

Comparison of denitrification characteristics among three habitat types of a large river floodplain: Atchafalaya River Basin, Louisiana

Amy E. Scaroni · John A. Nyman ·
Charles W. Lindau

Published online: 23 September 2010
© Springer Science+Business Media B.V. 2010

Abstract Mobile forms of nitrogen leach from upland environments into aquatic systems, often discharging to coastal zones. Addition of nitrogen to once N-limited systems results in a host of changes ranging from eutrophication to loss of biodiversity. Floodplains can ameliorate these changes by removing and sequestering nitrogen. In many coastal floodplains, sedimentation causes lakes to transition to baldcypress swamps, and ultimately to bottomland hardwood (BLHW) forests. These habitats differ in their contact with floodwater, which directly and indirectly affects their ability to process nutrients, but the effects of habitat change on denitrification at the floodplain scale cannot be predicted because of lack of suitable data. This study compared denitrification characteristics among the aforementioned habitats within the Atchafalaya River Basin (ARB). Microcosms were established in the laboratory, and the

acetylene block technique was used to estimate four denitrification characteristics: background denitrification rates, maximum rates, time to reach maximum rates, and the linear response of denitrification to nitrate concentration. There were significant differences in denitrification characteristics among the three habitat types; specifically, all habitats differed in the time required for denitrification to respond to nitrate in the overlying water, and denitrification in lake habitats differed from both BLHWs and baldcypress swamps. Landscape-scale models should account for different linear relationships between denitrification and nitrate concentrations, and different response times to nitrate concentrations for different habitats. Because denitrification characteristics differ across habitats within the ARB, continued habitat change within the floodplain will alter nutrient discharge to coastal waters.

Guest editors: M. Ford & J.A. Nyman / Interactions among rivers, floodplains, and coastal areas examined at the Atchafalaya River

A. E. Scaroni (✉) · J. A. Nyman
School of Renewable Natural Resources, Louisiana State
University Agricultural Center, Louisiana State
University, Baton Rouge, LA 70803, USA
e-mail: ascaroni@gmail.com

C. W. Lindau
School of the Coast and Environment, Louisiana State
University, Baton Rouge, LA 70803, USA

Keywords Acetylene blockage technique ·
Eutrophication · Mississippi River · Nitrate removal ·
Succession · Wetlands

Introduction

Humans have altered the nitrogen cycle in such a way that levels of reactive nitrogen released into the environment are degrading air, soil, and water quality worldwide (Kulkarni et al., 2008). Nitrogen has been

identified as a key limiting nutrient for biomass production. Consequently, the rate of nitrogen fertilizer application has been on the rise over the past century. While human manipulation of the nitrogen cycle has provided many benefits, a host of unintended problems have resulted. Consequences of increased nitrogen levels in the environment include eutrophication (Rabalais et al., 1996), and loss of ecosystem services (Tilman et al., 2001). Because of the highly mobile nature of nitrate-nitrogen, environmental problems extend beyond the area of application; adverse effects generally are seen downstream of the source. A prime example is the Mississippi River Basin (MRB) in the United States.

The MRB drains over 1.2 million ha of land, which constitutes 40% of the land area of the continental United States (Mitsch et al., 2001). Agriculture is one of the major land uses in this watershed. As a result, nitrogen loading to the MRB has increased substantially in comparison to a pre-industrial baseline (Howarth et al., 1996). This watershed is the primary source of freshwater and nutrients to the Gulf of Mexico (80 and 90%, respectively), and the last 30 years have seen a tripling in the flux of nitrate to the Gulf (Goolsby et al., 2001).

Increased nitrogen loadings to nitrogen-limited marine ecosystems can alter species composition, lower biodiversity, and stimulate biological productivity causing harmful algal blooms and hypoxia (Rabalais et al., 2001). The formation of a large area of hypoxia (oxygen $<2 \text{ mg l}^{-1}$) in the Gulf is primarily attributed to nitrogen loading from the Mississippi and Atchafalaya Rivers (Rabalais et al., 2002). While there is evidence of natural hypoxic events before artificial fertilizers were used in the Upper MRB, hypoxic conditions have intensified since the 1940s, and the extent and persistence of hypoxia are growing (Rabalais et al., 2002). Mitsch et al. (2001) recommend a combination of several best practices for reducing nitrogen loading to the Gulf; these include improvements in fertilizer-use efficiency, upstream wetland creation, and Mississippi River diversions.

Before the Mississippi River reaches the Gulf of Mexico, approximately 30% of the flow diverts through the Old River Control Structure where it joins with the Red River to form the Atchafalaya River. The control structure was built in 1962 to

prevent the Atchafalaya River from capturing the Mississippi River (Sparks, 1992). With an average annual discharge of $6,228 \text{ m}^3 \text{ s}^{-1}$ (Xu, 2006a), the 226-km Atchafalaya River is the fifth largest river in North America by volume. A combination of sedimentation, control structures, and levees ultimately reduce contact between floodwaters and floodplain sediments in the Atchafalaya River Basin (ARB). Once a river- and lake-dominated system, the ARB is now nearly 70% forested (Hupp et al., 2008), and the floodplain continues to fill in with sediment. As surface elevations increase, this will reduce connectivity between the river and sites that currently experience seasonal flooding. The ARB itself is a system in transition; habitats continue to change in response to hydrologic conditions and sediment loads.

The ARB removes 14%, or 50,544 Mg, of the total nitrogen that enters the system via the Atchafalaya River (Xu, 2006a). Processes responsible for removal include storage in the sediments, biomass uptake, and denitrification. The relative contribution of each of these processes is currently unknown for the ARB. Denitrification, the reduction of nitrate to di-nitrogen gas, is the only permanent removal process and is likely the dominant process for removal of river-borne nitrogen (Howarth et al., 1996). Reviews have compared denitrification rates among different wetland types (Groffman, 1994) and across terrestrial, freshwater, and marine systems (Seitzinger et al., 2006). Denitrification characteristics have been estimated in Louisiana freshwater lakes (Lindau et al., 2009), baldcypress swamps (Lindau et al., 2008), and bottomland hardwood (BLHW) forests (Hunter et al., 2008). However, individual studies use different methodologies for measuring denitrification. This makes habitat comparison difficult and limits their use in models to predict the effects of habitat change on denitrification at the floodplain scale. A previous study (Scaroni et al., 2010) identified trends in spatial variability by geographic position across the ARB, with increasing potential denitrification rates along a northwest–southeast trajectory. We hypothesize that this variability can be explained, in part, by habitat type. The objective of this study was to determine whether there are significant differences in denitrification potential among the major habitat types of the ARB using the acetylene block technique. This will contribute to our understanding of the spatial

distribution of denitrification rates in the ARB, and the effects of habitat change on nitrogen removal in the ARB. A secondary aim was to generate estimates of denitrification characteristics from these three habitats that could be used in models of denitrification at the floodplain scale.

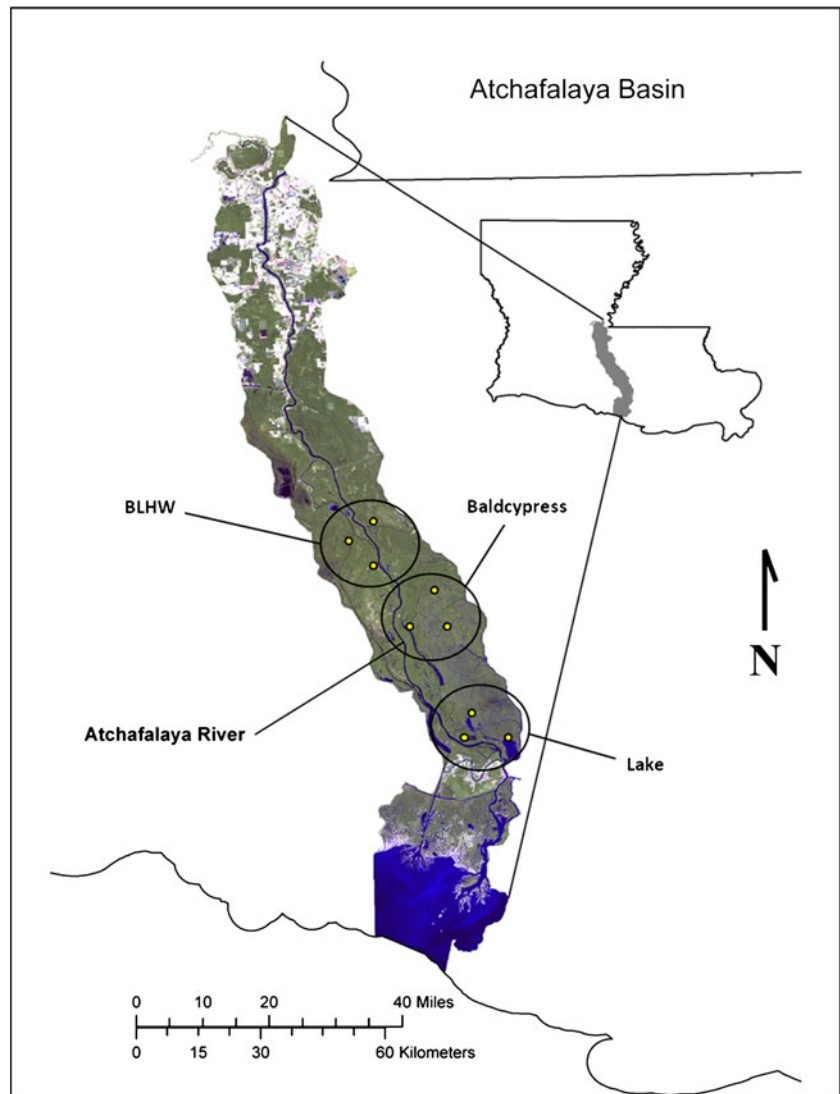
Methods

Sample collection

We identified BLHW, baldcypress swamp, and lake as the three major habitat types in the ARB, and

selected three sites within each habitat type based on accessibility and proximity to public land ($n = 9$) (Fig. 1). Sites within habitat type were considered replicates that were used to estimate variability in denitrification within habitats. It is important to note that there is considerable spatial and temporal variability both within and between habitats in respect to hydrology, and resulting biogeochemical properties of the sediment. The BLHW and baldcypress sites were not flooded during sampling, which reflects the seasonal nature of flooding in these areas. The three lake sites were flooded during sampling, which reflects their typical condition throughout the year. Water monitoring stations located in the main

Fig. 1 Location of sampling sites



channel indicate that the seasonal range in water levels is 9.5 m at the upstream end of the Atchafalaya Basin but only 0.9 m at the downstream end of the basin (Ford and Nyman, this issue). However, soil elevation data are unavailable, and even then water levels in forests and swamps may differ behind spoil banks and natural levees, wherein the majority of our sites were located.

Bulk sediment samples were collected (0–15 cm depth) between June and August 2006. All the samples were sealed in 4-l wide-mouthed heavy-duty polyethylene (HDPE) bottles, put on ice, transported to the laboratory, and stored at 2°C for 1 month. Despite extensive disturbance during extraction and transport of cores, prior study has shown good agreement between in situ and laboratory denitrification studies (Well et al., 2003). Subsamples were removed and analyzed for bulk density. We analyzed nitrogen (N) and carbon (C) content with a Leco C–N Analyzer (Leco Corp., St. Joseph, MI, USA).

Denitrification experiments

Sediments were mixed to homogenize samples. Duplicate microcosms were established for each site by adding approximately 576 g (5 cm) of wet sediment to the bottom of a 9-cm glass incubation jar ($n = 18$). Sediments were compacted to mimic field bulk densities as reported in the literature for these types of soils, and to remove excess gas. Final bulk densities ranged from 0.243 to 0.500 g cm⁻³. Sediments were then flooded with 240 ml (4 cm) of nitrate-free deionized water. Jars were encased in foil and loosely covered with tissue paper to discourage growth of plants. The microcosms equilibrated for approximately 1 week to allow for the development of a thin oxidized layer at the sediment–water interface as was observed in the field. This setup was repeated for each level of nitrate added.

We used the acetylene block technique (Groffman, 1994) to measure background levels of denitrification. Nitrous oxide (N₂O) emissions were measured from the microcosms with no nitrate added (day zero). To simulate low, slightly elevated, and high nitrate loading rates, microcosms were amended with 2 ml of 1, 5, and 50 mg NO₃-N l⁻¹ solution, respectively. Nitrate concentrations in the Atchafalaya River ranged from 0.31 to 2.80 mg N l⁻¹ over the last 25 years (Xu, 2006b).

Acetylene was added directly into the headspace (~10% v/v) and floodwater of each microcosm on each day denitrification was measured, which was capped to prevent gaseous exchange with the atmosphere. Gas samples were collected via a rubber septum in the lid with a 2-ml gas tight syringe at 2 and 4 h (for the 50 mg l⁻¹ addition), or 2 and 6 h (for the 1 and 5 mg l⁻¹ additions). Samples were injected into a Shimadzu GC-14A gas chromatograph (Shimadzu Scientific Instruments, Inc., Columbia, MD, USA) fitted with a 1-ml sampling loop, Porpak Q 1.8-m ss column, and electron capture detector (ECD), and calibrated with certified N₂O gas standards (Scott Specialty Gases, Inc., Plumsteadville, PA, USA). Ultra high-purity nitrogen was the carrier gas, and the instrument was operated at temperatures of 40, 100, and 290°C for the oven, injector, and ECD detector, respectively (Lindau et al., 1998).

Denitrification rates were measured until they began to decrease: 0–9 days (1 mg l⁻¹ NO₃-N), 0–13 days (5 mg l⁻¹ NO₃-N), and 0–29 days (50 mg l⁻¹ NO₃-N) after nitrate additions. This ensured that the system had sufficient time to respond to the nitrate loading and reach maximum rates. Microcosms were sealed only during 4–6 h incubations each measurement day. The highest rate of denitrification over time was defined as the “maximum denitrification rate,” and the time elapsed between nitrate additions and maximum denitrification rate was defined as “time to reach maximum emissions.” All tests were run at 22°C.

Calculations were performed using the Bunsen absorption coefficient (which corrects for solubility of N₂O in water) to determine N₂O–N that was recovered in the headspace and floodwater (Tiedje, 1982). We used the closed chamber equation of Rolston (1986) to calculate final N₂O flux, reported as g N ha⁻¹ day⁻¹.

Statistical analysis

Maximum N₂O emissions and time to reach maximum N₂O emission rates were analyzed to characterize denitrification for each habitat type and each level of nitrate added. Data were analyzed using PROC MIXED in SAS Institute (2006). The experimental design was completely randomized with nesting of site in habitat type to test the hypothesis that denitrification characteristics vary among habitat

types and with nitrate level. We performed pair-wise comparisons with a Tukey adjustment (SAS Institute, 2006). We estimated relationships between denitrification rates and nitrate levels for both initial (24 h following nitrate addition) and maximum emissions using the PROC REG function in SAS Institute (2006). For both regressions, background rates observed were subtracted from initial and maximum rates to force the regression lines through the origin (Nielsen, 1992). Associations between N_2O emissions and soil components (N, C) were tested using the PROC CORR function in SAS Institute (2006).

Results

Background denitrification rates

Background denitrification rates were the highest for the BLHW sites ($5.4 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.72$) followed by baldcypress ($3.9 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.52$), and lake sites ($1.4 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.52$) (Fig. 2). BLHW and lake sites differed ($P = 0.0077$), as did lake and baldcypress sites ($P = 0.0462$). BLHW and baldcypress sites did not differ ($P = 0.2023$).

Potential denitrification rates

The three habitats responded differently to nitrate addition, and the ranking differed from that observed in background denitrification. After addition of $1 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$, denitrification rates were the highest for lakes ($12.0 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.84$), followed by baldcypress ($10.0 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.60$) and BLHW sites ($8.1 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 0.57$) (Fig. 2). These rates differed among each of the habitats ($P = 0.0395$), but only BLHW and lake differed from each other ($P = 0.0143$). When amended with $5 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$, the same pattern was seen as with the 1 mg l^{-1} addition ($P = 0.0002$); however, denitrification rates were higher (lake = $109.4 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 6.81$; baldcypress = $34.1 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 4.43$; BLHW = $23.0 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 1.29$) (Fig. 2). Once again, BLHW and baldcypress did not differ ($P = 0.5247$). With a $50 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$ addition, lake sediments showed the highest rates of denitrification ($451.6 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 64.62$), followed by baldcypress

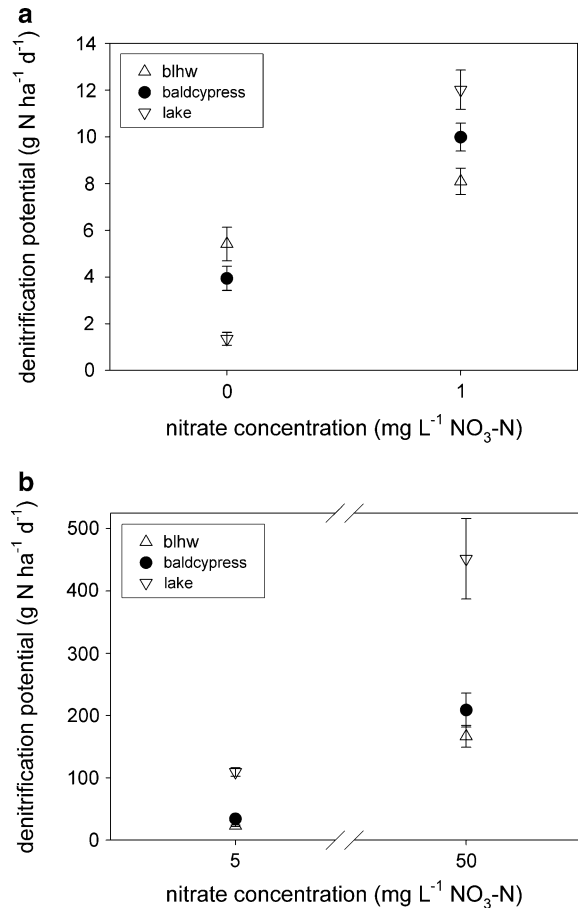


Fig. 2 Denitrification rates for three habitat types in the ARB, Louisiana following **A** 0 and $1 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$ nitrate additions, and **B** 5 and $50 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$ nitrate additions. Graphs show Least Squares Means and Least Squares Standard Error bars

($208.8 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 27.38$), and BLHW ($166.6 \text{ g N ha}^{-1} \text{ day}^{-1}$, $\text{SE} = 17.51$) (Fig. 2). There were significant differences among all comparisons except BLHW and baldcypress sites ($P = 0.8426$).

Response time to nitrate loading

With a $1 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$ addition, denitrification in the lakes peaked in 1 day ($\text{SE} = 0.00$), while baldcypress took 1.6 days ($\text{SE} = 0.42$) and BLHW sites took 3 days ($\text{SE} = 0.00$) (Fig. 3). After addition of $5 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$, lakes peaked in 0.5 days ($\text{SE} = 0.00$), baldcypress took 2.1 days ($\text{SE} = 0.61$), and BLHW took 3.8 days ($\text{SE} = 0.42$) (Fig. 3). After a $50 \text{ mg l}^{-1} \text{ NO}_3^- \text{-N}$ addition, it took 2.2 ($\text{SE} = 0.42$),

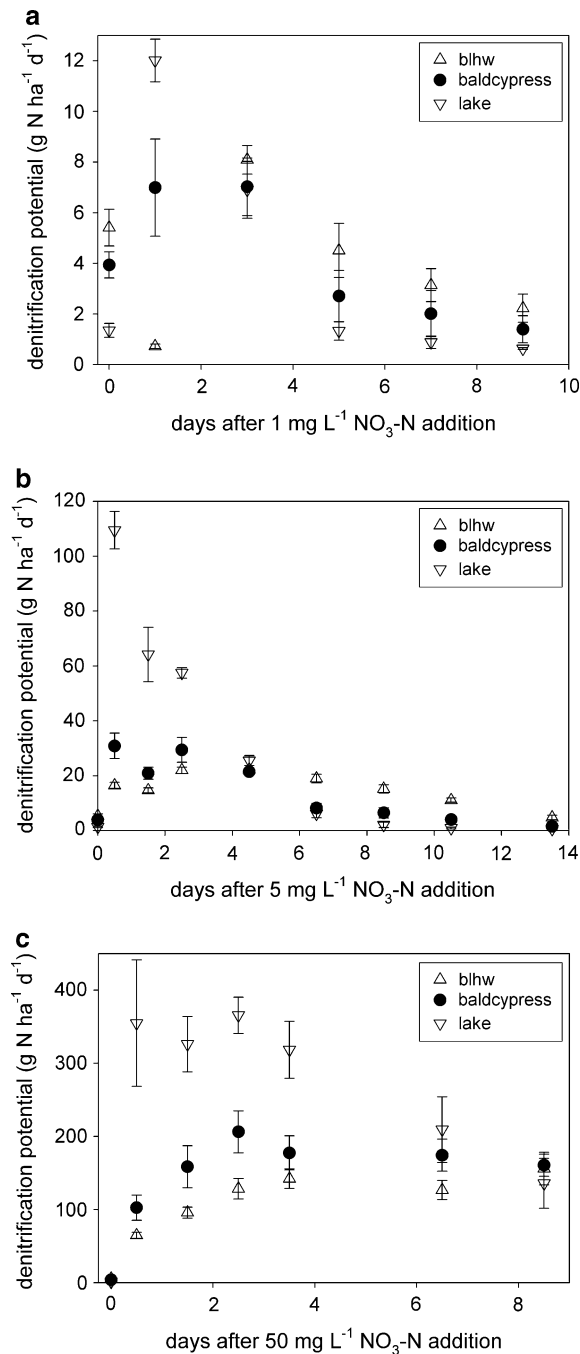


Fig. 3 Temporal dynamics of denitrification potential after additions of **A** 1 mg l⁻¹ NO₃-N nitrate, **B** 5 mg l⁻¹ NO₃-N nitrate and **C** 50 mg l⁻¹ NO₃-N nitrate to microcosms of each habitat type

3.0 (SE = 0.72), and 7.5 days (SE = 1.63) for lakes, baldcypress, and BLHW, respectively, to reach maximum emission rates (Fig. 3). We did not sample

Table 1 Slope of the regression line (g N ha⁻¹ day⁻¹/nitrate ppm) for initial (24 h after nitrate addition) N₂O emissions versus nitrate, and maximum N₂O emissions versus nitrate, by habitat type

	Initial N ₂ O emissions	Max N ₂ O emissions
BLHW	1.2	3.2
Baldcypress	2.0	4.1
Lake	7.2	9.1

Background N₂O emissions were subtracted from both initial and maximum emissions to force the regression line through the origin

multiple times per day, and acknowledge that peak emissions may have occurred between samples.

Relationship between nitrate and denitrification

When we regressed maximum N₂O emissions on nitrate, without accounting for habitat differences, there was a significant linear relationship between nitrate and N₂O, but the *r*-squared value was only 0.73 (slope = 5.49). When we ran this regression separately for each habitat, each of the habitat types proved to have a significant linear relationship between nitrate and maximum N₂O emissions, and more of the variation was explained (BLHW *R*² = 0.94; baldcypress *R*² = 0.91; lake *R*² = 0.89). We then repeated separate regressions for each habitat using initial (24 h after nitrate addition) instead of maximum N₂O emissions. For both regressions, background rates observed were subtracted from initial and maximum rates to force the regression lines through the origin (Nielsen, 1992). These regressions all underestimated the true response of denitrification to nitrate loading (Table 1).

Associations with soil composition

Total C and N content of the sediments differed among habitat types (*P* = 0.0171, *P* = 0.0146, respectively). BLHW sediments contained 1.8% C (SE = 0.36) and 0.18% N (SE = 0.03). Baldcypress swamp sediments contained 6.7% total carbon (SE = 1.8) and 0.66% total nitrogen (SE = 0.16). Lake sediments contained 2.4% total carbon (SE = 0.55) and 0.27% total nitrogen (SE = 0.06). However, neither total carbon nor total nitrogen content was

significantly correlated with denitrification rate ($P = 0.98$, $P = 0.95$, respectively).

Discussion

While denitrification can remove a considerable amount of nitrate from floodwaters, it only occurs under anaerobic conditions. Areas of the floodplain exposed to different flooding regimes thus differ in their anaerobic conditions, and their ability to denitrify. Intermittently wet and dry sediments, such as those found in BLHW forests, respond differently to flooding than sediments that are always submerged, such as those found in lakes (Wijler & Delwiche, 1954). With submerged sediments, flooding will likely bring in additional nutrients and an increased carbon supply. Nitrate does not accumulate, thus increased nitrate levels will stimulate denitrification. These sediments are primed to denitrify, so that nitrate can become limiting to denitrifiers, and background rates may remain low. The rate of N mineralization probably controls the background rate of denitrification in these habitats, as has been shown in other habitats (Seitzinger et al., 2006). With bottomland sediments, dry periods allow for nitrogen mineralization, increased carbon preservation, and nitrate accumulation. This sets the stage for a pulse of denitrification upon rewetting, as previously observed by Groffman (1994) and Baldwin & Mitchell (2000). Our results agree with this scenario; the rewetted BLHW sediments showed the highest rate of background denitrification, while the highly anaerobic lake sediments showed the highest rates of denitrification in response to nitrate additions. Lake sediments also reached their maximum denitrification rates more quickly than the other habitats when nitrate was added. Because anaerobic conditions were limiting denitrification in the field-dry sediments, we likely measured a hot moment (McClain et al., 2003), or reactivation of reactants necessary for denitrification, upon flooding them in the lab.

It is important to note, however, that the BLHW sites in this study rarely receive flooding from the Atchafalaya River. This would decrease the ability to remove inorganic nitrogen flowing in from the Atchafalaya River, and indicates that the background denitrification rate is stimulated by internal nitrogen cycling. These results do suggest that seasonal

diversions through spoil banks and natural levees could increase the capacity of the basin to remove nitrogen as sedimentation continues. BLHW and baldcypress sediments did not differ in their denitrification rates at any of the nitrate levels. Within the Atchafalaya Basin, both of these habitats are exposed to intermittent wet and dry cycles throughout the year, and probably maintain similar redox conditions in the sediment. Ongoing studies are attempting to determine background denitrification rates from unflooded BLHW sediments using unflooded microcosms; this also has implications for N_2O emissions from these areas.

Unlike Seitzinger et al. (1993), who observed no background denitrification from lake sediments when using the acetylene block technique, we observed significant background denitrification in all three habitats. Instead, our results were more similar to those of Groffman & Tiedje (1989), who observed significant background denitrification in forest soils when using this technique. These comparisons are consistent with our observation that background denitrification rates were higher in forest sediments than in lake sediments. Nonetheless, the acetylene block technique probably underestimated denitrification in the three habitats, especially the background estimates when there was no nitrate in the overlying water and only decomposing organic matter to supply nitrate (Groffman et al., 2006). Although acetylene inhibits nitrification, which can mask the importance of denitrification in systems with low external nitrate loading that rely on nitrification as a source of nitrate, this is less of a problem over the short term (Ryden & Dawson, 1982).

Gardner & White (2010) quantified denitrification potential in a Louisiana freshwater marsh receiving diverted Mississippi River water (estimated loading rate of $2 \text{ mg NO}_3^- \text{ l}^{-1}$) and their rates fell in between our denitrification rates in response to our 1 and 5 $\text{mg NO}_3^- \text{ l}^{-1}$ additions. Lindau et al. (2008) recently published denitrification rates (0 and $100 \text{ mg l}^{-1} \text{ NO}_3\text{-N}$ additions) for the same baldcypress swamp sites used in this study. Our background rates were similar to their background rates at 22°C . Our potential rates were similar, although slightly lower, because our highest addition was $50 \text{ mg l}^{-1} \text{ NO}_3\text{-N}$.

According to our results, denitrification increased linearly with nitrate concentrations, agreeing with previous research showing that denitrification in

terrestrial and aquatic ecosystems can be estimated with first-order reaction rates (Boyer et al., 2006). Our results indicate that a linear relationship between denitrification rates and nitrate concentration was fairly strong across habitat types, and even stronger within habitat type.

The time required for denitrification to respond to nitrate loading differed among habitat types. This suggests that future measurements of denitrification potential that include more than one habitat type should avoid measuring at an arbitrary time after adding nitrate. Failure to account for differences in time to reach maximum denitrification rates could inaccurately estimate relationships between denitrification and nitrate concentrations. In this study, BLHW sediments took up to 7 days to reach maximum emissions. This peak would have been undetected in a common 24-h study, and would have misrepresented the differences among habitat types.

A carbon source is a necessary prerequisite for the denitrification process. We detected a difference in total carbon among our habitat types, with baldcypress sites showing the highest C content. However, C and denitrification were not correlated. The lack of a correlation suggests that differences in denitrification among habitat types are not due to C limitation. We did not measure the proportion of C that was labile, and the fairly recalcitrant nature of baldcypress needles could explain why the habitat with the highest C content did not exhibit the highest rates of denitrification. N and C content were correlated with each other, suggesting that much of the N in the soil is bound up in organic matter and is not bioavailable until remineralized by decomposers.

Conclusion

The Atchafalaya River functions as a natural diversion of the Mississippi, and the large floodplain has been shown to remove C, N, and P from floodwaters (Xu, 2006b). Results from this study could be used to guide future planning efforts for diversions. We showed that lake habitats in the Basin differ from forested habitats in their ability to remove nitrate. The transition from lake to baldcypress and BLHW habitat is changing the overall dynamic within the ARB, potentially altering the fate of nutrients flowing toward the Gulf of Mexico. Our results indicate that

denitrification responds to nitrate in the floodwater differently across habitat types, and modeling efforts addressing watersheds containing these habitats should model denitrification separately for each habitat. Models specific to the ARB should use background N₂O emissions for the intercept of a regression line, and maximum N₂O emissions from each habitat type for the slope of a regression line.

Acknowledgements This study was funded by the Louisiana Board of Regents Research Competitiveness Subprogram. We thank D. Heckman, R. Cleary, V. Tobias, J. Conkle, and H. Brand for their assistance in the field and laboratory.

References

- Baldwin, D. S. & A. M. Mitchell, 2000. The effects of drying and re-flooding on the sediment and soil nutrient dynamics of lowland river-floodplain systems: a synthesis. *Regulated Rivers: Research and Management* 16: 457–467.
- Boyer, E. W., R. B. Alexander, W. J. Parton, C. Li, K. Butterbach-Bahl, S. D. Donner, R. W. Skaggs & S. J. Del Grosso, 2006. Modeling denitrification in terrestrial and aquatic ecosystems at regional scales. *Ecological Applications* 16: 2123–2142.
- Gardner, L. M. & J. R. White, 2010. Denitrification enzyme activity as an indicator of nitrate movement through a diversion wetland. *Soil Science Society of America Journal* 74: 1037–1047.
- Goolsby, D. A., W. A. Battaglia, B. T. Aulenbach & R. Hooper, 2001. Nitrogen input to the Gulf of Mexico. *Journal of Environmental Quality* 30: 329–336.
- Groffman, P. M., 1994. Denitrification in freshwater wetlands. *Current Topics in Wetland Biogeochemistry* 1: 15–35.
- Groffman, P. M. & J. M. Tiedje, 1989. Denitrification in north temperate forest soils: spatial and temporal patterns at the landscape and seasonal scales. *Soil Biology and Biochemistry* 21: 613–620.
- 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. Nielsen & M. A. Voytek, 2006. Methods for measuring denitrification: diverse approaches to a difficult problem. *Ecological Applications* 16: 2091–2122.
- Howarth, R. W., G. Billen, D. Swaney, A. Townsend, N. Jaworski, K. Lajtha, J. A. Downing, R. Elmgren, N. Caraco, T. Jordan, F. Berendse, J. Freney, V. Kudeyarov, P. Murdoch & Z. Zhao-Liang, 1996. Regional nitrogen budgets and riverine N, P fluxes for the drainage to the North Atlantic Ocean: natural and human influences. *Biogeochemistry* 35: 75–139.
- Hunter, R. G., S. P. Faulkner & K. A. Gibson, 2008. The importance of hydrology in restoration of bottomland hardwood wetland functions. *Wetlands* 28: 605–615.
- Hupp, C. R., C. R. Demas, D. E. Kroes, R. H. Day & T. W. Doyle, 2008. Recent sedimentation patterns within

- the central Atchafalaya Basin, Louisiana. *Wetlands* 28: 125–140.
- Kulkarni, M. V., P. M. Groffman & J. B. Yavitt, 2008. Solving the global nitrogen problem: it's a gas!. *Frontiers in Ecology and the Environment* 6: 199–206.
- Lindau, C. W., R. D. Delaune, J. W. Collins, P. K. Bollich, L. M. Scott & E. N. Lambremont, 1998. Methane and nitrous oxide evolution and N-15 and Ra-226 uptake as affected by application of gypsum and phosphogypsum to Louisiana rice. *Agriculture Ecosystems and Environment* 68: 165–173.
- Lindau, C. W., R. D. DeLaune, A. E. Scaroni & J. A. Nyman, 2008. Denitrification in cypress swamp within the Atchafalaya River Basin, Louisiana. *Chemosphere* 70: 886–894.
- Lindau, C. W., R. R. Iwai, R. D. DeLaune & R. P. Gambrell, 2009. Denitrification potential of lake sediment at the Davis Pond Mississippi River freshwater diversion site, Louisiana, USA. *Journal of Freshwater Ecology* 24: 181–190.
- McClain, M. E., E. W. Boyer, C. L. Dent, S. E. Gergel, N. B. Grimm, P. M. Groffman, S. C. Hart, J. W. Harvey, C. A. Johnston, E. Mayorga, W. H. McDowell & G. Pinay, 2003. Biogeochemical hot spots and hot moments at the interface of terrestrial and aquatic ecosystems. *Ecosystems* 6: 301–312.
- Mitsch, W. J., J. W. Day, J. W. Gilliam, P. M. Groffman, D. L. Hey, G. W. Randall & N. Wang, 2001. Reducing nitrogen loading to the Gulf of Mexico from the Mississippi River Basin: strategies to counter a persistent ecological problem. *Bioscience* 51: 373–388.
- Nielsen, L. P., 1992. Denitrification in sediment determined from nitrogen isotope pairing. *FEMS Microbiology Ecology* 86: 357–362.
- Rabalais, N. N., R. E. Turner, D. J. Justic, Q. Dortch, W. J. Wiseman & B. K. Sen Gupta, 1996. Nutrient changes in the Mississippi River and system responses on the adjacent continental shelf. *Estuaries* 19: 386–407.
- Rabalais, N. N., R. E. Turner & W. J. Wiseman, 2001. Hypoxia in the Gulf of Mexico. *Journal of Environmental Quality* 30: 320–329.
- Rabalais, N. N., R. E. Turner & W. J. Wiseman, 2002. Hypoxia in the Gulf of Mexico. *Annual Review of Ecology and Systematics* 33: 235–263.
- Rolston, D. E., 1986. Gas Flux. In Klute, A. (ed.), *Methods of Soil Analysis, Part 1, Physical and Mineralogical Methods*. American Society of Agronomy, Madison, WI: 1103–1119.
- Ryden, J. C. & K. P. Dawson, 1982. Evaluation of the acetylene inhibition technique for field measurements of denitrification in grassland soils. *Journal of the Science of Food and Agriculture* 33: 1197–1206.
- SAS Institute, 2006. Version 9.1.3. SAS Institute, Cary, NC.
- Scaroni, A. E., C. W. Lindau & J. A. Nyman, 2010. Spatial Variability of Sediment Denitrification Across the Atchafalaya River Basin, Louisiana, USA. *Wetlands*. doi: 10.1007/s13157-010-009-1.
- Seitzinger, S. P., L. P. Nielsen, J. Caffrey & P. B. Christensen, 1993. Denitrification measurements in aquatic sediments: a comparison of three methods. *Biogeochemistry* 23: 147–167.
- Seitzinger, S. P., J. A. Harrison, J. K. Bohlke, A. F. Bouwman, R. Lowrance, B. Peterson, C. Tobias & G. Van Drecht, 2006. Denitrification across landscapes and waterscapes: a synthesis. *Ecological Applications* 16: 2064–2090.
- Sparks, R. E., 1992. The Atchafalaya Basin. In Maurizi, S. & F. Poillon (eds), *Restoration of Aquatic Ecosystems*. National Academy Press, Washington, DC: 398–405.
- Tiedje, J. M., 1982. Denitrification. In Page, A. L., R. H. Miller & D. R. Keeney (eds), *Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties*, Agronomy Monograph No. 9, 2nd ed. American Society of Agronomy, Madison, Wisconsin, USA: 1011–1026.
- Tilman, D., J. Fargione, B. Wolff, C. D'Antonio, A. Dobson, R. Howarth, D. Schindler, W. Schlesinger, D. Simberloff & D. Swackhamer, 2001. Forecasting agriculturally driven global environmental change. *Science* 292: 281–284.
- Well, R., J. Augustin, K. Meyer & D. D. Myrold, 2003. Comparison of field and laboratory measurement of denitrification and N₂O production in the saturated zone of hydromorphic soils. *Soil Biology and Biogeochemistry* 35: 783–799.
- Wijler, J. & C. C. Delwiche, 1954. Investigations on the denitrifying process in soil. *Plant and Soil* 5: 155–169.
- Xu, Y. J., 2006a. Total nitrogen inflow and outflow from a Large River Swamp Basin to the Gulf of Mexico. *Hydrological Sciences Journal* 51: 531–542.
- Xu, Y. J., 2006b. Organic nitrogen retention in the Atchafalaya River Swamp. *Hydrobiologia* 560: 133–143.