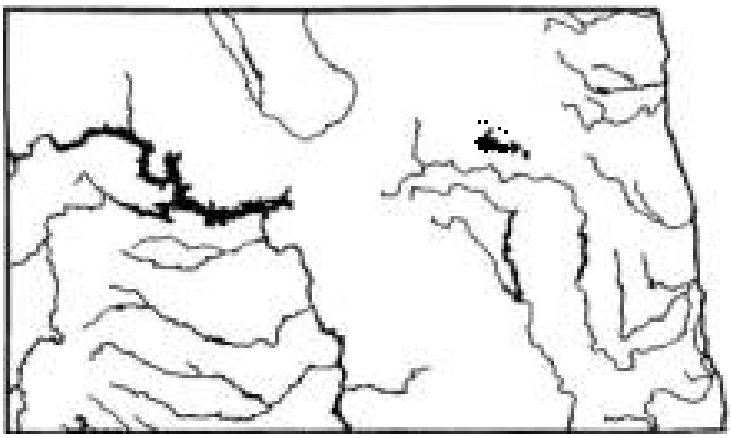

North Dakota Water Resources Research Institute

North Dakota State University
University of North Dakota

ANNUAL REPORT

For the Period March 2006 to February 2007

Fiscal Year 2006 Report to the U.S. Geological Survey

June 2007

Annual Report

Fiscal Year 2006 Report to the U.S. Geological Survey

Award No. 06-HQ-GR-0104
U.S. Geological Survey

G. Padmanabhan, Director
North Dakota Water Resources Research Institute
North Dakota State University
Fargo, ND 58105

June 2007

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INTRODUCTION

This report describes the activities of the North Dakota Water Resources Research Institute (NDWRRI) during the period of March 1, 2006 to February 28, 2007.

The ND WRRI is one of the 54 institutes known collectedly as the National Institutes for Water Resources (NIWR). The NDWRRI was founded in 1965, by authority of Congress (Water Resources Research Acts of 1964, 1972, 1984, and 1990), and is administrated through the United States Geological Survey. Section 104 of the Water Resources Research Act requires the NDWRRI to apply its Federal allotment funds to:

1. Plan, conduct or otherwise arrange for competent research that fosters: (A) the entry of new research scientists into the water resources field, (B) training and education of future water resources scientists, engineers, and technicians; (C) the preliminary exploration of new ideas that address water problems or expand understanding of water and water-related phenomena; and (D) the dissemination of research results to water managers and public.

2. Cooperate closely with other college and universities in the state that have demonstrated the capability for research, information dissemination and graduate training, in order to develop a statewide program designed to resolve State and regional water and related land problems.

3. Cooperate closely with other institutes and other organizations in the region to increase the effectiveness of the Institute and for the purpose of promoting regional cooperation.

This year, NDWRRI once again allocated its 104(B) resources to fund the Graduate Fellowship research projects. The institute also continued its efforts to enhance communications between the State and Federal agency personnel and university faculty and students. Three proposals were submitted by the faculty for the National Competitive 104(G) grant program. One of them was funded. NDWRRI also worked closely with the Environmental and Conservation Sciences program of North Dakota State University (NDSU), Energy and Environmental Research Center at University of North Dakota (UND), and the International Water Institute, Fargo, ND on water related research issues and collaboration. NDWRRI co-sponsored an international conference on water, March 13-15, 2007 with the International Water Institute, Fargo, North Dakota. The Director organized and chaired two sessions creating a forum for presentation of the WRRI Fellowship research to the public. Several WRRI Fellows and their advisors participated.

Program Management

The Institute continued the same administrative mechanism with a director managing the institute program with the help of a State Advisory Committee. Dr. G. Padmanabhan, Professor of Civil Engineering, is the director. Linda Charlton, a NDSU employee, has been working part-time for the Institute to assist the director with Institute finances, communications and information transfer. The State Advisory Committee consists of three members representing the three principal water agencies in North Dakota: State Water Commission, State Department of Health, and the USGS North Dakota District. In addition, the Institute also has a Technical Advisory Committee consisting of faculty from two universities, North Dakota State University and University of North Dakota.

State Appropriation

The State Water Commission continued again this year its support to the 2006 – 2007 federal 104(B) funding for the Graduate Research Fellowship program of NDWRRI. This is third year the SWC provided 15% match in support of the Fellowship program.

University Support

North Dakota State University and the University of North Dakota administrations consider the NDWRRI activities important and are supportive of its efforts.

Institute Location

The Institute continues to operate from the Administrative Building of the College of Engineering and Architecture of North Dakota State University in Fargo, North Dakota, The director may be reached at:

ND Water Resources Research Institute
North Dakota State University
Department of Civil Engineering
Fargo, ND 58105
Phone: (701) 231-7043
Fax: (701) 231-6185
E-mail: G.Padmanabhan@ndsu.edu

State Advisory Committee

The State Advisory Committee provided guidance on water resources research priorities in the State and region, and participated in the review and evaluation of research proposals and projects. The current committee members are:

Gregg Wiche, District Chief, U.S. Geological Survey, Water Resources Division, Bismarck, North Dakota

William Schuh, Water Appropriation Division, North Dakota State Water Commission, Bismarck North Dakota

Mike T. Sauer, Environmental Health Section, North Dakota Department of Health, Bismarck, North Dakota

The committee members are senior officials in the three agencies in North Dakota responsible for much of the water resources research done outside of NDSU and UND in North Dakota.

RESEARCH PROGRAM

ANNUAL BASE GRANT (104-B)

In the past several years NDWRRI has offered competitive fellowships to NDSU and UND graduate students for research on water resources topics under a Graduate Research Fellowship (GRF) program effectively using the modest amount of the 104(B) annual base grant. The program meets the requirements of Section 104 of the Water Resources Research Act of 1984. The fellowship program encourages entry of young university faculty and new research scientists into the water resources field; provides training and education to future water resource scientists and engineers; promotes exploration of new ideas that address water problems or expand understanding of water quantity, quality and related phenomena; and engages university faculty in collaborative research programs seeking supports from entities concerned with water problems.

This year, the NDWRRI continued the GRF program and applied bulk of the federal allotment to it. The GRF program is administrated and monitored by the director. Applications are invited from the graduate students and their advisors of the two research universities of the State, NDSU and UND. A rigorous review by the State Advisory Committee and other water professionals in the state determines the awards. Active participation of the academic advisors of the students in meeting matching requirement and seeking co-funding from local, state and other sources is another positive aspect of the program. Periodical review of the progress of the students in meeting the fellowship expectations is ensured by seeking reports from the students and by encouraging them to make presentations in local and regional technical seminars and conferences.

Guidelines for the 2006-2007 Graduate Research Fellowship competition were posted on the Institute website in September 2005, and the competition was announced in the faculty news publications of the two university campuses in October. The following is the request for application that was published on the UND and NDSU campus newsletters, and distributed by e-mail lists:

ND WRRI Calls for Applications for Graduate Research Fellowships

The ND Water Resources Research Institute announces its 2006 Graduate Research Fellowship program. NDSU and UND Graduate students who are conducting or planning research in water resources areas may apply for fellowships varying from three summer months to a full year in duration. Stipends may range from $800-$1,400 per month. The fellowship funds must be applied between March 2006 and February 2007.

Projects proposed for fellowship support should relate to water resources research issues in the state or region. Regional, state, or local collaborations or co-funding will strengthen an application. Fellowships have a matching requirement of two non-federal dollars to one federal dollar. Applicants should have a plan of study filed and should have a thesis research topic chosen at the time of applying. Applications need to be prepared in consultation with advisors. Advisors of the applicant should co-sign the applications.

Applications are due in the office of the director by 5:00 p.m., November 18, 2005. The proposals will be reviewed by a panel of faculty and state water resources research professionals. Announcement of awards will be made by early January.

Consult the ND WRRI website, http://www.ndsu.edu/wrri, for background on the program, and guidelines for preparation of applications. Applicants and advisors who are new to the program are urged to contact ND WRRI Director, G. Padmanabhan, at 231-7043, or G.Padmanabhan@ndsu.nodak.edu.

Send your applications to Dr. G. Padmanabhan, Director, ND Water Resources Research Institute, North Dakota State University, CIE 201E, Department of Civil Engineering and Construction, P.O. Box 5285, Fargo, ND 58105.

NDWRRI GRADUATE RESEARCH FELLOWSHIPS

Twelve fellowships were awarded in FY2006. The titles of the fellowship projects awarded are given below and details are provided for each project under separate project sections. Six of the Fellowships are renewals, three M.S. and three Ph.D. The renewals are Ali Tackett, Kendall Goltz, William Lenarz, Dan McEwen, Michael Newbrey, and Tedros Tesfay.

2006-07 Fellows and their projects:

Graduate Student	Project Title	Advisor
Ali Tackett, M.S. Biological Sciences, NDSU	Molecular phylogeography of Etheostoma nigrum (Rafinesque) in the upper Midwest.	Dr. Craig Stockwell, Assistant Professor, Biological Sciences, NDSU
Ara Anderson*, M.S. Biological Sciences, NDSU	The life history of Hexagenia limbata (Serville) (Ephemeroptera:Ephemeridae) in North Dakota and Minnesota streams	Malcolm Butler, Professor of Zoology Department of Biological Sciences, NDSU
Christopher Hill, M.S. Civil Engineering, NDSU	Using Entrapped Cell Systems for Treating Supernatant from Anaerobic Digester of the Moorhead Wastewater Treatment Plant	Dr. Eakalak Khan, Assistant Professor, Civil Engineering, NDSU
Dan McEwen, Ph.D Biological Sciences, NDSU	Stoichiometry and the transfer of mercury from benthic macroinvertebrates into game fish	Malcolm Butler, Professor of Zoology Department of Biological Sciences, NDSU
Kendall Goltz, M.S. Natural Resources Management, NDSU	The Impact of Wetlands and Wetland Easements on North Dakota Land Values	Dr. Jay Leitch, Professor of Agricultural Economics, NDSU
Mary Schuh, M.S. Soil Science, NDSU	Farm-scale reconnaissance of estrogens in subsurface waters	Dr. Frank Casey, Assistant Professor of Soil Physics, NDSU
Michael Newbrey, Ph.D Biology, NDSU	Comparative Study of fossil and extant fish growth: Including analyses of Mean annual temperature in the geologic record	Dr. Allan Ashworth, Professor, Geology, NDSU
Ryan Klapperich, M.S. Geology , UND	Analysis of Associated Bedrock-Aquifer System Sediments: Origins of Electron Donor-Rich Aquifers in Eastern North Dakota	Dr. Scott Korom, Professor, Geology and Geological Engineering, UND
Tedros Tesfay, Ph.D. Geology, UND	Modeling Groundwater Dentrification by Ferrous Iron using PHREEQC	Dr. Scott Korom, Professor, Geology and Geological Engineering, UND
Wei Zheng, Ph.D. Biological Sciences, NDSU	Classification of Macroinvertebrate Communities across Red River Drainage Basin	Malcolm Butler, Professor of Zoology Department of Biological Sciences, NDSU
William Lenarz, M.S. Geology, UND	Effect of flow path processes on the geochemistry and quality of water discharged along the seepage face at Pigeon Point, Sheyenne delta aquifer, Ransom County, North Dakota	Dr. Phil Gerla, Geology and Geological Engineering, UND
Yuhui Jin, Ph.D. Chemistry, UND	Rapid and Sensitive Determination of Bacteria in Water Using Nanoparticles	Julia Zhao, Assistant Professor, Department of Chemistry, UND

* Ara Anderson had to leave the graduate program due to unavoidable circumstances before much progress was made and duly informed us so. The amount was re-awarded to another deserving NDSU graduate student of Dr. Malcolm Butler:
Name: Melissa Konsti
Major: MS Student in Zoology
Title: Top-down and bottom-up effects on the abundance of periphyton in shallow lakes

NATIONAL COMPETITIVE PROGRAM (104-G)

The Director, G. Padmanabhan, again this year, encouraged several faculty members from NDSU and UND to submit good proposals for the NIWR-USGS National Competitive Grant program (104-G). Three proposals (one from NDSU and two from UND) were submitted through the ND Institute. The proposal “Collaborative Research on In Situ Denitrification and Glyphosate Transformation in Ground Water: NAWQA Eastern Iowa Basins Study Unit” of Dr. Scott Korom, UND was funded for three years for a total of $91,988.

INFORMATION DISSEMINATION

Information dissemination is done through an annual newsletter initiated in 1992, a website initiated in 1999, and presentations and publications by grant and fellowship recipients. The institute's website address is http://www.ndsu.edu/wrri. The newsletter is usually issued in the month of December of each year. Past newsletters can be accessed through the institute web site. Technical reports of Fellowship projects authored by the Fellows and advisers are also placed on the institute web site.

NDWRRI continued its sponsorship of the Biotic Resources Seminar Series at North Dakota State University.

ND WRRI and Civil Engineering Department of North Dakota State University cosponsored a seminar on “Dam-Break Flood in Natural Channels” by Dr. Arup K. Sarma, Associate Professor of Civil Engineering, Indian Institute of Technology (IIT)-Guwahati, India. Dr. Sarma has worked on dam-break flood wave propagation modeling of some of the Himalayan rivers in the state of Assam located in the northeast part of India. Dr. Sarma was here in the USA to participate in the Environmental and Water Resources Institute conference of the American Society of Civil Engineers in Omaha, Nebraska.

NDWRRI co-sponsored the Third International Water Conference titled "Research Education in an International Watershed: Implications for Decision Making" in Grand Forks, North Dakota, in March 13-15, 2007. The conference organized by the International Water Institute brought administrators, researchers, professionals and educators to Grand Forks, North Dakota to discuss water resources, flood control and water quality management issues related to the Red River of the North. The WRRI Director worked with the IWI in the planning and organization of the conference. He developed and chaired two sessions in the conference which provided a forum for presentation of the WRRI Fellowship research to the public. Several WRRI Fellows and their advisors participated. Ten past and present WRRI Fellows and three advisers made presentations at the conference. The Institute affiliate faculty chaired three session in all:

Concurrent Session 4C: **Water Quality in Rivers and Streams**
*Moderated by **G. Padmanabhan**, Director, ND Water Resources Research Institute, North Dakota State University, Fargo, ND.*

1. Upper Souris River TMDL Background Study. *Wei Lin, Civil Engineering and Environmental and Conservation Sciences, Bernhardt Saini-Eidukat, Geosciences and Environmental and Conservation Sciences, Joseph Super, Environmental and Conservation Sciences, North Dakota State University, Fargo, ND, and Michael Ell, Environmental Administrator, North Dakota Department of Health, Bismarck, ND.*

2. Simulation of Constituent Transport in the Red River of the North Basin, North Dakota and Minnesota, During Unsteady-Flow Conditions, 1977 and 2003-04. *R.A. Nustad, Environmental Engineer, USGS, North Dakota Water Science Center, Grand Forks, ND, and J.D. Bales, Hydrologist, USGS, North Carolina Water Science Center, Raleigh, NC*

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3. Application of Entrapped Cell Systems for Treatment of Anaerobic Sludge Digester Supernatant. *Christopher Hill and Eakalak Khan, Associate Professor, Department of Civil Engineering and Construction, North Dakota State University, Fargo, ND.*

Concurrent Session 6A: **Soils, Bacteria and Wastewater Treatment.**
*Moderated by **G. Padmanabhan**, Director, ND Water Resources Research Institute, North Dakota State University, Fargo, ND.*

1. The Fate of Manure-borne, Land-Applied Hormones. *Francis Casey, Associate Professor, Mary Schuh, graduate student, Dept. of Soil Science, Gerald L. Larsen, Research Chemist/Research Leader, Heldur Hakk, Research Chemist, Bioscience Research Lab, USDA-ARS, Fargo, ND, Zhaosheng Fan, graduate student, Dept. of Soil Science, North Dakota State University, Fargo, ND.*

2. Denitrification in the Red River Basin and Beyond: How Aquifer Sediments Influence Water Quality. *Scott F. Korom, Associate Professor, Department of Geology and Geological Engineering, University of North Dakota, Grand Forks, ND.*

3. Sensitive Determinatrion of Bacterial Cells Using Fluorescent Nanoparticles. *Yuhui Jin, graduate student, Dept. of Chemistry, Jenna Parisien, student, Dept of Chemistry, Min Wu, Assistant Professor, Dept. of Biochemistry, School of Medicine and Health, and Julia Xiaojun Zhao, Associate Professor, Dept. of Chemistry, University of North Dakota, Grand Forks, ND.*

Concurrent Session 6C: **Flood, Droughts and Water Management.**
*Moderated by **Phil Gerla**, Professor, Department of Geology and Geological Engineering, University of North Dakota, Grand Forks, ND.*

1. Characteristics of Effective Local Water Management Organizations in the Red River Basin. *Craig C. Kritsky, graduate student and Robert R. Hearn, Assistant Professor, Department of Agribusiness and applied Economics, North Dakota State University, Fargo, ND*

2. The Red River Management Consortium. *Daniel J. Stepan, Senior Research Manager, Energy & Environment Research Center, University of North Dakota, Grand Forks, ND.*

The presentations by the Fellows and Institute affiliate faculty can be viewed at the International Water Institute's web site:

http://www.internationalwaterinstitute.org/2007proceed.htm

Research results of NDWRRI Graduate Research Fellows were published and presented in various other conferences also.

PUBLICATIONS AND PRESENTATIONS

From 2006-07 Fellows and PIs

Journal papers

1. **Jin, Y., Zhao, X**., Wu, M. Determination of bacteria using luminescent nanoparticles, Pittcon 2006, Orlando, FL, March 2006

2. **Jin, Y.**, Parisien, J., Wu, M., **Zhao, X.**, Simultaneous Detection of Multiple Bacterial Cells Using Fluorescent Nanoparticles, Pittcon 2007 Chicago, Feb 2007

Presentations

- **Anderson, Ara** and **Butler, M. G**. 2006.The life history of Hexagenia limbata (Serville) (Ephemeroptera:Ephemeridae) in two Minnesota streams, Poster Presentation, North American Benthological Society (NABS) annual meeting. Anchorage, AK. 6 June.
- **Anderson, Ara** and **Butler, M. G.** 2006. The life history of Hexagenia limbata (Serville *Ephemeroptera:* Ephemeridae) in the Straight, Shell, and Toad Rivers, MN." Oral Presentation, Northern Plains Biological Symposium, Fargo, ND. 7 April.
- **Hill, Christopher** 2006. Using Entrapped Cell Systems to Remove Nitrogen from Anaerobic Digester Supernatant. North Dakota State University Civil Engineering Graduate Seminar.
- **Hill, Christopher** and **Eakalak Khan.** 2007. Application of Entrapped Cell Systems for Treatment of Anaerobic Sludge Digester Supernatant. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Jin, Y.**, Jenna Parisien, M. Wu, and **Julia Zhao**. 2007. Sensitive Determination of Bacterial Cells Using Fluorescent Nanoparticles. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Konsti, M. K.**, K.D. Zimmer, B.R. Herwig , M.A. Hanson, J.A. Younk, and **M.G. Butler.** 2007. Effects of Macrophytes, Nutrients, and Fish on Periphyton Abundance in Shallow Lakes. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Lenarz, William** and **Philip J. Gerla**. 2007. Effect of Land Cover and Pattern on the Quality of Groundwater Discharged from Springs at The Nature Conservancy's Pigeon Point Preserve, Southeastern North Dakota. Third

International Water Conference, International Water Institute, Grand Forks, March 13-15.

- **McEwen, Daniel C**. and **Butler, M. G**. 2007. Macroinvertebrate Responses Associated with a Change in Water-Level Regulation of Lakes in Voyageurs National Park, Minnesota. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Casey, F**., **Mary Schuh**, G. L. Larsen, Heldur Hakk and Zhaosheng Fan. 2007. Fate of Manure-borne, Land-Applied Hormones. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Tackett Ali**, 2006. 54th annual meeting of the North American Benthological Society, June.
- **Tesfay, T**. and **Korom, S. F**. 2006. The relative roles of electron donors in aquifer denitrification reactions: insights from geochemical modeling. 40th annual meeting, North-Central section, the Geological Society of America, University of Akron, Akron, Ohio, April 20-21, 2006.
- **Zheng, Wei**. 2006. A baseline study of aquatic macroinvertebrates on the Red River of the North, NDSU Environmental and Conservation Sciences seminar Fargo, ND May 25.
- **Zheng, W**. and **Butler, M. G**. 2007. Composition and Seasonal Pattern of Invertebrate Drift in the Red River of the North near Fargo, ND. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Korom, S**. 2007. Denitrification in the Red River Basin and Beyond: How Aquifer Sediments Influence Water Quality. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Hearne R**. 2006. Water Quality Monitoring Among Local Agencies in the Red River Basin. Presented at the 2006 National Water Quality Monitoring Conference. San José CA. May 9 2006.
- Kritsky, C. C. and **Robert Hearne**. 2007. Characteristics of Effective Local Water Management Organizations in the Red River Basin. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Hearne R**. 2006. Criteria and Indicators for Effective Water Management Institutions Presented at the Red River Basin Conference Winnipeg, Manitoba, January 12.
- **Hearne R**. 2006. Interbasin Transfer to Reduce Flooding: The Case of Devils Lake, North Dakota. Presented at eht University Council on Water Resources Conference. Santa Fe, NM. July 18.

From Prior Fellows and PIs

Journal papers

- J. R. Williams, W. L. Harman, M. Magre, **U. Kizil**, J. A. Lindley, **G. Padmanabhan**, and E. Wang. APEX Feedlot Water Quality Simulation, Transactions of American Society of Agricultural and Biological Engineers, Vol. 49(1): 69-73, January 2006
- **U. Kizil, J. A. Lindley, and G. Padmanabhan**. 2006**.** Verification of Nutrient Transport Modelling of a Bison Feedlot, Biosystems Engineering, Vol. 94 (3): 453-460**.**

Presentations

- Larsen, A., Michael Ell, and **Christina Hargiss**. 2007. Macroinvertebrate Methods Comparison Between the North Dakota Department of Health and the Minnesota Pollution Control Agency. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Potthof, A. J.**, K.D. Zimmer, B.R. Herwig, M.G. Butler, M.A. Hanson, J.R. Reed, B.G. Parsons, M.C. Ward, D.W. Willis. 2007. Effects of Piscivore Introduction on Prairie Wetland Ecosystems: Trophic Interactions Can Alter Ecosystem Structure. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Nustad, R. A.** and J. D. Bales. 2007. Simulation of Constituent Transport in the Red River of the North Basin, North Dakota and Minnesota, During Unsteady-Flow Conditions, 1977 and 2003-04. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Lin, W.**, B. Saini-Eidukat, J. Super, and Michael Ell. 2007. Upper Souris River TMDL Background Study. Third International Water Conference, International Water Institute, Grand Forks, March 13-15.
- **Hargiss, C.L.M.**, E.S. DeKeyser, **D. Kirby**. 2006. Development and Evaluation of an Index of Plant Community Integrity for Assessing Wetland Plant Communities. 2006 Annual Society of Range Management Meeting, Vancouver, BC.

THESES AND DISSERTATIONS

- **Gautam, Brajesh**. 2006. M.S thesis, Department of Civil Engineering, College of Engineering and Architecture, North Dakota State University, Fargo, North Dakota.
- **Tesfay, Tedros**. 2006. Ph. D Dissertation, Department of Geology and Geological Engineering, University of North Dakota, Grand Forks, North Dakota.

FELLOWSHIP and 104 (G) PROJECTS

Description of WRRI Graduate Research Fellowship and 104(G) projects follow:

THE IMPACT OF WETLANDS AND WETLAND EASEMENTS ON NORTH DAKOTA LAND VALUES

GRF Project 2005ND78B
Fellow: Kendall Goltz
Adviser: Jay Leitch
Department of Agricultural Economics
North Dakota State University
Fargo, ND 58105

A Synopsis of the Problem and Its Importance

Wetlands are ubiquitous across the Prairie Pothole region of North and South Dakota and Minnesota and numerous federal, state, and non-governmental agencies are actively involved in purchasing wetland conservation easements from private landowners. Common examples are the Small Wetland Acquisition Program of the United States Fish and Wildlife Service (USFWS), the Wetlands Reserve Program (WRP), and the Natural Resource Conservation Service (NRCS). To ensure that such programs are effective in encouraging landowners to place wetlands under easement while minimizing the expenditure of public funds, it is necessary that the impact of both wetlands and wetland easements on land values be fully understood.

Research Accomplishments to Date (June 17, 2006)

4,332 agricultural land sales from the years 2000 to 2004 have been collected from county courthouse public records in all 53 North Dakota counties. 775 of the 4,332 total land sales were found to contain wetland easements. The boundaries of all sales tracts have been digitized into a GIS database by identifying and selecting field boundaries in the common land unit (CLU) database of the NRCS based on the legal descriptions of the database. Land uses (acres of cropland vs. pastureland) have been calculated through spatial overlay of the year 2003 or 2004 NASS CDL coverages. The relative productivity of the sale parcels is represented by spring wheat yields for cropland, and pounds of forage per acre for pastureland. These soil productivity measures were calculated through spatial overlays of tracts and the SSURGO soils database. The type of wetlands found within the sale tracts has been quantified using spatial overlays of the NWI Basin. Conservation easement acres within sale tracts were determined by overlaying USFWS easement boundaries with the sale tract and NWI wetland coverages. The hydrologic (wetness) condition of all wetlands and easements has been determined using spatial overlays of water classifications of the NASS CDL, and visual inspections of overlaid NAIP color aerial photography. Three comparable sales without easements have been

identified for each of the 775 sales with easements. An extensive background review of the literature relating to wetland easements has been completed.

In addition, 210 land purchasers that reside either in one of the larger North Dakota cities or outside the state were identified as potential recreational land buyers. Survey information about the nature of the sale was obtained from 152 of the 210 land purchasers. These estimates are expected to be used by the USFWS, the USDA-NRCS, rural appraisers, land owners, and land buyers/sellers to evaluate fair market prices for land purchases with and without different types of wetlands.

Work Remaining Under This Project

Two counties in the prairie pothole region have been selected for an in-depth analysis of the impact of wetlands on the value of North Dakota agricultural land, one in the northern prairie pothole region and one in the southern prairie pothole region. Within each region, the county containing the most USFWS easements was chosen for further study because it is assumed that a greater number of easements indicate a county with a greater value from the perspective of social welfare. Towner County in the north portion contains the most sales with USFWS easements with 65 (and 60 non-easements sales) and McIntosh County in the south contains the most sales with USFWS easements with 105 (and 113 non-easement sales).

Similar sales will be evaluated on the basis of the attributes of the sale:

1. sale size
2. predominant land use
3. spring wheat yield
4. forage yield
5. % wet wetlands
6. distribution of wet wetlands
7. % dry wetlands
8. distribution of dry wetlands

Parcels will be grouped according to four sale sizes: quarter section, half section, section, and larger than one section. For each sale the predominant land use will be noted. If the sale contains 70% cropland or more, the sale is classed as a crop sale. If the sale contains 70% grassland or more, it is classed as a pasture sale. If the sale contains less than 70% cropland or less than 70% grassland it is considered a mixed sale. Sale size, predominant land use, spring wheat yield of the crop land, and the forage yield of the grassland will be used to determine sales comparable to the subject sale (contains wetlands), but that do not contain wetlands.

The percentages and distributions of the wet wetlands and the dry wetlands will then be used to determine the value of wet wetlands and dry wetlands by comparing the per acre price of the subject sale with per acre price of the comparable sales.

Using Entrapped Cell Systems for Treating Supernatant from Anaerobic Digester of the Moorhead Wastewater Treatment Plant

GRF Project 2006ND99B
Fellow: Christopher Hill
Adviser: Eakalak Khan
Department of Civil Engineering
North Dakota State University
Fargo, ND 58105

Brief Project Description

The scope of this research consists of studying the use of entrapped cells to treat supernatant from Moorhead Wastewater Treatment Plant sludge digesters. There are a number of benefits to side stream treatment of this supernatant: Accommodation of lower flows; prevention of shock to the mainstream biological process; and prevention of the bypass of ammonia to the river. Cell entrapment is a process which has been applied to domestic wastewater for the removal of both organic carbon and nitrogen. This is the first study to apply the cell entrapment process to highly concentrated wastewater. The main focus of this research is on the removal of nitrogen from Moorhead Wastewater Treatment Plant supernatant. Typically nitrogen is removed through a series of biological processes, nitrification followed by denitrification. Entrapped cell systems can follow this conventional configuration or nitrification/denitrification can be performed in a single reactor. The oxygen diffusion limitation of the entrapment matrix creates an environment in which both nitrifying and denitrifying bacteria can coexist. Both conventional two-step and simultaneous nitrogen removal are investigated in this research.

Progress of Work

In addition to the comprehensive literature review which began before and will continue throughout this fellowship, a substantial amount of time and energy has been spent in the Environmental Engineering Laboratory, North Dakota State University and at the Moorhead Wastewater Treatment Plant. The research progress thus far is briefly described below.

The characteristics of the supernatant are of importance. Therefore, a statistical analysis was performed, using the SAS Enterprise Guide, on data obtained from the plant. The results are presented in Table 1. The data used were based on the total population of samples collected and tested between January 1, 2005 and September 19, 2005.

Table 1 Statistical Analysis Results

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Summary Statistics Results

The MEANS Procedure

Variable	Label	Mean	Std Dev	Minimum	Maximum	N	Lower 95% CL for Mean	Upper 95% CL for Mean
TS	TS	0.2319048	0.0247476	0.2100000	0.3300000	63	0.2256722	0.2381374
TVS	TVS	55.8555556	3.1936822	50.0000000	61.0000000	63	55.0512371	56.6598740
COD	COD	2426.73	324.3843403	1624.00	3400.00	63	2345.04	2508.43
TBOD5	TBOD5	49.3555556	18.4147187	27.6000000	87.6000000	63	44.7178687	53.9932424
TSS	TSS	464.7619048	100.2393449	260.0000000	940.0000000	63	439.5169527	490.0068569
NH3-N	NH3-N	2055.75	301.1676721	1535.00	2348.00	63	1979.90	2131.59

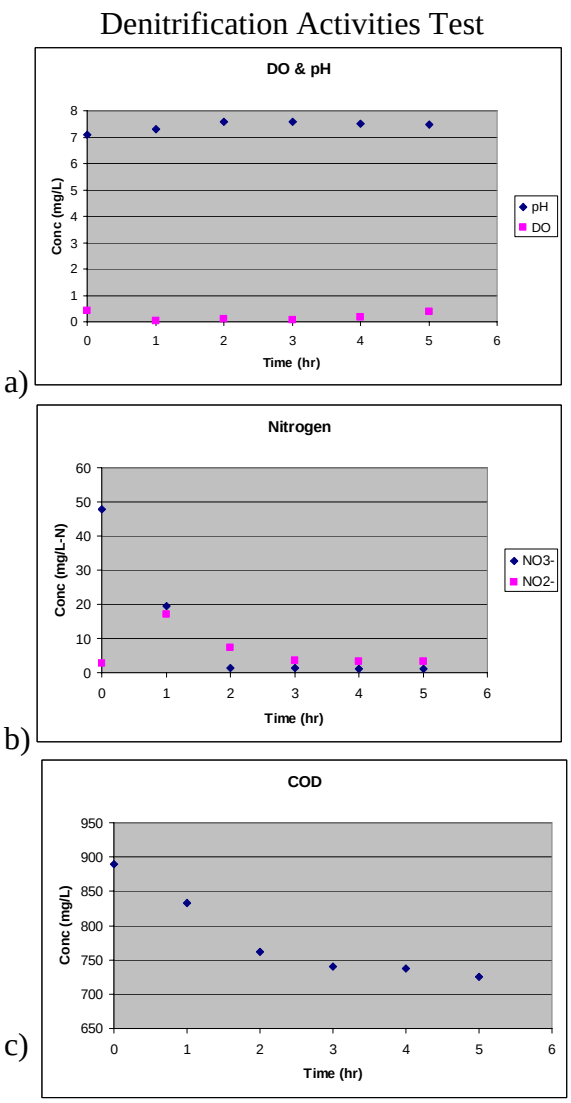
Note: TS, are in %; TVS, are in % of TS; COD, are in mg/L; TBOD5, are in mg/L; TSS, are in mg/L; NH3-N, are in mg/L as nitrogen

Nitrifying and denitrifying bacteria were cultivated initially using synthetic wastewater until sufficient biomass was obtained, approximately 3 months. After testing the activity of the bacteria (data not shown), they were then entrapped both separately and combined (1.5 nitrifier mass:1 denitrifier mass) into a cellulose triacetate matrix. The activity of the nitrifying bacteria after entrapment was virtually zero and suggests that the procedures for entrapping nitrifying bacteria in cellulose triacetate may substantially reduce their activity. It was decided that calcium alginate matrix should be explored in further entrapment experiments.

Once again, nitrifying and denitrifying bacteria were cultivated until sufficient biomass was obtained, approximately 3 months. For the denitrifying bacteria, synthetic wastewater was used as before but for the nitrifying bacteria the supernatant from Moorhead Wastewater Treatment Plant was used. After testing the activity of the bacteria, they were entrapped, both separately and combined, into a calcium alginate matrix. The pre-entrapment activity test results are shown in Figures 1 and 2.

Both activity tests illustrate that the bacteria are performing as expected. It should be noted that the nitrifying culture appears to be composed of mainly ammonia oxidizing bacteria because there is an accumulation of nitrite. This is often seen in high concentrated ammonia wastewater due to the fact that ammonia levels are toxic to nitrite oxidizing bacteria.

Denitrifying bacteria require organic carbon as electron donors to remove nitrogen from wastewater. It can be seen from the statistical analysis (Table 1) that there is a significant amount of chemical oxygen demand (COD) in the supernatant. However, based on corresponding biochemical oxygen demand (BOD), which was very low, most of COD was biorecalcitrant. “Hard” COD is the description typically given to the COD found in sludge digester supernatant. “Hard” referring to the difficulty microorganisms have in consuming this organic carbon. The source of organic carbon is currently being investigating for the denitrifying bacteria in an entrapped cell system. A study of the co-entrapped systems for nitrogen removal with and without the addition of methanol as a carbon source is the latest experiment. Due to the fact that the experiment is not complete the results will be reported at a later date.



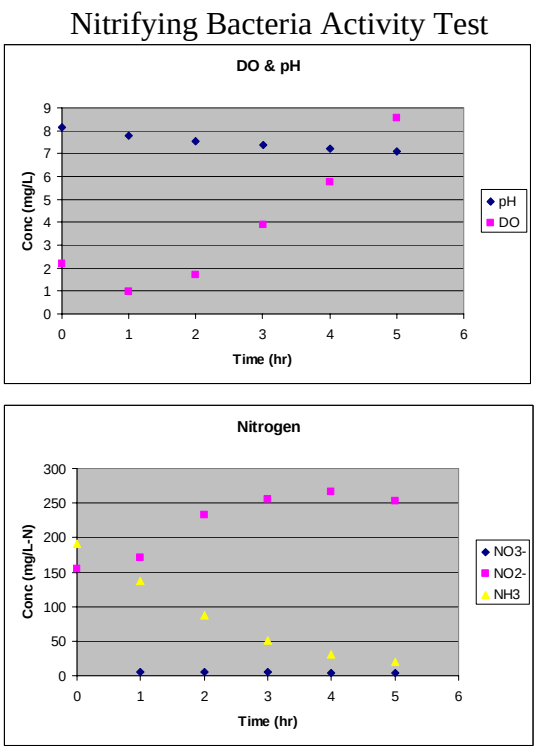


Figure 2 Nitrification Activity Test a) pH & DO b) Nitrogen

Figure 1
Denitrificaiton Activity Test a) pH & DO b) Nitrogen c) COD

Work Remaining

The work remaining for this fellowship includes the completion of the activity and carbon utilization kinetics. Two experiments for the activity and carbon utilization kinetics have been complete. It is estimated that two or three more experiment are require to obtain solid data for analysis. After each experiment, a representative sample of entrapped cells is collected and stored. These samples are to be analyzed using fluorescence-antibody labeling, detecting the spatial location of nitrifying and denitrifying bacteria and possible shedding light on the interaction between the bacteria in the entrapped cell system. After or during the fluorescence experiments, the reactors will be setup and operated in a CSTR configuration and data collected. The data shall be analysis to determine to most feasible configuration and a paper will be submitted to report the findings.

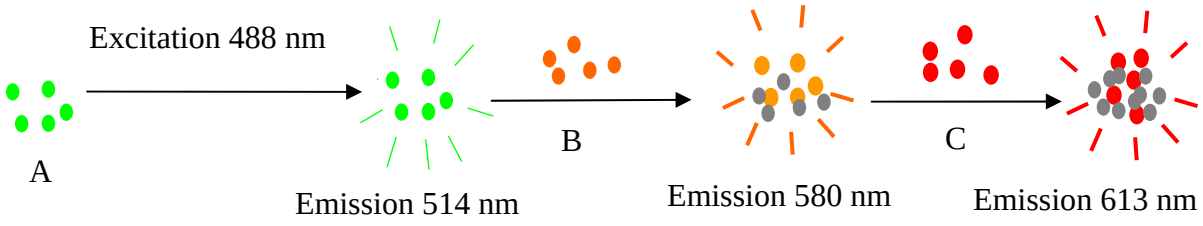
RAPID AND SENSITIVE DETERMINATION OF BACTERIA IN WATER USING NANOPARTICLES

GRF Project 2006ND98B
Fellow: Yuhui Jin
Adviser: Julia Xiaojun Zhao
Department of Chemistry
University of North Dakota
Grand Forks, ND 58202

1. Significance of the project

The rapid and sensitive determination of pathogenic bacteria is important in the fields of biotechnology, medical diagnosis, and water analysis. Currently, conventional methods either are low sensitive or require long detection time. A rapid and sensitive bacteria determination method is critically needed. Quantum dot (QD) is a novel nanomaterial that has attracted researchers' great interest due to its unique advantage, which is simultaneously giving different emission wavelengths based on varying sizes when a single excitation source is used. However, quantum dots are restricted by their low quantum yield that results in limited signals when they are used as luminescent labeling materials. Meanwhile, most of QDs are toxic to living systems which limits their applications in biological and medical fields. Luminescent silica nanoparticle containing high quantum yields of organic dye molecules emits much stronger fluorescent signals than QDs, which has been successfully used for trace amount of DNA detection and other biochemical analyses. However, luminescent nanoparticles can only emit within a fixed narrow range of wavelengths. The proposed nanoparticles will possess the advantages of quantum dots and luminescent nanoparticles, and overcome their disadvantages. Meanwhile, the silica based fluorescent nanoparticles are not toxic to living systems according to current research results. This nanoparticle is expected to be of great interest as a novel labeling nanomaterial for simultaneous analysis of multiple biomolecules, especially for labeling cells.

The key idea of this design is to use energy transfer of fluorescence molecules to produce various dye-doped nanoparticles with different colors using a single excitation source. For instance, as shown in below schematic diagram, the emission wavelength of fluorescence molecule A is near the absorbance wavelength of fluorescence molecule B. As molecule A's excitation light is applied to a solution containing molecule A only, molecule A will emit at 514 nm. Subsequently, molecule B is added to the molecule A solution and thus form a mixture of A and B (with a proper ratio of A to B), the emission



energy of molecule A is absorbed by molecule B. So, the emission of molecule B is observed and molecule A is quenched. Furthermore, molecule C is added to the mixture of A and B, the emission energy of molecule B will be absorbed by molecule C. Then, the mixture will emit C's spectrum and A and B are all quenched. Therefore, different mixtures will simultaneously emit different colors using a single excitation source. By doping a mixture of dyes into a silica matrix, quantum dots-like nanoparticles will be produced.

2. Progress Review

2.1 Material used: Tetraethylorthosilicate, triton X-100, succinic anhydride, Z-morpholinoethane sulfonic acd (Mes), BSA, 1-ethyl-3-3(3-dimethylaminopropyl) carbodiimide hydrochloride, and *N*-hydroxy-succinimide were purchased from Sigma-Aldrich. Avidin conjugated fluorescein, and dextran (Mw 3000) conjugated tetramethylrhodamine biotin were purchased from Molecular Probe, Invitrogen. Ammonium hydroxide (28-30 wt%) was purchased from Fisher Scientific. Klebsiella pneumoniae antibody was purchased from Biogenesis (NH, USA). Pseudomonas aeruginosa was donated from Dr. Wu, department of biochemistry and molecular biology, UND. Pseudomonas aeruginosa (PA) and Klebsiella pneumoniae (KP) are the lung bacteria which can induce lung infection. These bacteria were got from Dr. Wu, department of biochemistry and molecular biology, UND. All the bacteria operation was performed in the department of biochemistry and molecular biology of UND.

2.2 Synthesis of carboxyl coated silica nanoparticles

The carboxyl coated nanoparticles were first prepared using a microemulsion method. FRET dyes, avidin conjugated fluorescein (Av-F) and dextran conjugated tetramethylrhodamine with biotin (B-TMR), were doped inside nanoparticles with different ratios.

2.3 Preparation of antibody conjugated nanoparticles

0.5 mg nanoparticles were activated using 0.5 ml of 100 mg/ml 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride (EDC) and 0.5 ml of 100 mg/ml N-hydroxy-sccinimide (NHS) in a Z-morpholinoethanesulfonic acid (Mes) buffer (pH 6.0), for 25 min at room temperature with continuous stirring. Water-washed nanoparticles were dispersed in 5 ml of 0.1 M PBS (pH 7.3). To covalently immobilize antibody onto the nanoparticle surface, 5 ml of 0.1 mg/ml nanoparticles was reacted with 2 ml of 5 μg/ml antibody for 2 hours at room temperature with continuous stirring to form the resultant antibody-conjugated nanoparticles, followed by washing with a PBS buffer. Finally, these antibody conjugated nanoparticles were dissolved in 2ml 0.1 M PBS (pH 7.3) with 0.1% BSA to reduce the effect of nonspecific binding in the subsequent immunoassay. The solution was stored at 4 ^{o}C.

To determine the conjugation efficiency, UV spectra of these bioconjugated nanoparticles (without BSA) were measured. A strong absorption peak was found at 254 nm from both pure antibody and antibody conjugated nanoparticles indicating present of antibody on the nanoparticle. The conjugation efficiency was then determined.

The fluorescent resonance energy can be transferred from Av-F to B-TMR when they are in the proper distance (<10 nm). Thus, the Av-F doped, Av-F and B-TMR doped silica nanoparticles can be illuminated simultaneously. The fluorescence spectra of FRET dye doped nanoparticles were obtained. Excited by a single excitation wavelength (495 nm), both the Av-F doped nanoparticle (FRET-A-NP) and Av-F and B-TMR doped nanoparticle (FRET-B-NP) were illuminated exhibiting different emission spectra. Under the confocal fluorescence microscope, different colored nanoparticles were observed with a single excitation source (excitation wavelength: 488 nm).

Bioconjugation of antibody to nanoparticles is one of the key steps in the whole project. To get a well dispersed bioconjugated nanoparticles, EDC needs to be dissolved at room temperature to prevent self-polymerization. EDC and NHS solutions should be fresh. To reduce crosslinking, the concentration of nanoparticle solution needed to be lower than 0.1 mg/ml. The conjugation efficiency and the dispersion of the nanoparticles were characterized by UV spectra and SEM images. The bioconjugated nanoparticles were well dispersed.

2.4 Determination of *Pseudomonas aeruginosa* (PA)

The developed nanoparticles have initially used to detect bacteria, PA. PA were first incubated with PA antibody conjugated FRET-A-NP. The hold process was monitored in the real time through confocal fluorescence microscope. Results showed PA bacteria were successfully tagged with bioconjugated nanoparticle, the accumulation of green colored (pseudocolor) nanoparticle was observed after 20 min of incubation. The results demonstrated that the bioconjugated silica nanoparticles were competent for rapid bacteria determination.

The linkage of bioconjugated nanoparticles onto bacteria also can be observed in SEM images. PA were incubated in agar plate overnight at 37 ^{o}C. A single clone was collected and cultured in 3 ml of lysogeny broth (LB) media at 37 ^{o}C for 5 h. 10^{8} bacteria were then washed and incubated with 1 ml PA antibody conjugated nanoparticles for 1 h at room temperature. The mixture was separated by centrifuging. The pellet was resuspended in 1ml PBS and filtrated through a 0.2 μm, 0.01% polylysine pretreated membrane. After fixation and dehydration steps, the bacteria on the membrane can be observed under SEM.

4. Work remaining under this project

The initial result has been reported in Pittcon 2006. The most recent result will be reported in Pittcon 2007. A manuscript is in the preparation. Currently, the Fellow is working on the goal 2.

ANALYSIS OF ASSOCIATED BEDROCK-AQUIFER SYSTEM SEDIMENTS: ORIGINS OF ELECTRON DONOR-RICH AQUIFERS IN EASTERN NORTH DAKOTA

GRF Project 2006ND101B
Fellow: Ryan Klapperich
Adviser: Scott Korom
Department of Geology and Geological Engineering
University of North Dakota
Grand Forks, ND 58202

Project Background

Nitrate pollution has long been recognized as the most prevalent form of groundwater pollution. The only effective process to remediate nitrate contaminants is denitrification, typically through natural attenuation. This process reduces nitrate (NO_3^-) to harmless nitrogen gas. The process occurs naturally, requiring only an oxygen-limited environment, the presence of nitrogen digesting bacteria, and the availability of electron donors. The three most common electron donors are organic carbon, sulfide (typically as pyrite, FeS_2), and ferrous iron minerals. Research has also shown that the controlling factor in this reaction has typically been the availability of suitable electron donors within the aquifer sediments.

The long-term goal of this research is to develop an aquifer nitrate vulnerability index based on the supply of electron donors in the aquifer sediments. This project contributes toward that goal by considering the abundance of electron donors in the likely parent materials comprising the aquifer sediments. Based on the experience of previous UND denitrification research and literature reports, shale units are more likely to have large supplies of electron donors than other regional rock types. Shale is relatively soft and easily weathered and comprises much of the bedrock in eastern North Dakota. Therefore, shale units are the most likely contributors to sediments in nearby aquifers. My hypothesis is that regional bedrock units rich in electron donors should correlate to nearby aquifer sediments that are potentially rich in electron donors. The practical utility of this work is that an initial list of aquifers with potentially high denitrification capacities in North Dakota may be made using geological investigations. Later, more extensive and more expensive geochemical analyses would be necessary to confirm the denitrification capacity of specific aquifers and specific aquifer sites. However, much of the initial work could be done with data already available and in conjunction with future drilling performed by the North Dakota State Water Commission (NDSWC).

Progress

Geochemical analysis and literature review are ongoing. The bedrock samples were collected this previous summer in conjunction with the State Water Commission. They were collected in a general east to west transect across Stustman, Foster, and Steele counties. Two to three samples were collected when the shale was penetrated, representing an upper and lower, and occasionally mid-level sample. Samples from Kidder, Benson, Nelson, and Cass Counties have also been provided from other drilling projects. In total there are 39 samples representing 21 different sites and seven lithologic units. XRD analysis has been preformed on many of the samples and shows that the shale units are rich in quartz, clay minerals, and occasionally calcite, as was expected. Pyrite, micas, and iron bearing minerals have also been detected. XRD has also verified that the lithologic units are generally uniform in space and depth, meaning a single sample can be considered representative. Work has also begun to analyze organic carbon content in the samples. There are not enough data to date to be conclusive. Analysis of other electron donors, sulfide and iron, will begin in earnest.

Significance:

This research will investigate the link between the denitrification capacities of eastern North Dakota aquifers with the electron donor composition of the surrounding bedrock. If such a link is possible, a qualitative index (low, medium, and high) of aquifer denitrification capacity based on the probable source of the parent material will be developed. This index could then be used to focus, in a cost-effective fashion, more extensive and expensive geochemical analysis on specific aquifers or zones in specific aquifers.

TOP-DOWN AND BOTTOM-UP EFFECTS ON THE ABUNDANCE OF PERIPHYTON IN SHALLOW LAKES

GRF Project 2006ND136B
Fellow: Melissa Konsti
Adviser: Malcolm G. Butler
Department of Biological Sciences
North Dakota State University
Fargo, ND 58105

Regional Water Problem

Shallow lakes are the most common lake type in North America, yet our ecological understanding of these systems is poor relative to deeper "sport fish" lakes. Throughout the Prairie Pothole Region (PPR), landscape alterations have directly and indirectly altered the character and quality of regional waterbodies. The ecological value of these shallow aquatic ecosystems decreases as conditions favor a turbid, phytoplankton-dominated condition with low abundance and diversity of invertebrates and submerged aquatic plants (Scheffer 1998). Waterbodies in the turbid state are considerably less valuable to migrating waterfowl than clear-water, plant-dominated systems. Much evidence points to nutrients as a cause of high periphyton biomass, just as nutrient loading enhances planktonic algae. Fish presence in a shallow basin may also favor increased algae, both planktonic and periphytic. Periphyton is detrimental to macrophytes, and ultimately may contribute to a basin shifting from the clear-water state to the less valuable turbid state. We need to better understand what controls periphyton, and the role it may play in shifts from the clear-water state to the turbid-water state in shallow lakes within the PPR.

Scope and objectives

This study will provide better understanding of interactions contolling to periphyton within shallow lake ecosystems, by accomplishing these objectives:

1. Sample the epiphyton, invertebrate, fish, macrophyte, phytoplankton, and nutrient/light interactions within 75 shallow lakes in Minnesota.
2. Sample shallow lakes of varying water clarity (clear- vs turbid-water state) and nutrient input (LO & HI impact landscapes).
3. Test various combinations of explanatory variables in models to determine if periphyton is controlled from the top-down, from the bottom-up, or by a combination of both.
4. Determine which variables best predict periphyton biomass (nutrients, light, fish, invertebrates).

5. Determine how periphyton biomass and its predictor variables differ between study landscapes, among lakes within each landscape, and with depth in each lake.

Methods

Two areas in western Minnesota have been chosen for study. The northern area is located in eastern Polk County, while the southern area spans the region from southern Grant County to the northern edge of Stevens County. Each study area comprises approximately 560 km^2. Study sites were selected by randomly choosing from groups of candidate lakes conforming to criteria identified as part of a broader study. While compiling candidate lakes, surface area, depth, distance to roads, adjacent upland cover and other criteria were taken into account. A total of 75 wetlands were chosen using this method, roughly half within the northern study area and remaining sites in the southern study area. All sites selected are semipermanent or permanent (type IV or V) with regard to duration of flooding (Stewart and Kantrud 1971). Periphyton, invertebrate, macrophyte, and fish abundance, plus nutrient and light availability were sampled throughout the 2005 and 2006 summers.

Periphyton biomass (Chl *a*) is determined by deploying artificial substrates for five weeks. Sampling devices were set out in mid-June and collected in late-July each year (average time in water is 5 weeks). These devices consisted of a polyester braided rope (¼" thick, 1½ m long) with a brick anchor attached to one end and a float on the other, with three vinyl microscope slides attached using zip-ties at individual depths along the rope (10, 50, and 90cm from surface). The total height of the sampling device was approximately 1.5m. Using artificial substrata instead of collecting living plants permits a uniform surface type, area, and orientation, and therefore less variation in the sampling of periphyton biomass. Some species of plants may be able to alter the periphyton community, so by using an artificial substrate the species of macrophytes present will not be a factor. Cattaneo and Kalff (1979) concluded that periphyton production did not differ between natural and artificial plants. Substrates were placed vertically in the water column at a depth of ~1.5 m. Since periphyton and grazer biomass varies with depth (James et al. 2000), deploying substrates at specific depths controlled for this influence. Three devices were deployed in each lake, near the same locations where invertebrates were sampled. Upon collection, each sample was removed from the lake with care to limit disturbance to the periphyton, placed in a container with tap water, and stored in a dark cooler until be processed in the lab within 12 hours.

Periphyton biomass will be estimated from chlorophyll *a* analyses (APHA 1989). Periphyton is scraped off slides into a dish with a razor blade, and a sub-sample was filtered onto a glass fiber filter (Whatman GF/C). Filters were frozen until processing in the lab. Each sample is allowed to steep for 24 hours in a separate tube with 90% acetone. Fluorometry is used for determining chlorophyll *a*, since it is more sensitive than spectrophotometry and thus requires fewer samples. Fluorescence is measured at 430 nm and 663 nm, and calibration factors are derived to convert fluorometric readings to concentrations of chlorophyll *a*.

Zooplankton were sampled twice per year, once in early-June, and again in Late-July, by collecting two replicate vertical column samples (Swanson 1978a) at six open water locations in each wetland. Estimates will be made of density, biomass, and taxon

richness of zooplankters. Relative abundance of free-swimming invertebrates was obtained with submerged activity traps (ATs) (Swanson 1978b, Murkin et al. 1983, Ross and Murkin 1989). Six ATs were deployed at the interface of open water and emergent macrophytes, and left in each wetland for 24 hours. Abundance (counts of dominant forms) and taxon richness of macroinvertebrates will be determined, paying special attention to identifying taxa considered to be grazers/scrappers.

Density and trends in abundance of submerged macrophytes were assessed using a modified technique of Jessen and Lound (1962) and Deppe and Lathrop (1992). In each wetland, submersed macrophytes were sampled at 20 stations in early August each year. Four transects were established perpendicular to the longest axis of the lake, with 5 stations established along each transect. Therefore, sampling stations were apportioned among 3 depth strata (open water, transition, nearshore). Two samples were collected from each station using a weighted plant rake, with frequency of occurrence calculated for each plant species and all taxa combined. The first sample was weighed to determine the relative abundance (mass) of macrophytes overall. Metaphyton (e.g. *Cladophora* spp.) and macroalgae (e.g. *Chara* spp.) were assessed along with vascular plant species during these surveys.

Three surface water samples were taken along the middle of each wetland during early-June, and again in late-July each year. These samples were stored on ice and transported immediately to the Minnesota Department of Agriculture chemistry lab (St. Paul, MN) for analysis of chlorophyll *a*, total and Kjeldahl nitrogen, and total phosphorus. Turbidity and specific conductance were measured in the field with a portable nephelometer and conductivity meter, respectively. Phytoplankton biomass were estimated from chlorophyll *a* (Strickland and Parsons 1972). Collection of samples for chlorophyll *a* simultaneously with measurement of turbidity allows assessment of the contribution of phytoplankton to turbidity, and ultimately to light attenuation. A secchi disk was also used to determine water clarity, by sampling the middle of each lake once in early-June, and again in late-July each year.

All fish sampling was conducted during late-July each year. Three mini-fyke nets (9.5 mm bar mesh with 4 hoops, 2 throats, 7.62 mm lead, and a 0.69 X 0.99 rectangular frame opening into the trap) were set overnight in each lake. One experimental gill net (76.2 m multifilament net with 19, 25, 32, 38, and 51-mm bar meshes) was set along the deepest depth contour available in wetlands less than 2-m deep or parallel to shore along a 2-m contour in wetlands with sufficient depth. This protocol has been shown to be effective in sampling fish assemblages in small lakes from other regions (Tonn and Magnuson 1982, Rahel 1984, Jackson and Harvey 1989, Robinson and Tonn 1989). This should enable us to capture both small- and large-bodied fish, and species from all the major trophic guilds (e.g. planktivores, benthivores, and piscivores) potentially present in the study wetlands. All species of fish sampled were counted and returned alive to the wetlands if possible. Voucher specimens were retained for laboratory identification when field identification could not be made.

Multivariate techniques will be used to interpret the interactions between biotic and abiotic variables. Specifically, two types of gradient analysis, principle components analysis (PCA) or correspondence analysis (CA) will be used (ter Braak 1995). These techniques are able to identify and summarize major patterns in the data, and by considering all species and sites at once, they permit detection of patterns that univariate

techniques are unable to identify. Such patterns include which species vary the most among sites, which species have positive/negative associations, which species are most abundant in specific sites, as well as which sites are most similar/dissimilar in terms of species abundance, and how much individual sites differ in community composition. Abundance and composition of fish, invertebrates, and macrophytes may be correlated with the various variables, such as nutrients, light, and periphyton biomass.

Finally an information-theoretic (IT) approach, a type of model selection, will be used to determine which variables are related to periphyton biomass. Models will be selected *a priori* to ensure inclusion of appropriate variables, and comprised of various combinations of the variables. This approach will then be used to select the model best supported by the data, and dismiss others that are less plausible. Model selection considers both problems associated with overfitting the data (using too many parameters) and underfitting the data (using an insufficient number of parameters).

Deliverables

This study will improve understanding of the fish-invertebrate-periphyton-macrophyte cascade, including direct impacts of nutrients and light on periphyton biomass, and the indirect impact of periphyton on macrophyte abundance. An understanding of which variables most influence periphyton abundance can help managers maintain these shallow lakes in the clear-water state. Manuscripts will be prepared and submitted describing these findings.

Progress

Field data has already been collected for the full two years of the study. We sampled fish, macrophytes, nutrients, phytoplankton, periphyton, and invertebrates. Many invertebrate samples still need to be processed, but other lab work is well underway. Work is in the early stages of analyzing data and writing manuscripts.

EFFECT OF FLOW PATH PROCESSES ON THE GEOCHEMISTRY AND QUALITY OF WATER DISCHARGED ALONG THE SEEPAGE FACE AT PIGEON POINT, SHEYENNE DELTA AQUIFER, RANSOM COUNTY, ND

GRF Project 2006ND125B
Fellow: William Lenarz
Adviser: Philip Gerla
Department of Geology and Geological Engineering
University of North Dakota
Grand Forks, ND 58202

Project Background:

The large seepage face at Pigeon Point, Ransom County, North Dakota, provides an opportunity to trace the evolution of groundwater geochemistry back to its source as infiltrating precipitation. Previous work delineated pathlines and the capture zone in the groundwater flow system, which extends several kilometers upgradient. The contrasting land cover within the spring and seep capture zone consists of dunes, native grass pasture, wetland, and irrigated cropland that lie above the phreatic Sheyenne delta aquifer. Previous work revealed that the seepage face shows a wide variation in mineralization and oxidation-reduction conditions, with strikingly more reduced and mineralized water discharging from higher areas indicating a shorter groundwater pathline. We hypothesize that the groundwater quality relates to vadose and shallow phreatic geochemical processes, which are largely controlled by differences in soils and land cover, and that water composition remains generally unchanged along deeper pathlines.

Project Objectives:

1. Install monitoring/sampling equipment along a groundwater pathline.
2. Select five springs to sample to represent best the varied water quality found within the discharge zone.
3. Collect samples of water during different seasons (Fall, Spring, and Summer) to determine if groundwater quality and chemical transport vary temporally across the study area. Seasonal samples will reveal how dissolved constituents in groundwater change during different times of the year.
4. Sample and analyze pH, conductivity, and dissolved oxygen in the field.
5. Analyze major anions, cations, and total and organic carbon content at UND's Environmental Analytical Research Laboratory using flame atomic absorption spectrometry (FAAS), total organic carbon analysis (TOC), and ion chromatography (IC).
6. In conjunction with soil water and groundwater sampling and analysis, work to better understand the physical conditions of infiltration and recharge will be completed during the project. At and near the instrumentation sites, matric potential, moisture content, and soil permeability will be estimated using transducer tensiometers, portable time-domain reflectometry surveys, and a

disk infiltrometer. These data will be used to create basic numerical models of infiltration and recharge using the USDA Salinity Laboratory's HYDRUS-2D code.

Progress to Date:

Installation of the monitoring/sampling stations along the selected groundwater flow pathline has been completed. Further site development was conducted to ensure successful water collection during the first and subsequent sampling events. All samples have been collected at this point, and analyses completed. All field work is done. Thesis writing has also begun, with a first draft of the thesis being half completed at this point.

Results:

Upon initial observations and comparisons of the results from all three sample collections, our initial hypothesis is holding true. The springs that are discharging at higher elevations (indicating shorter travel times and pathlines) are producing results indicating a much more reduced environment. The springs that are discharging at lower elevations (indicating longer travel times and longer pathlines) on the other hand are producing results indicating higher levels of oxidation. The common processes would lead to the idea that groundwater traveling over a shorter distance and shorter time span should discharge with a higher level of oxidation, while the groundwater traveling over the longer distance and taking more time should discharge more reduced. As our sample results are indicating, the spring and fens at Pigeon Point are unique in that the groundwater discharging is reversed from what would commonly be expected.

Significance:

Although the variation of geochemical composition of groundwater on a regional basis is generally understood, much less is known about the detailed, local processes that lead to the spatial variation of groundwater chemistry along a flowline. The groundwater flow system at Pigeon Point is well constrained physically, and shows remarkable variation across the seepage face. Results will provide a conceptual model on how groundwater composition evolves within this shallow aquifer flow system. The model will be used to explain the unusual variation of water quality at the seepage face and help predict changes in water quality following alteration of land cover. The groundwater flow system maintains wetlands that host rare and unusual flora found only in boreal region hundreds of kilometers to the northeast. The north-facing slope, shaded by some of the only remaining old-growth riparian forest in North Dakota, creates a unique environment where continuously moist and cool hardwood forest borders an extensive xeric plant community developed in nearby dunes. This valuable natural resource exhibiting an unusually large biodiversity, however, is threatened by the growing demand for fresh water in the region. The Sheyenne delta aquifer constitutes an important source of high quality groundwater in southeast North Dakota and may provide water for Fargo in the future.

STOICHIOMETRY AND THE TRANSFER OF MERCURY FROM BENTHIC MACROINVERTEBRATES INTO GAME FISH

GRF Project 2005ND74B
Fellow: Dan McEwen
Adviser: Malcolm G. Butler
Department of Biological Sciences
North Dakota State University
Fargo, ND 58105

Research

Mercury (Hg) damages the central nervous system, altering the way that nerves conduct electrical impulses and divide, leading to lowered cognitive and metal functioning or in especially acute circumstances cerebral palsy, mental retardation or death. Its effects are especially harmful to fetuses or infants during development of the nervous system. Hg emissions have continued to increase since the industrial revolution, entering aquatic food chains via atmospheric precipitation where they suspend in the water column and are taken up by phytoplankton or settle to the bottom where they become available to bacteria. Phytoplankton, with acquired Hg, is either ingested by zooplankton or sinks to the substrate as detritus. In turn, detritus and bacteria provide a food source to macroinvertebrate benthic organisms (i.e., benthos) and bioaccumulation occurs up through trophic levels eventually to game fish that are consumed by humans. As a result, the EPA and various state agencies issue advisories for fish consumption where impairment occurs. The North Dakota Department of Health currently has consumption advisories listed for Devils Lake, Red River, Lake Oahe/Missouri River, Lake Sakakawea, and other water bodies. Research is prposed for addressing taxonomy, stoichiometry, and benthic-pelagic coupling in the context of benthic contribution to Hg accumulation to fisheries in Minnesota and North Dakota lakes along a trophic gradient from oligotrophic to eutrophic.

Objectives and Methods

The objectives of this study are as follows:

1. Characterize the benthic community of lakes under a variety of nutrient regimes
2. Quantify the biomass of the constituent members of the benthic communities in those lakes
3. Quantify carbon, nitrogen, phosphorus, and mercury ratios for those communities as a whole
4. Model how those ratios impact rates of mercury accumulation and concentrations in piscivorous game fish likely to inhabit those lakes (e.g., lake trout, walleye, smallmouth bass, northern pike, etc.)

5. Use appropriate regression models to test whether there is a relationship between Hg and P (as a limiting nutrient) in the benthos
6. Provide applicable information for managers of mercury-susceptible systems

Project Progress

Twenty-four lakes, varying in nutrient status, have been sampled to date for benthic invertebrates ranging from shallow- eutrophic wetlands in North Dakota to deep, oligotrophic trout-lakes in the northeast Minnesota. Benthic samples have been dried for 24 h and dry mass was recorded for each taxonomic group. All organisms were homogenized and ground and subsamples are in the process of being analyzed for carbon, nitrogen, phosphorus, and mercury. These data will be used in a bioenergetics model to predict game fish species mercury concentration levels using literature-derived values for parameters not measured in the field.

Significance of Research

A framework will be provided by which lakes can be assessed for susceptibility of Hg concentrations. A stoichiometric underpinning can provide managers with a relatively easy method of assessing fish susceptibility to Hg by measuring a subsample of benthos for C:N:P.

COMPARATIVE STUDY OF FOSSIL AND EXTANT FISH GROWTH: INCLUDING ANALYSES OF MEAN ANNUAL TEMPERATURE IN THE GEOLOGIC RECORD

GRF Project 2003ND25B
Fellow: Michael Newbrey
Adviser: Allan Ashworth
Department of Geosciences
North Dakota State University
Fargo, ND 58105

DESCRIPTION OF THE REGIONAL WATER PROBLEM

It is important to consider the implications of climatic change on surface water resources in light of potential consequences of global warming. North Dakota boasts some of the best long-term data sets in the form of a fossil record to measure the effect of climatic warming on a single population of fish. Very little is known about growth and the life history characteristics of fish in the fossil record. Fossils can provide valuable information about growth of extinct forms of fish, thereby providing insight into their life histories and ecology. A fossil lake bed near Jamestown, ND will provide perhaps thousands of years of continuous data of fish growth during a warming climate. This research will provide insight for fishery biologists and wetland ecologists concerning the long-term response of contemporary fish growth in North Dakota given potential climatic changes.

PROJECT OBJECTIVES

The project entails an examination of the relationships between age, growth, longevity, and climate on a geologic scale. The objectives of this study are to: 1). examine the age and growth patterns of fossil freshwater hiodontids, esocids, and the percid, *Perca flavescens* from all fossil localities known to produce these taxa in North America; and 2). quantify patterns of growth of extant hiodontids, esocids, and the percid, *Perca flavescens* from a range of latitudes and ambient mean annual temperatures (MAT) to understand the effects of MAT on fish growth; 3) contrast the growth patterns from fossil fish to that of extant populations to examine evolutionary patterns.

PROGRESS

In previous research, we have contrasted growth of living forms of pike (*Esox*) to that of fossils. More recently, we have been working with yellow perch (*Perca flavescens*). The research has shown that growth of living and extinct closely related species are similar. By examining the growth patterns of contemporary pike and yellow perch across their ranges, we found that mean annual air temperature describes variation in growth. Furthermore, changes in age and growth of Esociformes and Hiodontiformes since the Cretaceous show trends in time that are correlated with climate change.

Pleistocene fossils can also be used to examine the effects of climate change on fish. We reexamined a well-preserved late Pleistocene to early Holocene fossil fish assemblage from lake deposits on the Missouri Coteau, near Buchanan, North Dakota. Our findings were published in the Canadian Journal of Fisheries and Aquatic Sciences. We reported that fossil fish abundance, stratigraphy, pollen, and charcoal provided information about postglacial colonization and the subsequent population fluctuations during a time of climatic warming. The fossil fish included complete specimens of *Perca flavescens, Hybognathus hankinsoni, Notropis heterolepis, Fundulus diaphanous, and Culaea inconstans*. The sequence of colonization for each species was correlated with individual thermal and relative water velocity tolerances. We found that fish abundance fluctuates six times during an approximate 1000 year depositional history. Charcoal abundance, representing fires, was inferred to represent episodic droughts during which nutrient levels were reduced and fish abundance declined. The fluctuations followed an overall trend to increased abundance during a time when the lake-margin vegetation changed from a spruce to a deciduous forest in response to climatic warming. The research provides insight into the effects of a changing climate on fish populations and demonstrates the potential of using fossils to examine long-term processes regarding contemporary fish species.

The research so far has 1) correlated contemporary yellow perch (Perca flavescens), muskellunge (Esox masquinongy), northern pike (Esox lucius), chain pickerel (Esox niger), goldeye (Hiodon alosoides), growth to temperature, 2) showed that climatic events such as that at the Cretaceous / Tertiary boundary and the Early Eocene Thermal Maximum are coincident with evolutionary events, 3) showed the effects of climate change on fossil fish colonization, biogeography, and population trends of fishes.

Some example results: centra from Cretaceous (70 million years old) hiodontids and are significantly smaller than those of 50 million year ago. Those centra in the Cretaceous and Eocene are both significantly smaller than those from ND today. As the climate has been cooling since the Cretaceous, hiodontids have been getting significantly larger, a pattern also seen in the pike family. Furthermore, major climatic shifts in the geologic record have been correlated with evolutionary shifts in the hiodontids and the pikes, which both inhabit North Dakota today. Life spans have been getting longer for both groups, and growth rates have been changing. The interesting correlation is that more ancestral, living members of these groups show similar significant trends and characteristics in growth, lifespan, total length at age four and other ages, and ultimate total length with regard to temperature across their ranges today. These significant relationships provide numerical models with which to quantify the effects of climate change on fish. The data suggests that as the climate warms fish will mature sooner at smaller sizes thereby suggesting effects on population dynamics, predator-prey relationships, and ultimately our fishery stocks!

A part of this research addresses the trends in fish movement across latitude and through time, which is the Fellow's dissertation research objective 2. The results are being used to interpret and corroborate the results from my other dissertation objectives concerning evolutionary shifts in growth patterns. The largest freshwater fossil fish database to date

has been constructed, which is nearly double the size of previous fish reviews published in 1981 and 1986 and consists of nearly 400 taxonomic entries. Preliminary results are very interesting and suggest that fish dispersal is tracking with climate. The age – latitude relationships for 54 taxa within 37 families of freshwater fishes were examined from over 150 fossil localities from the Late Cretaceous to the Pliocene (~100 to 2 mya) in North America. To examine the long-term effects of climate, we compared the changes in latitudinal data with changes in paleotemperatures based on the oxygen isotope analyses of benthic foraminifera. A regression analysis indicates the paleolatitude distribution of fishes is negatively correlated with paleotemperature. This relationship suggests that fish populations are shifting in response to changing thermal conditions, which may help explain many of the patterns in long-term fish dispersal. WRRI was acknowledged in both presentations and will be in the subsequent manuscript. The manuscript will be submitted to the journal Nature.

SIGNIFICANCE

Ultimately, this research will document the changes in evolution of growth of extinct species during climate change and help to understand how contemporary species respond to climate change.

FARM-SCALE RECONNAISSANCE OF ESTROGENS IN SUBSURFACE WATERS

GRF Project 2006ND100B
Fellow: Mary Schuh
Adviser: Francis Casey
Department of Soil Sciences
North Dakota State University
Fargo, ND 58105

RESEARCH PROBLEM

Estrogens are very potent endocrine disrupting chemicals and are naturally present in animal manures applied to the soil. In areas where there are concentrated animals, such as at confined feeding operations, there is increasing concern on both state and regional levels that hormones may be transported to surface water or to groundwater. Research has shown that parts per trillion 17ß-estradiol concentrations can cause male fish to express female characteristics. Humans are not as toxicologically sensitive to hormones compared to aquatic organisms, yet the amount of environmental exposure to hormones is still unknown. Because of this, the fate and transport of estrogens and quantification of estrogens introduced into the environment from animal operations is an important problem of state, national, and international concern.

Previous to this study, work was done at a swine (*Sus scrofa*) farm in North Dakota, including the installation of lysimeters to monitor the amount of 17ß-estradiol leached through soil and the installation of wells to monitor the surficial aquifer. For this reason, the above location was elected as the research site for this project. In a prior study various types of manure were applied to the surface of lysimeter plots. Based on known application rates in manure and laboratory-determined degradation rates, it was anticipated that most of the hormones would be metabolized or bound to the top 0.10 m of soil. Instead, data (unpublished) consistently shows low amounts of 17ß-estradiol leached through the soil and detected in lysimeter water (0.6 m below the surface; Fig. 1) and in the surficial aquifer (~0.5 – 3m below the surface; Fig. 2). 17ß-Estradiol was even found beneath a control plot, where no manure was applied, which provided evidence for the antecedent existence of this hormone in the soil or that it was leached from somewhere else.

OBJECTIVES

The objective of this project is to identify the causes of the unexpected detections of 17ß-estradiol. It is known that the farmer injects his swine liquid manure slurry from a lagoon holding pond into the field soil at this research location. It has also been determined that the soils in this area are aquic and exhibit redoximorphic features (i.e. faint mottling) within 0.16 m of the soil surface, suggesting large fluctuations in the water table. These high water tables are more prevalent in the springtime when the temperatures are still low. The combination of cool and wet anaerobic conditions inhibit microbial activity that is

necessary to degrade these hormones and may promote their persistence in the soil. Moreover, these soils are derived from water deposited sand, which decreases the sorption of 17ß-estradiol and further exacerbates the transport of this hormone. The following hypotheses were proposed for causes: (1) There exists high levels of background 17ß-estradiol from the lagoon material the farmer injected in the field; (2) High water-table conditions cause a cool and anaerobic environment in which 17ß-estradiol persists; and (3) The ground water that contains the 17ß-estradiol is transporting exogenous hormones into the lysimeters in the soil horizon.

PROJECT PROGRESS

The proposed sample design has been implemented and samples have been taken. The samples have been prepared for analysis; however, the chemical analysis has not yet been initiated. Once this analysis is completed, the data can be evaluated to make further assertions. Additionally, wells and lysimeters from this research location have been sampled in an ongoing manner. The samples have been analyzed and the data has been summarized. This data will help support and interpret the main objective of the current research project.

Sampling Strategy

A sampling strategy was formulated to allow the comparison of spatial and temporal variability of 17ß-estradiol in soil at the swine farm selected for this research. Five locations for soil cores were chosen in and around a field where liquid lagoon slurry was to be injected as fertilizer. The soil core sites were chosen based on nearness to regularly monitored wells – so soil concentrations could be easily compared to groundwater concentrations – and contact with surface conditions deemed as having potential to compromise groundwater quality, i.e. next to an uncovered compost heap, near a holding pond for lagoon slurry, and in a field injected with lagoon material. Also, samples were taken through time to observe any temporal fluctuations in 17ß-estradiol concentration due to temperature and high water tables, or manure application. Soil core samples of the field soil before and after the lagoon manure was applied will provide information about the fate and persistence of manure-borne hormones.

Soil Coring Method and Procedure

Samples were taken using a Giddings probe with a 2-5/8-inch bit. The core barrel was lined with a 4-foot polyethylene liner tube with a 2-3/8-inch diameter that encased the sample. After removal of the sample tube from the barrel, the ends of the tube were sealed with plastic end caps and waterproof tape to prevent moisture loss. Cores were subsequently labeled and placed upright under a tarp to minimalize disturbance and prevent photodegredation due to exposure to light.

Geographic Positioning Sensor (GPS) coordinates were recorded to allow replication of the sites, and the holes were filled with a mixture of bentonite and sand. Cores were transported to the USDA-ARS Biosciences Research Laboratory where they were placed in a freezer until ready for extractions and analysis using Liquid Chromatography with tandem Mass Spectrometers (LC-MS-MS).

Sampling Accomplished

To date, duplicate core samples were taken from each location except for the injection field. Here four cores were taken, since this is where the hormone concentrated slurry is actually injected below the surface, making it the prime site of interest. The lagoon material that was applied to this site was sampled for hormone characterization and quantification. Additionally, the soil core locations in this field were directed towards topological high and depression features (where subsurface water recharge is directed). Thus, between the five sites, we planned to obtain a total of 12 core samples for each set.

On May 9, 2006 a total of six cores were taken from the compost site, the lysimeter site, and the west side of the injection field. Time restrictions and wet field conditions prevented us from obtaining cores from the other sites. On May 30, 2006, we returned for the remaining samples. At this time, cores were collected from the lagoon site, the field near the state wells, and the east side of the injection field. The hormone concentrations from the soil cores from the injection field will serve as baseline levels and it will allow us to compare profile concentrations of 17ß-estradiol both before and immediately following the waste injection and to see whether there is any indication of it moving down the profile in the later sets of samples

On June 6, 2006 a second set of cores was taken from the injection field only. A handheld GPS unit was used to locate the approximate locations where the first four cores were taken. These were taken within three days after the field was injected with the lagoon slurry from the holding pond.

Corn had been planted on the injection field in early June and was beginning to sprout. In order to minimize disturbance of the farmer's crop, we waited to take the next set of cores. On June 14, 2006 another twelve cores were collected from the previously recorded GPS locations.

An end-of-season set of soil cores is planned to be collected from the injection field once the corn has been harvested. This is expected to be in either the last week of October or the first week of November.

Laboratory Methods

Back in the lab, core samples were removed from the freezer and the liners sliced open longitudinally. Sample lengths, soil horizonation, and other observations were noted at this time. Photos were taken of all cores. Next, each was divided into six-inch increments to facilitate pore water extraction. Each six-inch segment was weighed and mixed in a sealed plastic bag to promote homogeneity within each sample, especially with respect to water distribution. There were a total of 82 samples. A portion of each was placed in a moisture can and oven-dried overnight at 105°C to determine the residual moisture in each soil sample. Soil not needed for extractions was used to determine additional physical properties, such as density, organic matter content, and texture (using the hydrometer method of particle size analysis).

Separate pore water samples were sequentially extracted from each core segment. According to the devised procedure, 100 g of moist soil were mixed with 200 mL of 0.01M $CaCl_2$ solution in a 500 mL Erlenmeyer flask and placed on a shaker for 20 min. After shaking, the dispersed soil in the flasks were refrigerated and allowed to settle for

an hour. After an hour, the clearer liquid from the upper portion of the flask was decanted and filtered into narrow-mouthed 250 mL Nalgene bottles.

The purpose of the $CaCl_2$ was to enhance flocculation and to allow us to obtain a clear extraction in a reasonable amount of time. The clayey nature of the soil made it nearly impossible to filter the solution without the addition of the calcium solution. In test extractions, even after centrifuging and vacuum filtration, the fine particles clogged up the filter paper. The binding quality of the calcium allowed attainment of a relatively clear solution, using regular 18.5cm-diameter No. 2 filter paper and gravity filtration with a Buchner funnel.

At the time of this progress report, all samples are refrigerated and awaiting chemical analysis by LC-MS-MS, when the USDA lab is ready for them. This should be soon, as a new machine has recently been purchased to concentrate the water samples by automation, thus saving a substantial amount of “hands-on” time. The concentration of the water samples is necessary to measure hormone concentrations at the part-per-trillion level. The new machine will be installed and new procedural methods determined within in the first two weeks of November, 2006.

EXPECTED BENEFITS AND RESULTS

The data obtained from this study will allow the mechanisms of 17ß-estradiol and its metabolites to be studied more effectively. It will help to connect the ongoing field and laboratory research on this subject, and it will help to assess the effectiveness of the natural restorative processes in the environment.

The knowledge that is obtained will also help to further discern the factors that control the retention and transport 17ß-estradiol, and thus may be useful in the development of remediation or manure handling strategies.

Another important outcome is to quantify the amount 17ß-estradiol contributed to the environment, and whether this amount is a concern to human and animal health. This research will provide new information on 17ß-estradiol and will be valuable for risk evaluation of this estrogen as well as related hormones.

MOLECULAR PHYLOGEOGRAPHY OF *ETHEOSTOMA NIGRUM* (RAFINESQUE) IN THE UPPER MIDWEST

GRF Project 2006ND120B
Fellow: Ali Tackett
Adviser: Craig Stockwell
Department of Biological Sciences
North Dakota State University
Fargo, ND 58105

WATER PROBLEM AND OBJECTIVES

The geologic history and abundant potential study sites of the upper Midwest provide a unique opportunity for the assessment of spatial genetic diversity. The Johnny Darter, *Etheostoma nigrum* (Rafinesque), with its large range and abundant populations, is an excellent species to study to answer phylogeographic questions about North Dakota and Minnesota. In this study, the genetic diversity of *E. nigrum* will be examined by using microsatellite PCR primers designed initially for other species of *Etheostoma* and recently optimized for *E. nigrum*. This information will provide not only the inferred gene flow among the darters but will also provide a baseline against which to evaluate gene flow for other fish species located in the same water bodies. For instance, many game fish are stocked and transferred within and among watersheds with no genetic monitoring. By studying a benthic fish with a small home range, it will be possible to uncover the phylogeographic structure among the various watersheds of the upper Midwest. In turn, this information can be used by managers for conserving genetic diversity within and among watersheds.

This project uses molecular markers and their application in conservation ecology and as such the project spans the fields of ecology and genetics. More specifically, microsatellite markers will be used to examine the genetic relationships among Etheostoma nigrum (Johnny Dater) populations in the northern Midwest, specifically North Dakota and Minnesota.

PROJECT PROGRESS

During 2005, 406 individuals were captured and assayed from the Upper Missouri River, Red River of the North, and Upper Mississippi River watersheds. DNA extracted from fin clips taken from each fish is used in conjunction with the optimized microsatellite makers to examine genetic diversity within and among the sampled populations. The most recent data results from a comparison of four populations; one from each of the watersheds and one from a hydrologically isolated lake. Of the nine optimized primers six are polymorphic, one is monomorphic, one has a high frequency null allele, and the remaining locus requires more study. The preliminary data indicates that there is little gene flow between the basins of the upper Midwest. This is consistent with the low vagility of the species and glacial history of the region. Also, at this point, it appears that

sufficient diversity exists to map the landscape genetics for use as reference for translocated game fishes.

SIGNIFICANCE OF RESEARCH

The genetic evaluation of E. nigrum populations may have management implications. Most of the fish populations in the upper Midwest have been isolated since the end of the Pleistocene. Managers often transfer and stock game fish from one water body to another with little to no regard for the genetic structure of the systems. This practice is occasionally based upon the idea that gene flow will occur in systems that are hydrologically connected; but in actuality, gene flow is largely influenced by the migratory habits of individual species. Species with small home ranges may have little gene flow between closely located populations. Over time, these populations develop a unique genetic identity, often adapting to local conditions. When fish transfers are planned without consideration of this diversity, populations become genetically homogenous. This results in a loss of genetic variation among populations and perhaps even outbreedig depression. This is especially important if populations are locally adapted. Understanding the current diversity and gene flow of E. nigrum in the watersheds of the upper Midwest will aide in the establishment of management and conservation units as well as help managers plan for the transfer and stocking of fishes. E. nigrum are particularly useful for evaluating phylogeographic structure because they are non-migratory and unlikely to be accidentally transferred due to their use of benthic habitats. The optimization of published primers for use on E. nigrum will provide an excellent opportunity for not only this study but for future molecular work on Johnny Darters throughout their range. This project will provide a baseline with which other species and populations may be compared. Once the genetic relationships among this species are better understood and the diversity is used to help managers and scientists in this area, the information could potentially be used to manage congeners throughout the range of Etheostoma nigrum.

MODELING GROUNDWATER DENTRIFICATION BY FERROUS IRON USING PHREEQC

GRF Project 2003ND27B
Fellow: Tedros Tesfay
Adviser: Scott Korom
Department of Geology and Geological Engineering
University of North Dakota
Grand Forks, ND 58202

PROBLEM DESCRIPTION

Nitrate is one of the most common groundwater contaminants. Denitrification converts nitrate irreversibly into harmless nitrogen gas. It is a natural process that requires an anaerobic environment, denitrifying bacteria, and sufficient and reactive electron donating species. Numerous researchers show that the availability of electron donors within aquifer sediments limits the denitrification potential of aquifers. The three common electron donors for denitrification are organic carbon, sulfide (usually as pyrite), and ferrous iron. Reduced manganese may also contribute to denitrification, but it has never been shown to be a significant electron donor for denitrification in an aquifer. Our denitrification research team show organic carbon and sulfide are active electron donors for denitrification in North Dakota and Minnesota. We also believe ferrous iron is an active electron donor; however, the geochemical evidence for ferrous iron is more difficult to demonstrate and requires comprehensive knowledge of the hydrogeochemistry of the research sites.

SCOPE AND OBJECTIVES

Denitrification in aquifers involves numerous hydrogeochemical processes with both the water and sediment phases. Theses include dilution, ion exchange, dissolution, precipitation, and oxidation-reduction reactions. Knowledge of the above reactions will enable us to decipher the denitrification capacity of aquifers, particularly when ferrous iron minerals are involved. Therefore, our objective is to use PHREEQC in order to gain a more comprehensive understanding of the hydrogeochemical environment that governs denitrification by ferrous iron and associated aquifer reactions. This research complements the previous works by investigating the role of Fe(II) in the regional aquifer denitrification processes. Geochemical modeling, PHREEQC, is employed to gain insight into the in situ denitrification processes that take place via all major electron donors.

SUMMARY OF RESEARCH RESULTS

The major reasons that led to the ignorance of the role of Fe(II) in previous regional studies were two: 1) The fact that geochemical evidences for Fe(II)-supported denitrifcation is hard to comprehend and, 2) in the event where both inorganic carbon and Fe(III)-oxyhydroxides were precipitating, the role of Fe(II) was masked by that of the organic carbon. Therefore, two important measures were taken to tackle these problems.

First, the abundance of Fe(II) and the minerals that host it were determined using multiple complementary analytical techniques: wet chemical extractions, x-ray diffraction and Mössbauer spectroscopy. The results of these analyses confirmed that the sites where pyrite and organic carbon did not seem to be dominant are found to be relatively rich in ferrous iron minerals.

Then PHREEQC was used to resolve the intricacies between the two precipitating denitrification reaction products. First, PHREEQC simulated the amount of inorganic carbon precipitated out from solution indirectly through the co-precipitating Ca^{2+} and Mg^{2+} that were released into solution by cation exchange reactions. In some of the sites, Ca^{2+} and Mg^{2+} also decreased in solution. Therefore, computing the mass balance of Ca^{2+} and Mg^{2+} provided the maximum fraction of these cations lost from both the solid phase and solution. If all these cations were assumed to be co-precipitated together with the inorganic carbon, which is not likely, it provides the upper limit for the inorganic carbon that was possibly produced in the N-ISMs. By process of elimination the net nitrate lost due to denitrification, but not accounted for by reactions with pyrite and organic carbon, was attributed to Fe(II) and substantiated by the subsequent evolution on the water in the N-ISMs.

Validation of the modeling work by comparing output files with the target solutions of different time steps demonstrated that dilution, CEC, and reversible reactions were apparently responsible for the geochemical evolution observed in the C-ISMs. Whereas for the N-ISMs, in addition to dilution, CEC, and reversible reactions, denitrification reactions involving FeS_2, CH_2O, and Fe(II)-amphibole were the main processes influencing the geochemical environment of the N-ISMs. Therefore, all aqueous analytical data, mineralogy and chemistry of sediments and geochemical modeling works are evidently showing the proportional role of the common electron donors (Fig. 17) and Fe(II)-supported denitrification has a significant role as a natural remediation process.
Moreover, observation of the hydrochemical data of the ISMs also demonstrated that denitrification rates were higher for those sites with higher concentrations of electron donors and vice versa.

CLASSIFICATION OF MACROINVERTEBRATE COMMUNITIES ACROSS RED RIVER DRAINAGE BASIN

GRF Project 2006ND124B
Fellow: Wei Zheng
Adviser: Malcolm G. Butler
Department of Biological Sciences
North Dakota State University
Fargo, ND 58105

Research Problem

Aquatic macroinvertebrates living in large rivers generally use different habitats and respond differently to the hydrological regime. As one of the major rivers in western United States, the Red River of the North provides multiple uses for local people. However, little is known about the macroinvertebrate fauna of the Red, largely because rivers of this size and type are difficult to sample. Research on macroinvertebrate communities in major habitats of the main-stem Red River is needed to determine what types of animals predominate in the various habitats within this river. Identification of ecologically significant taxa (in term of abundance or functional importance) in the river will aid in selection of candidate indicators for future biomonitoring, a need increasingly recognized by state and national management agencies. Also needed is assessment of sampling techniques that can efficiently monitor those taxa that may be significant indicators of the river's ecological function and condition.

Scope and Objectives

The objectives of this research are to 1). Inventory the macroinvertebrate communities of different habitat conditions in the main stem Red River 2). Explore the temporal changes of these benthic communities through the open water season and 3). Compare the results of outcomes provided by different sampling techniques. These objectives will allow me to evaluate potential relations between aquatic invertebrate communities and environmental conditions.

Methods

Sites will be randomly selected in three reaches of the main stem Red River: above Fargo (Oxbow), within Fargo (Oak Grove), and below Fargo (Harwood). Sampling periods will be chosen from May to September, during which different sampling techniques will be applied to each site according to habitat: main-channel using a Ponar sampler, channel slopes using a core sampler, and sweep net to collect from snags. If possible, drift nets will be deployed during the night time, and retrieved them the next day. Site variables will be measured on each sampling occasion using meters, water samplers, direct observations or visual estimations. Water quality samples will be taken back and analyzed by chemistry laboratory. Macrobenthic invertebrate samples will be preserved

in 70% ethanol and separated from river sediments using a surcrose-floatation procedure. Organisms will be identified to the lowest practicable level by a stereoscope and with appropriate taxonomic keys. The identification results will be compared and complied with published and known records.

Deliverables

The macroinvertebrate faunas of large, rivers throughout the world have been poorly studied by comparison to the vast literature on smaller, upland streams. There has been a handful of studies on large rivers around the world, including rivers in Australia, Austria, France, North America, Spain, and Russia. Very little research has been done to benthic marcroinvetebrates in the Red River. Ultimately, this work will be available for use in developing large scale international monitoring assessment of Red River, and advance our understanding of aquatic ecology to the benefit of biomonitoring and water resources in North Dakota and Minnesota.

Progress of Work

Drift net sampling was conducted from May 27, 2006 to November 4, 2006. Nets were deployed from bridges over the main-stem Red River at three different locations (within Fargo, an interval of two weeks. We are now starting to identify and quantify the macroinvertebrates from the drift samples. Infaunal benthos was sampled with a Ponar grab at five cross-channel transects at different sites along the river. We also deployed artificial substrates (standard Hester-Dendy samplers plus a custom-built ash log substrate design) for 5 weeks beginning Sept.1, 2006.

Preliminary results from the drift nets, indicate that drift densities reached a maximum during summer, peaking in late August. Community composition differed between sampling sites and over time. Dominant taxa over the season included chironomid larvae, elmid beetle larvae, adults of several aquatic beetle families, as well as terrestrial Coleoptera. Chironomidae displayed the highest drift rates from August to early September, when other taxa were relatively sparse in the drift. The burrowing mayfly *Ephoron* (Polymitarcidae) had a notable emergence from mid-July to early August. Several drifting taxa (Chironomidae) were significantly negatively related to discharge, but not to other hydrological parameters. Seasonal variability in drift likely results from the life histories of individual species, as well as fluctuations in discharge. Terrestrial insects made a substantial contribution to invertebrate drift in this river reach.

ASSESSING THE EFFECTIVENESS OF LOCAL WATER INSTITUTIONS IN WATER MANAGEMENT

GRF Project 2005ND86G (104-G)
PI: Robert Hearne
Department of Agricultural Economics
North Dakota State University
Fargo, ND 58105

Abstract

There are a variety of formal and informal local institutions that are involved with water resources management in rural areas. These governmental and non-governmental institutions have different objectives, different legal statuses, and different affiliations with state and local governments. Research is needed to assess the roles and effectiveness of local water institutions. As new initiatives to improve water quality are being proposed, it is important to assess the capacity of existing institutions to meet new and evolving needs. The objective of this research is to improve local management of water resources by providing policy makers and agencies with an improved understanding of the characteristics of successful local institutions. This research will focus on the Red River of the North basin in Minnesota and North Dakota although some assessment of Manitobas institutions will be included. The basin is fairly homogeneous in terms of land use and geographic features, but features three completely different sets of water law, which makes it an excellent case study of institutions. The overall objective of this research is to strengthen local water management institutions so that they may better meet evolving local and basin wide needs, especially the maintenance of water quality. Specific objectives of the research include: 1) Develop a set of objective and subjective criteria and indicators to evaluate local water management institutions; 2) Provide a review of the different governmental and nongovernmental institutions in the basin, classify their goals, activities and chartered purposes, and identify overlaps and functions that are not being addressed; 3) Identify and evaluate the characteristics of local water institutions that have a demonstrated capability to meet local goals and wider goals of the greater river basin; 4) Assess the use of: scientific and technical information; extension education and training programs; and other support provided by governmental and non-governmental agencies; 5) Analyze institutions and agencies likely behavior in a decision-making situation and further develop decision-making support tools; 6) Identify the characteristics of institutions that successfully evolve to meet new challenges; 7) Analyze preferences of a sample of residents and stakeholders toward watershed management issues and the types of institutions that they trust; and 8) Disseminate results to various forums including local workshops and scientific journals. Objective and subjective criteria and indicators for local water institutions will be refined for local circumstances by interviewing and surveying assorted State and Federal agencies who work on water management issues. A survey of local water institutions will be used to: identify goals, activities, and accomplishments; assess their understanding and use of technical information and extension training; and provide an understanding of how these institutions have evolved to meet changing needs. This survey will be supported by another survey of local leaders, county commissioners, and mayors. The Legal-Institutional Analysis Model will be used to assess negotiation strategies. And choice experiments, a stated preference technique that can estimate the non-market value for environmental goods and services, will be used to analyze residents and leaders preferences towards water management programs and institutional frameworks.

Benefits of the Research

By identifying the attributes of local institutions that effectively achieve their own goals and/or further goals of water quality maintenance, this research will:1) ascertain whether existing institutional frameworks should be adapted to meet evolving needs or new institutions should be developed to address emerging issues such as water quality monitoring and enforcement; 2) support local institutions by identifying key characteristics that facilitate effectiveness; 3) assess the benefits and costs associated with having water resource institutions defined along county lines as opposed to watershed lines; 4) support the development of extension and education programs that strengthen local institutions by specifically addressing key characteristics of effectiveness; and 5) help policy makers in the design strategies to monitor and enforce nonpoint source pollution abatement initiatives. Based upon this research and subsequent reviews and comments, recommendations will be made to political leaders and lawmakers, agency officials, and local stakeholders.

This project will collaborate with the North Dakota State University's (NDSU) M.S. programs in Agribusiness and Applied Economics, and Natural Resources Management and is expected to support at least two M.S. students. It should produce at least two M.S. theses, a number of extension reports, at least two peer-reviewed scientific journal articles, and at least one workshop to present results to regional and local water management leaders.

Planned First and Second-Year Goals

During the first year period of September 2005 – August 2006 planned goals were to:

1) Develop a set of objective and subjective criteria and indicators to evaluate the effectiveness of local water management institutions;
2) Review the different governmental and nongovernmental institutions in the basin, classify their current goals and activities as well as their chartered purposes, and identify overlaps as well as functions that are not being addressed; and
3) Identify and evaluate the characteristics of local water institutions that have a demonstrated capability to meet local goals and wider goals of the greater river basin, including water quality monitoring and participation in the establishing TMDLs.

During the period of September 2006- August 2007 planned goals included:

4) Assess the use of: 1) scientific and technical information provided by USGS and other agencies; 2) extension education and training programs; and 3) other support provided by governmental and non-governmental agencies and organizations;
5) Analyze institutions' and agencies' likely behavior in a decision-making situation, such as watershed and basin planning, or conflict resolution and further develop decision-making support tools;
6) Identify the characteristics of institutions that successfully evolve to meet new challenges; and
7) Analyze preferences of a sample of residents and stakeholders toward watershed management issues and the types of institutions they trust.

Progress towards Goals

A review of water management organizations and institutions has been conducted.

A set of criteria and indicators for effective public water management organizations. These criteria and indicators were used to develop a survey instrument 2006. A surveys of organization managers and board members was conducted in December 2006 – January 2007. Survey data is currently being analyzed. This analysis will: i) identify and evaluate the characteristics of local water institutions that have a demonstrated capability to meet local goals and wider goals of the greater river basin; ii) assess the use of scientific and technical information, extension education and training programs, and other support provided by governmental and non-governmental agencies and organizations; and iii) identify the characteristics of institutions that successfully evolve to meet new challenges. A MS thesis from this work will be presented and defended in June or July 2007.

An analysis of institutions' and agencies' likely behavior in a decision-making situation has not been conducted. This objective was to have been completed with the support of USGS personnel from Ft. Collins Research center. However, given that grant funds can not be used for USGS travel, we have not been able to complete this task. Currently there are no plans to complete this activity.

A second survey of informed stakeholders will be conducted in late 2007. Currently the population to be sampled is being identified and initial consultations to support the development of the survey instrument are being conducted.

A further analysis of measures of "effectiveness" is being initiated. The results of this analysis should be used with the data on organizational activities and attributes.

COLLABORATIVE RESEARCH ON IN SITU DENITRIFICATION AND GLYPHOSATE TRANSFORMATION IN GROUND WATER: NAWQA EASTERN IOWA BASINS STUDY UNIT

GRF Project 2006ND126G (104-G)
PI: Scott Korom
Co-PI: Capel, Paul D. (USGS)
Department of Geology and Geological Engineering
University of North Dakota
Grand Forks, ND 58202

RESEARCH PROBLEM

Contamination of ground water by nitrate and pesticides is widespread in some areas of the country and can threaten drinking water supplies. It is well known that the most important removal mechanism of nitrate and most pesticides from ground water is biodegradation, but the in situ transformation rates are largely unknown. In this study, two 180-L stainless steel chambers forming in situ mesocosms (ISMs) of aquifer sediments will be installed below the water table at the NAWQA agricultural chemicals study sites in the glaciated part of Iowa. This work will extend the use of this technique to examine denitrification in an area characterized by high dissolved iron concentrations and to measure the transformation rate of the extensively-used herbicide, glyphosate.

OBJECTIVES

The objectives for the research are:

1. Measure the denitrification and glyphosate transformation rates in the two ISMs.
2. Determine whether the denitrification is better fit by zero-order or first-order reaction rates.
3. Determine what donors are contributing electrons for the denitrification and their relative amounts.
4. Incorporate the results of the two ISMs into the existing databank of nine other ISM sites in glacial outwash aquifers in the Upper Midwest.
5. Update the available data of the apparent isotopic enrichment factor for 15N in nitrate versus denitrification rate among of ISM sites.
6. Update the nitrate vulnerability index and extrapolate the findings to similar, unmonitored agricultural and environmental settings.

Aquifer sediment samples will be collected from the Iowa site and analyzed for grain-size distributions, mineralogy, and major e- donors (organic carbon, sulfide, and ferrous iron) to determine optimum locations for installation of the ISMs, provide insights on the types and heterogeneity of e- donors at the site, and provide the e- donor supply data at the Iowa site that can be compared to previous ISM studies in the Upper Midwest. After the

ISM chambers are installed, they will be purged and then amended with nitrate and bromide, which serves as a tracer for nitrate. Any loss of nitrate beyond that explained by dilution of the bromide tracer is attributed to denitrification. The ISMs will be sampled over time (months) and the decreases in nitrate concentrations will be used to calculate rates of denitrification. Modeling of the evolution of the geochemistry in the ISMs will provide insights into what donors contributed electrons to the denitrification and their relative amounts. The field experiment will be repeated a second time; however, in addition to nitrate and bromide, glyphosate will be added. Denitrification information from the second test will provide insights into the variability of the results from the first test. More importantly, the attenuation and transformation of glyphosate (with the dominate metabolite, AMPA) will be studied in both the presence and absence of nitrate to determine the fate of glyphosate in oxidizing and reducing conditions. The results of this study will provide site-specific transformation rates for nitrate and glyphosate and extend the aquifer nitrate vulnerability index that was developed in earlier studies. This information is vital for the development of tools and quantitative methods to characterize the transport and fate of agricultural chemicals in the Eastern Iowa Basins Study Unit, the Upper Midwest, and beyond.