatic environments, rates of denitrification in rivers are highly variable (Piña-Ochoa & Álvarez-Cobelas 2006). Efficient denitrification in river sediments requires that sufficient nitrate and labile organic matter occur in combination with the proper redox conditions. Temperature also may limit the rate of denitrification, and seasonal changes in denitrification rates often are driven by temperature (e.g., Pfenning & McMahon 1996). Denitrification can occur over large areas of a stream channel or may be limited to micro-sites that include the right combination of conditions. If any one of the requirements for denitrification (nitrate, organic matter, redox conditions, temperature) at a particular location is insufficient, however, rates will be suppressed.

Because the potentially limiting factors for denitrifying bacteria vary spatially and temporally within river networks, rates of denitrification can vary spatially and temporally, even over short periods of time and over short distances. Whole-reach estimates of denitrification are possible with isotopic tracers (e.g., Mulholland et al. 2004), but estimates with ^{15}N have been limited to small streams due to the prohibitive cost of isotopic tracer additions in large rivers. Whole-reach estimates of denitrification also are possible through mass balance of transport and transformation rates (Hill 1981, Sjodin et al. 1997, Pribyl et al. 2005), but accumulation of measurement errors can affect the precision for estimates of denitrification with this approach (Cornwell et al. 1999). Recently, an open-channel N_2 approach has been developed for the estimation of denitrification in running waters (Laursen & Seitzinger 2002, McCutchan et al. 2003). This method, which is analogous to the open-channel method for estimation of oxygen metabolism, has been tested extensively on the South Platte River in Colorado (Pribyl 2002, McCutchan et al. 2003, Pribyl et al. 2005). The open-channel method provides high precision and is well suited to the study of spatial and temporal patterns of denitrification at the reach scale.

The purpose of this study is to describe the spatial and temporal patterns of denitrification in the South Platte River below Denver, Colorado. Although the open-channel N_2 method has simplified estimation of denitrification, there are still relatively few system-level estimates of denitrification for running waters (Piña-Ochoa & Álvarez-Cobelas 2006). Examination of the spatial and temporal patterns of denitrification in the South Platte River may contribute to a better understanding of the controls on denitrification in running waters and may improve predictions of denitrification across a wide range of running waters.

Key words: denitrification, dissolved organic carbon, open-channel method, river, spatial patterns, temperature

Study site

The South Platte River flows from the southern Rocky Mountains onto the Great Plains south of Denver, Colorado (Fig. 1). The flow regime of the South Platte is dominated by snowmelt runoff but has been modified by transbasin diversions and by a series of storage reservoirs upstream of Denver. Municipal wastewater from the city of Denver and agricultural runoff further augment the flow of the river downstream (Knopf & Scott 1990, Saunders & Lewis 2003, Cronin et al. 2007). Over the 69-km reach from 64th Avenue, just upstream of Denver's wastewater treatment outfall, to the confluence with St. Vrain Creek (Fig. 1), the South Platte flows over a bed of coarse sand and fine gravel at an average gradient of 0.0016 m m^{-1}. Near Denver, where the river flows through an urban setting, the channel has been substantially modified to maintain bank stability. Downstream, the channel is wider and shallower and is freer to meander naturally over its floodplain.

Nutrient concentrations in the South Platte below Denver are high (Fig. 2). During the study period, the concentration of nitrate-N increased gradually over the first 30 km of the study reach. High rates of nitrification account for much of the increase in nitrate concentration and for the concurrent decrease in the concentration of ammonia-N (Pribyl 2002, Pribyl et al. 2005). Concentrations of soluble reactive phosphorus and dis-

0368-0770/08/1531 $ 0.00

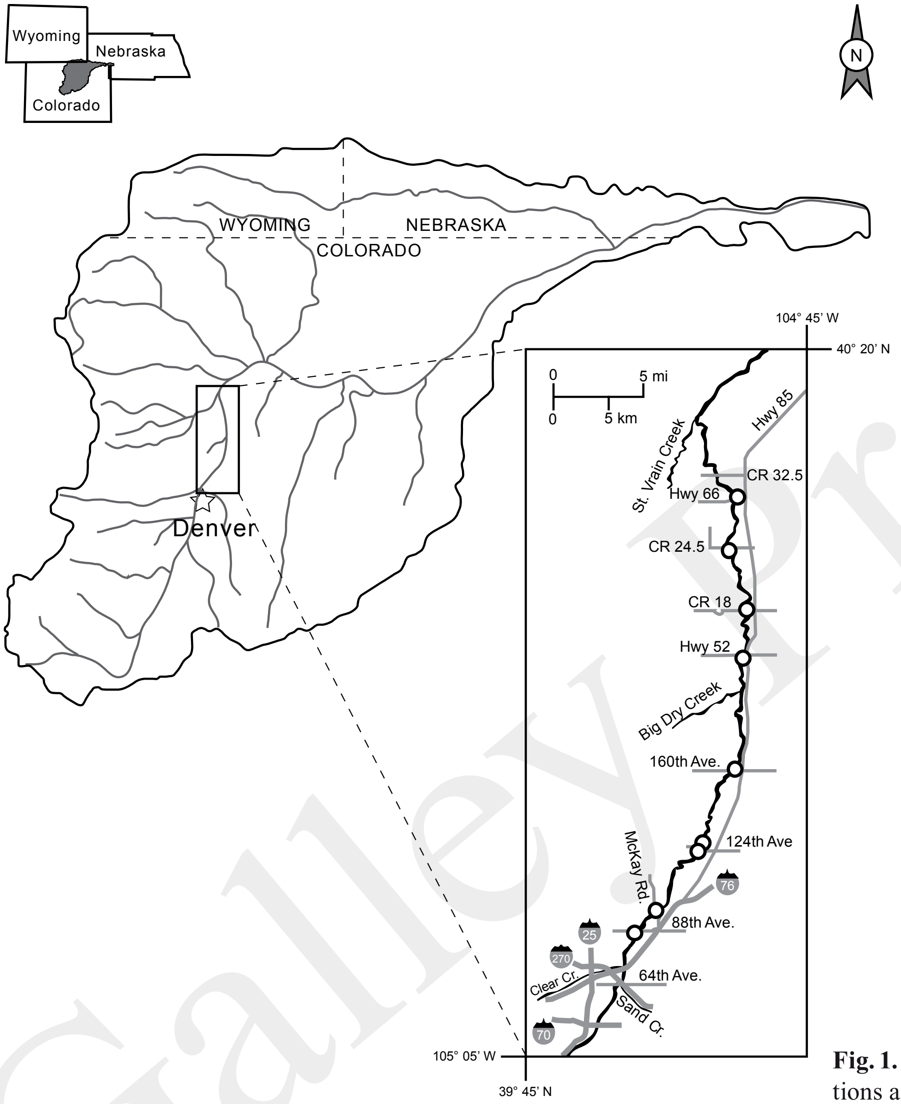

Fig. 1. Map of the study reach. Sampling locations are indicated by circles.

solved organic carbon (DOC) also declined in the downstream direction. Nutrient concentrations varied seasonally, with annual minima occurring in June, around the time of peak snowmelt runoff, and maxima occurring between December and March, when discharge and rates of biological activity were low.

Methods

Rates of denitrification were estimated by a single-station application of the open-channel N_2 method (McCutchan et al. 2003, Pribyl et al. 2005, McCutchan & Lewis 2006) within a 69-km reach of the South Platte River (Fig. 1). Concentrations of dissolved N_2 were measured by membrane-inlet mass spectrometry on 8 dates over a 12-month period (Jul 2004-Jun 2005). Depth, velocity, rate of seepage accrual, reaeration coefficient, temperature, barometric pressure, and the concentration of N_2 in groundwater also were measured, as necessary for open-channel estimation of denitrification.

Channel geometry, discharge, and reaeration

The study reach was divided into short (typically 30–300 m) subreaches for the purpose of modeling spatial changes in channel geometry, discharge, groundwater seepage, and the reaeration coefficient on each sampling date. Channel-geometry relationships were available for 3 locations within the study reach where repeated measurements of discharge, width, average depth, and average velocity have been made (source: U.S. Geological Survey and Colorado Division of Water Resources). It was assumed that slopes for each set of channel geometry relationships could be applied to nearby cross sections.

Discharge was measured at 3 gaging stations within the study reach and discharge also was measured for 3 tributaries

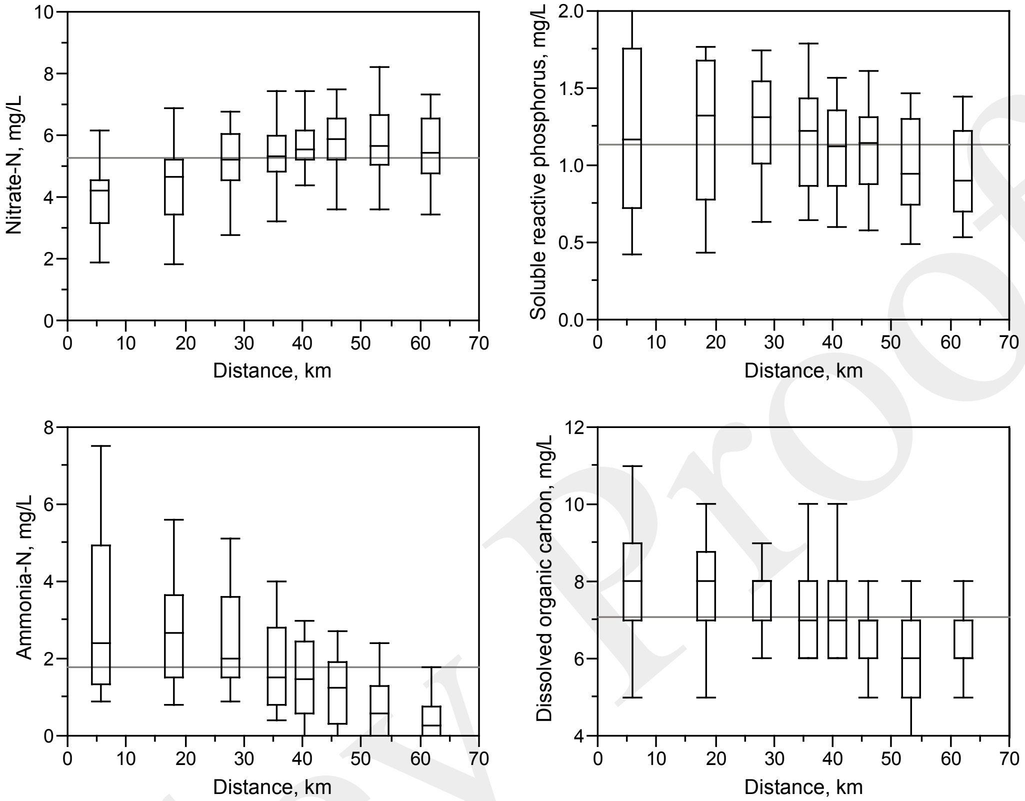

Fig. 2. Concentrations of nitrate-N, ammonia-N, soluble reactive phosphorus, and dissolved organic carbon in the South Platte River during the study period. Boxes show medians, 25th, and 75th percentiles, and whiskers show ranges. Data were provided by the Metro Wastewater Reclamation District, Denver, Colorado.

that contribute to the South Platte main stem within the study reach. Denver's municipal wastewater treatment plant and 3 smaller plants discharge effluent to the river within the study reach, and water is diverted from the river at 10 locations. Daily records of flow for gages, effluent discharges, and diversions were used to construct a daily flow model for the study reach. Groundwater seepage and other ungaged flows were calculated from flow residuals and applied as a distributed source within each segment of the study reach. Thus, on a given day, discharge could be estimated for any point along the study reach, and velocity, channel width, and mean depth were estimated from discharge according to the channel-geometry relationships derived from measurements of channel cross sections.

The reaeration coefficient for oxygen at 20 °C was predicted from channel slope (Cronin et al. 2007); this equation was derived from an analysis of reaeration coefficients within the study reach and is based on empirical measurements from tracer studies with propane conducted by the method of Kilpatrick et al. (1989). Estimates of the reaeration coefficient for oxygen were converted to coefficients for N_2 according to the method of Gulliver et al. (1990) and coefficients were adjusted for temperature as described by Thomann & Mueller (1987).

Temperature and barometric pressure

River temperatures were measured at 1-hour intervals with 4–6 recording digital thermometers. Temperatures at other locations were interpolated from measured values. Barometric pressure was measured with a high-precision barometer on multiple dates at 8 locations within the study reach. The difference in pressure between the sampling locations and Denver International Airport (DIA) varied linearly ($r^2 = 0.998$; $p < 0.0001$) with distance below Denver; this relationship and an hourly record of barometric pressure for DIA (source: National Climatic Data Center) were used to estimate hourly variations in pressure at any point on the South Platte below Denver on each of the sampling dates.

Sampling and estimation of denitrification rates

On each sampling date, river water was collected once at 4 locations spaced evenly across the channel (McCutchan et al. 2003). Samples of alluvial water also were collected with a peristaltic pump from permanent groundwater wells (5 locations) and piezometers (1 location) adjacent to the river channel. Concentrations of dissolved N_2 were measured with a

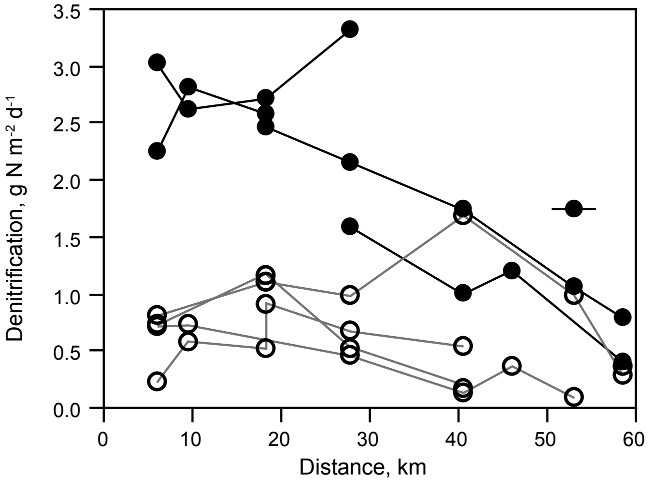

Fig. 3. Spatial and temporal patterns of denitrification rate within the study reach. Solid circles are for the warm months (Jun–Aug) and open circles are for cool months (Oct–Mar).

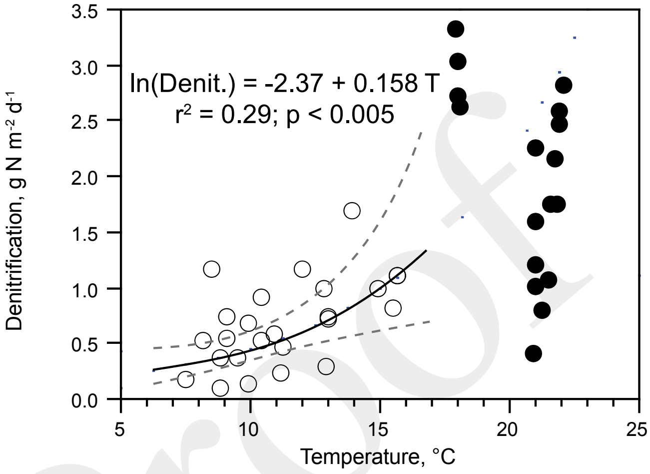

Fig. 4. Effects of temperature on the rate of denitrification. Solid circles are for the warm months (Jun–Aug) and open circles are for cool months (Oct–Mar).

membrane-inlet mass spectrometer (Kana et al. 1994, McCutchan et al. 2003).

For each sampling date, a simulation model was used to predict changes in N_2 concentration at each sampling location over a 24-hour period (Pribyl et al. 2005). These predictions were based on the measured N_2 concentrations, temperature, barometric pressure, channel depth, and the rate of groundwater accrual. It was assumed that the rate of denitrification was constant over the day, and that the N_2 concentration was the same at the beginning and end of each 24-hour modeling period. The rate of denitrification was adjusted in the metabolism model to obtain the best fit with measured concentrations over the 24-hour period.

Results

During the warm months (Jun-Aug), rates of denitrification generally decreased downstream (Fig. 3). During the cool months (Oct-Mar), rates of denitrification showed little spatial pattern. Near Denver, rates were much higher during the warm months, but near the confluence with St. Vrain Creek, rates were similar across the year.

During the cool months there was a weak relationship between temperature and the rate of denitrification, but there was no apparent relationship between temperature and the rate of denitrification during the warm months (Fig. 4). Although rates of denitrification varied considerably across sampling locations during the warm months (Fig. 3), this pattern cannot be attributed to downstream changes in temperature, which were minimal during summer.

Nitrate concentrations were higher during the cool months than during the warm months, but there was not a significant linear relationship between nitrate concentration and the rate of denitrification for the warm months ($r^2 = 0.03$, $p = 0.52$) or for the cool months ($r^2 = 0.06$, $p = 0.25$).

During the warm months, the rate of denitrification increased linearly with the concentration of DOC (Fig. 5). During the cool months, however, the rate of denitrification did not vary significantly with DOC concentration. The lowest rates recorded during the warm months, when the DOC concentration was ~5 mg/L, were similar to the rates of denitrification during the cool months.

Discussion

When water temperatures were above 17 °C, rates of denitrification in the South Platte River were very high near Denver, but declined downstream as the concentration of DOC decreased. During the study period, the nitrate concentration in the river remained high (> 3.5 mg/L), and it is unlikely that the nitrate supply limited rates of denitrification. The relationship between denitrification rate and DOC concentration suggests that labile organic carbon limited rates of denitrification in the South Platte during summer and that much of the DOC in the South Platte was unavailable to denitrifying bacteria (Fig. 5). The source of DOC for denitrifying bacteria, however, remains unclear; the concentration of DOC in the South Platte was highest near Denver's effluent outfall and decreased downstream (Fig. 2), but effluent rich in nitrate does not always support high rates of denitrification be-

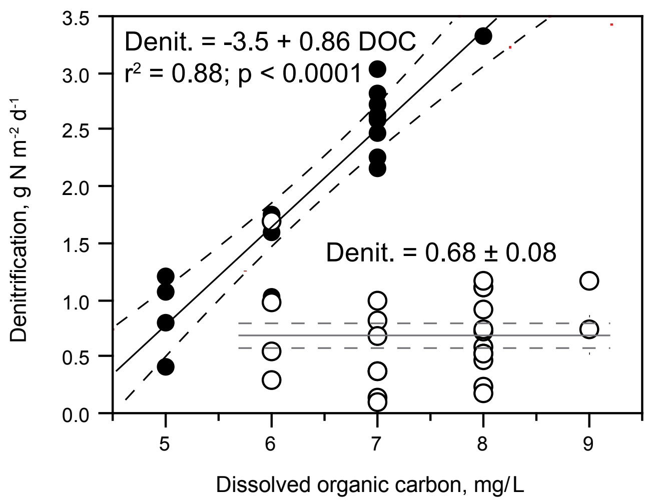

Fig. 5. Effects of dissolved organic carbon on the rate of denitrification. Solid circles are for the warm months (Jun–Aug) and open circles are for cool months (Oct–Mar). DOC data were provided by the Metro Wastewater Reclamation District, Denver, Colorado.

cause the lability of DOC in effluent is variable (Aravena & Robertson 1998). In addition to DOC from effluent, algal production may have been an important source of labile organic carbon for denitrifiers, especially as the labile component of organic carbon derived from wastewater effluent became depleted.

From October through March, when the temperature in the river remained below 17 °C, temperature appeared to be an important control on the rate of denitrification. Numerous studies have demonstrated relationships between temperature and the rate of denitrification (e.g., Pfenning & McMahon 1996, Saunders & Kalff 2001). Low temperatures may regulate metabolic rates for denitrifying bacteria. Rates of denitrification also may be limited indirectly through temperature, which affects the solubility of oxygen and rates of aerobic respiration within the sediments. The combination of increased oxygen solubility and decreased rates of aerobic respiration during winter may limit the volume of the hyporheic zone that has redox conditions favorable to denitrification. Rates of oxygen metabolism in the South Platte River are greatly suppressed during winter (Cronin et al. 2007), but it is not clear whether reduced rates of aerobic respiration in the sediments and increased solubility of oxygen are the main causes of reduced rates of denitrification during winter.

In the South Platte and other rivers with high rates of denitrification, the open-channel N_2 method can estimate rates of denitrification at the reach scale with high precision and with modest effort (McCutchan et al. 2003). Because high precision can be achieved with the open-channel method, it is well suited to the study of subtle variations in rates of denitrification over time and space. Although denitrifying bacteria were first isolated over a century ago, a complete understanding of the factors that control rates of denitrification in river sediments has remained elusive (Davidson & Seitzinger 2006). The open-channel method stands to add considerably to a quantitative understanding of the controlling factors for denitrification in running waters.

Acknowledgements

This work was supported by the Metro Wastewater Reclamation District, Denver, Colorado (MWRD). We thank Jim Dorsch, who organized data provided by MWRD, and cross-section measurements for the upper portion of the study reach. We also thank Steve Lundt, who helped with groundwater sampling, and Laura Tucker and Claire McGrath, who helped with gas sampling and channel surveys.

References

Aravena, R. & W.D. Robertson. 1999. Use of multiple isotope tracers to evaluate denitrification in ground water: study of nitrate from a large-flux septic system plume. Ground Water **36**: 975–982.

Cornwell, J.C., W.M. Kemp & T.M. Kana. 1999. Denitrification in coastal ecosystems: methods, environmental controls, and ecosystem level controls, a review. Aquat. Ecol. **33**: 41–54.

Cronin, G., J.H. McCutchan, Jr., J. Pitlick & W.M. Lewis, Jr. 2007. Use of Shields stress to reconstruct and forecast changes in river metabolism. Freshw. Biol. **52**: 1587–1601.

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

Gulliver, J.S., J.R. Thene & A.J. Rindells. 1990. Indexing gas transfer in self-aerated flows. J. Environ. Eng. **116**: 503–523.

Hill, A.R. 1981. Nitrate-nitrogen flux and utilization in a stream ecosystem during low summer flows. Can. Geogr. **25**: 225–239.

Kana, T.M., C. Darkangelo, M.D. Hunt. J.B. Oldham, G.E. Bennett & J.C. Cornwell. 1994. Membrane inlet mass spectrometer for rapid high-precision determination of N_2, O_2, and Ar in environmental water samples. Anal. Chem. **66**: 4166–4170.

Kilpatrick, F.A., R.E. Rathbun, N. Yotsukura, G.W. Parker & L.L. DeLong. 1989. Determination of stream reaeration coefficients by use of tracers. Techniques of Water-Resources Investigations of the U.S. Geological Survey 3-A18.

Knopf, F.L. & M.L. Scott. 1990. Altered flows and created landscapes in the Platte River headwaters, 1840–1990,

p. 47–70. *In* J.M. Sweeney [ed.], Management of dynamic ecosystems. North Cent. Sect, The Wildlife Society.

Laursen, A.E. & S.P. Seitzinger. 2002. Measurement of denitrification in rivers: an integrated, whole-reach approach. Hydrobiologia **485**: 67–81.

McCutchan, Jr., J.H. & W.M. Lewis, Jr. 2006. Groundwater flux and open-channel estimation of stream metabolism: response to Hall and Tank. Limnol. Oceanogr. **4**: 213–215.

McCutchan, Jr., J.H., J.F. Saunders, III, A.L. Pribyl & W.M. Lewis, Jr. 2003. Open-channel estimation of denitrification. Limnol. Oceanogr. **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.

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. 2002. The estimation of denitrification in a plains river. M.S. thesis, University of Colorado, Boulder.

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

Saunders, D.L. & J. Kalff. 2001. Denitrification rates in the sediments of Lake Memphremagog, Canada-USA. Water Res. **35**: 1879–1904.

Saunders, III, J.F. & W.M. Lewis, Jr. 2003. Implications of climate variability for regulatory low flows in the South Platte Basis, Colorado. J. Am. Water Resour. Assoc. **39**: 33–45.

Sjodin, A.L., W.M. Lewis, Jr. & J.F. Saunders, III. 1997. Denitrification as a component of the nitrogen budget for a large plains river. Biogeochemistry **39**: 327–342.

Thomann, R.V. & J.A. Mueller. 1987. Principles of surface water quality modeling and control. Harper Collins Publishers.

Authors' address: J.H. McCutchan, Jr. (corresp. author), W.M. Lewis, Jr., Center for Limnology, Cooperative Institute for Research in Environmental Sciences, University of Colorado, Boulder, CO 80309-0216, USA.