USGS

# WATER RESOURCES RESEARCH GRANT PROPOSAL

**Title:** Denitrification Rates and Controls in Sediments of Illinois Surface Waters

**Focus Categories:** CL, SED, NPP

**Keywords:** Agriculture, Denitrification, Nitrogen, Nutrients, Rivers, Sediments, Streams, Water Quality

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**FY 1999 NON-FEDERAL MATCHING FUNDS:** $48,091

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**Congressional District:** 15th

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## Statement of Critical Regional or State Water Problems

Recent estimates have shown that agriculture dominates the nitrogen (N) budget of Illinois, with large inputs (fertilization and N fixation, Fig. 1) and outputs (grain harvest,

Fig. 2) in the state (M. David, unpublished data). The budget is unbalanced, however, with a large surplus of N estimated each year (about 650,000 Mg N), primarily due to larger inputs of N compared to crop uptake and export. Some of this surplus N is transported to rivers and exported from the state (~200,000 Mg N $y^{-1}$ for 1994 to 1996), ending up in the Mississippi River. Studies in Illinois have clearly linked agricultural practices, subsurface tile drainage, and river nitrate concentrations (David et al., 1997; Gentry et al., 1998). Many Illinois surface waters, which are often used as drinking water supplies, have nitrate concentrations greater than the EPA standard of 10 mg N $L^{-1}$. In 1996, the difference between N inputs and outputs was estimated at 650,000 Mg N, and approximately 188,000 Mg of this N was exported from Illinois by the major river systems (Fig. 2). Yet, a large amount of N (461,000 Mg N) was still unaccounted for after subtracting the export by rivers, and determining its fate is critical to better understand what is happening to this large N surplus. Due to concern about the hypoxic zone in the Gulf of Mexico and possible linkages to N in the Mississippi River, along with drinking water problems, we need to fully understand the N budget of Illinois and controls on river N concentrations and export.

Currently, N losses from agricultural fields in much of the state are greater than N exports estimated at the mouths of the six major river systems (all expressed on a per ha basis), thereby linking in-stream processing and loss of N. River estimates of N export are made where the rivers enter the Mississippi or Ohio Rivers, and may not reflect all inputs made along long flow paths. Denitrification by microbes in surface waters could account for much of the fate of this missing N, implying larger losses from agricultural fields than would be estimated solely by river N fluxes. Therefore, denitrification could be a major process in reducing the export of N from Illinois rivers, and of critical importance. Estimates of in-stream denitrification rates would help resolve the linkage between agricultural losses of N to surface water export from the state. We also recognize, however, that a large amount of the surplus N estimated each year in Illinois may be lost through field denitrification, and never reach surface waters. By providing initial estimates of denitrification rates in Illinois surface waters in this study, we will help to put bounds on possible field loss rates of N as well.

**Statement of Results or Benefits**

We will use our measurements of denitrification rates to reevaluate N loss from agricultural systems and export in rivers using data from a range of sources. As described previously, N budgets have been and are being made for Illinois agricultural watersheds. Surface water denitrification rates will allow imbalances in budgets to be resolved, and help reduce uncertainty in the fate of N. These data are needed to fully understand the magnitude of N loss from agricultural fields, so that changes can be made to reduce the inputs. In addition, we will know the controls on denitrification rates, which will be needed to better estimate how future N inputs to surface waters may be reduced by this process.

This type of information is critically needed by state and federal agencies that are now considering surface water nutrient criteria and standards, and perhaps management

regulations to achieve them. The US EPA, through the President's Clean Water Action Plan of March 24, 1998, is mandated to establish nutrient criteria that reflect different water bodies in the US and to assist States and Tribes in adopting water quality standards based on these criteria (US EPA, 1998). To effectively determine nutrient criteria and standards in agricultural midwestern streams and rivers, we need to know in-stream processing rates of N. Denitirification could be a major factor in reducing nitrate concentrations in surface waters, but few data are available on rates and process controls. Our study will begin to provide this type of data and understanding.

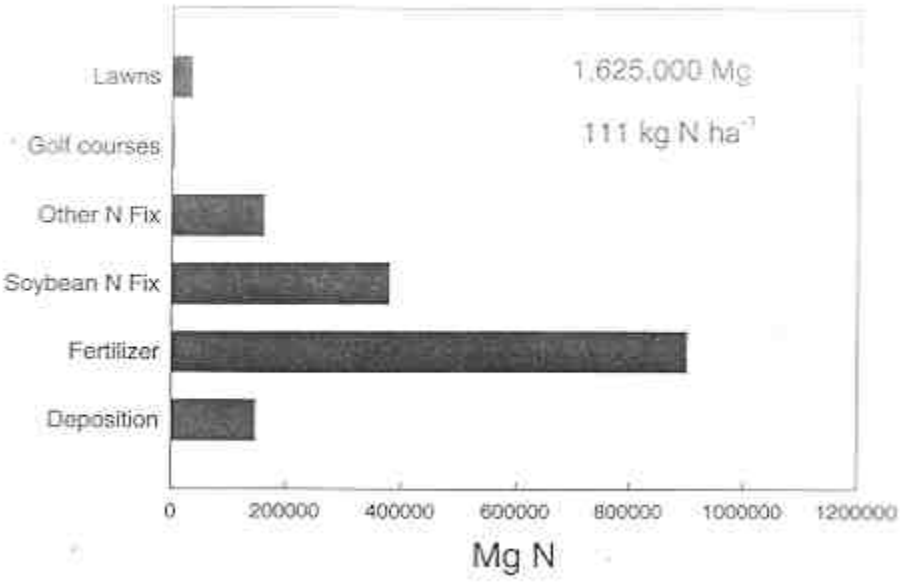


**Fig. 1.** Estimated human-derived inputs of N to Illinois in 1996. A total of 1,625,000 Mg N was added to the state, or 111 kg N $ha^{-1}$, mostly from fertilizer and N fixation.

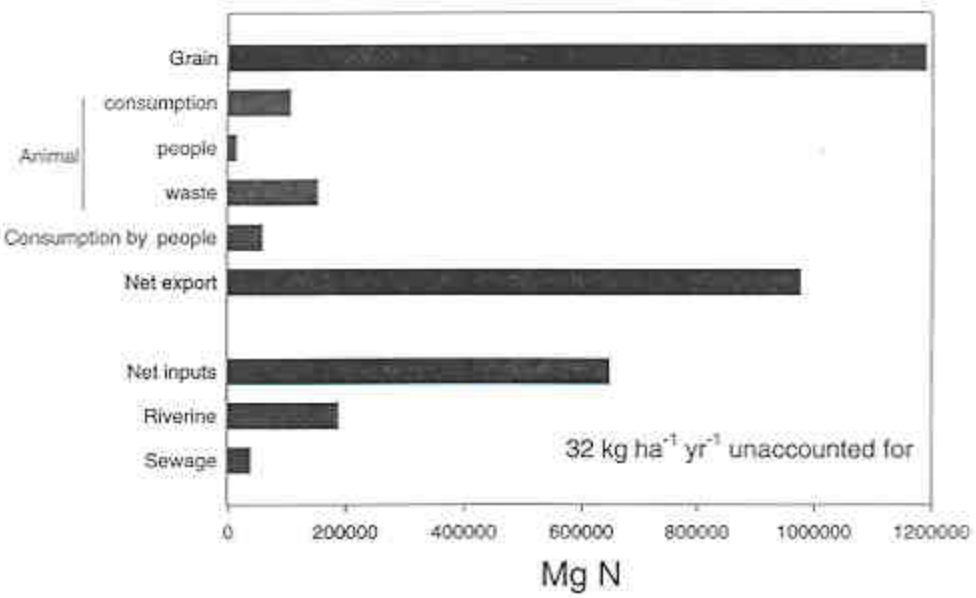


**Fig. 2.** Estimated outputs (by grain export) and conversions of N (consumption by animals and people), which are subtracted from total inputs (Fig. 1) to give a net input. Also shown are riverine N flux (estimated for all Illinois rivers) and a maximum sewage contribution. In this budget 32 kg N $ha^{-1}$ $yr^{-1}$ were unaccounted for.

**Nature, Scope and Objectives of Research**

We will establish denitrification rates and controls in sediments in a range of Illinois surface waters that currently transport large amounts of N, primarily as nitrate. During the first year we will use acetylene inhibition techniques to determine denitrification rates on sediment samples collected from 40-50 sites in Illinois streams, rivers, and reservoirs. Additional chemical and physical data will be collected to better understand the controls on denitrification rates. In the second year of the study, we will focus on a subset of sites using the acetylene inhibition technique combined with $^{15}N$ field additions to confirm denitrification rate estimates.
Our objectives are to:

1. measure denitrification rates in sediments sampled from a range of Illinois surface waters, determining regulating factors and identifying representative sites for more detailed studies; and

2. conduct detailed studies to determine denitrification rates seasonally at selected representative sites and make estimates of N loss through denitrification, comparing the loss to estimated inputs and river export.

**Methods, Procedures and Facilities**

The first year of funding will involve a screening of denitrification potential at 40-50 sites in Illinois streams, rivers, and reservoirs. We will make denitrification estimates of river sediments using the acetylene inhibition of $N_2O$ reductase method (Balderston et al., 1976; Yoshinari et al., 1976; Smith et al., 1978; Limmer & Steele, 1982). Sediment cores (0 –5 cm in depth) will be collected during spring to early summer (when most river export of nitrate occurs) from representative river habitats at each site (e.g., run, riffle, backwater pool areas) and brought back to the laboratory. The denitrification assay will follow the modified procedure of García-Ruiz et al. (1998). Samples will be incubated at the field temperature of the sediment. The amount of $N_2O$ will be quantified by gas chromatography using an electron-capture detector. We recognize certain limitations of the acetylene inhibition technique (e.g., Seitzinger et al., 1993), but feel it will provide the critical preliminary data needed to better understand denitrification rates in a wide range of surface waters and support later work. Additionally, samples from the water column just above each sediment sample will be collected and analyzed for nitrate (ion chromatograph (IC)), ammonium (colorimetric, auto-analyzer), phosphate (colorimetric, manual), sulfate (IC), and DOC (persulfate oxidation and UV detection, Dohrmann DC-80) using standard methods (APHA, 1995). Particle size (hydrometer), organic C (combustion, trapping and LECO C analyzer), total N (Kjeldahl digestion, ammonium by auto-analyzer), and extractable nitrate and ammonium (KCl extraction, colorimetric using auto-analyzer) will be determined on each sediment sample (Klute, 1986; Sparks, 1996).

In addition to the 1st year screening study, we will have replicate sites at several locations and habitat types on the Embarras River, which we have studied intensively since 1992. This will allow us to assess spatial variability in denitrification rates in a single surface water system. However, we predict that our water column and sediment measurements will explain much of the variation in denitrification rates across our wide range of sites.

During the second year we will focus efforts on a subset of sites to determine seasonal variability in denitrification rates throughout the year using the acetylene inhibition method in the laboratory as well as $^{15}N$ field additions to confirm rate estimates under realistic field conditions. We have recently used both of these techniques successfully in wetland studies of denitrification rates (Xue et al., 1999), and believe they will provide the data needed here. In contrast to acetylene inhibition methods conducted in the lab, $^{15}N$ additions will be made in field mesocosms, which will allow *in situ* transformations of nitrate to be made. The $^{15}N$ technique involves sealing off a column of water and its associated benthic sediments using a capped section of PVC pipe placed in the stream and driven up to 20 cm into the sediments. A known amount of 99% pure $^{15}N$ is then added as $Ca(^{15}NO_3)_2$ to serve as the tracer while not increasing the ambient nitrate concentration in the water. Gas samples are taken from the head-space of the sealed pipe over time and $^{15}N$ content determined using a mass spectrometer. Denitrification rates are calculated as amount of $N_20$ and $N_2$ emitted divided by total duration of experiment and cross sectional area of PVC pipe. Bi-monthly estimates of denitrification rates at multiple habitat types (pool, run, riffle) will be determined using both the field and laboratory

techniques to determine spatial and temporal variability in denitrification rates in surface waters.

As a first approach to convert areal denitrification rates in rivers to mass loss we will use the model of Kelly et al. (1987) as applied by Howarth et al. (1996). This model uses water residence time, mean depth, and an average mass transfer coefficient that describes the height of the water column where nitrate is removed by denitrification per unit time. We will be able to determine all of the variables needed to apply this model, allowing us to convert our denitrification measurements to a mass loss at each site.

Inputs of N to each watershed will be estimated (using IL Dept. of Agriculture statistics and atmospheric deposition rates) and compared to the river flux of N (determined using daily flow rates from USGS gaging stations and ILEPA chemistry data supplemented, where possible, with our own N measurements) and other outputs (harvest export of N), methods we have previously used for the Embarras River watershed at Camargo (David et al., 1997). Our estimates of denitrification will then be added to these budgets, which should begin to close the gap between inputs and outputs.

**Facilities Available**

The Department of Natural Resources and Environmental Sciences at the University of Illinois has the facilities and equipment needed to carry out this project. The PI's have well equipped environmental chemistry laboratories for much of the analyses needed (surface water and sediment chemistry), and have access to other equipment that may be needed (gas chromatograph). We have worked with Dr. Richard Mulvaney in the past for $^{15}N$ measurements, and will in this project. He has a customized mass spectrometer that we have previously used for $^{15}N$ gas measurements (Xue et al., 1999). Available equipment includes:

Dionex ion chromatograph
Dohrmann DC-80 C analyzer
Technicon auto-analyzer
Perkin-Elmer atomic absorption spectrophotometer
Perkin-Elmer dual beam UV/Vis spectrophotometer
Refrigerated incubators for controlled temperature studies
LECO total C analyzer
Hewlett Packard gas chromatograph
Aluminum block digester
Laboratory pH and conductivity meters
Field pH, oxygen and temperature meters
Complete wet chemistry analytical facilities
Pentium laboratory and office computers
Laptop computers for field work

**Related Research**

Using nutrient data from river systems around the world, Howarth et al. (1996) found a positive relationship between anthropogenic N inputs and riverine N flux (Fig. 3). If Illinois river export is placed on this figure (estimated for the entire state), there is a higher N flux per unit area compared to other river systems with similar N inputs (M. David, unpublished data). When a small Illinois watershed is added (Camargo, Illinois), a large export of N is shown. The role of in-stream processes in determining N flux is unknown, however, but may in part explain the decrease in river export per unit area from small watersheds to the mouth of large rivers.

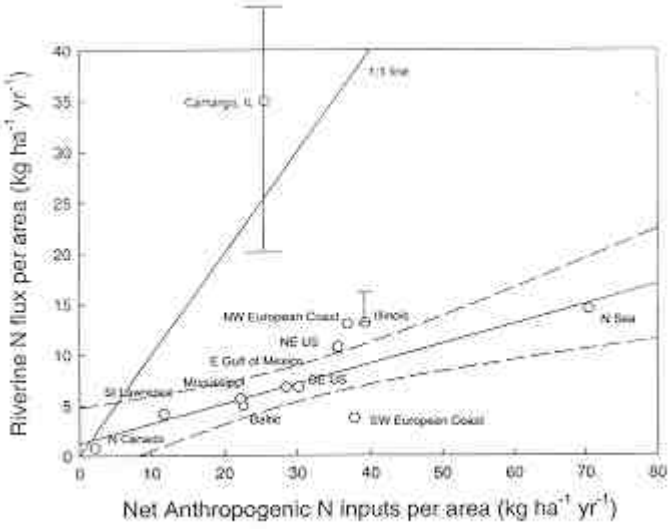


**Fig. 3.** River N flux versus net human-derived N inputs to each of the regions with surface waters draining to the North Atlantic. Net inputs are equal to sum of anthropogenic NOy deposition, fertilizer inputs, N fixation by crops, and the net import or export of N in food and feed. Illinois inputs and river flux (with range for 1994-1996) is shown, along with the Embarras River at Camargo, Illinois (river flux from 1992 through 1997). Adapted from Howarth et al. (1996).

Howarth et al. (1996), in summarizing N processing in large rivers as part of a regional study, suggested that as much as 50% of large river N is lost to in-stream processes, but typically might be 10-20% of total N inputs. They indicated that in well oxygenated surface waters (most of the North Atlantic rivers) benthic denitrification is the major process that removes N. However, most of these estimates are based on mass balances, and there have been few direct measurements of denitrification rates. For streams and rivers of the Midwest, these types of measurements are needed to help resolve imbalances in current N budgets as discussed previously.

Few studies have focused on in-stream denitrification. Recently, García-Ruiz et al. (1998) determined denitrification rates at 50 sites in northeast England. Denitrification was detected at all sites yet they found a wide range in denitrification rates, with water nitrate and sediment C and N concentration explaining much of the variation. Denitrification

rates were also correlated with increasing river size where there were greater rates of sedimentation resulting from higher current velocities, as well as increased nutrient concentrations in the water column (Garcia-Ruiz et al., 1998). Howarth et al. (1996) summarized 11 studies that measured denitirification rates in rivers. However, all had concentrations of less than 10 mg N $L^{-1}$ as nitrate, with most less than 5. There is a need for denitrification measurements in streams of the Midwest with high nitrate concentrations, since denitrification rates are generally considered to follow first order kinetics.

Detailed studies of the factors controlling denitrification have been conducted in terrestrial systems (both agricultural and forest habitats). Increasing water contents in soils tends to inhibit oxygen diffusion, creating a favorable environment for denitrification (Weier et al., 1993). Finer textured soils were found to have greater water retention leading to anaerobic conditions and denitrification (Groffman and Tiedje, 1989). In contrast, in aquatic systems, the water content of sediments does not vary as all sediments are saturated. Factors controlling oxygen concentrations, and N and C availability, will likely control denitrification rates but further study is needed to confirm these predictions.

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