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**Kentucky Water Resources Research Institute
Annual Technical Report
FY 2011**

Introduction

The FY2011 Annual Technical Report for Kentucky consolidates reporting requirements for the Section 104(b) base grant award into a single document that includes; 1) a synopsis of each research project that was conducted during the period, 2) citations for related publications, reports, and presentations, 3) a description of information transfer activities, 4) a summary of student support during the reporting period, and 5) notable awards and achievements during the year.

Project 2011KY166B (Sewage to fertilizer: a proposed solution to water pollution) was not initiated because the principal investigator (Dr. Rebecca Evans Kelley) resigned from the Department of Biological Sciences at Northern Kentucky University between the time the application was originally submitted and when funding was eventually made available through the program to conduct the project. As a result, the project was cancelled, no research was conducted, and no report for this project is provided.

Project 2011KY213B (Use of gene expression in longear sunfish (*Lepomis megalotis*) and green sunfish (*Lepomis cyanellus*) as a biomarker of polychlorinated biphenyl and metal exposure) was delayed in starting. As a result, no significant progress was completed during FY 2011. The project report will be included in the FY 2012 Annual Technical Report.

Research Program Introduction

The activities supported by the Section 104(b) program funds and required matching are interwoven into the overall program of the Kentucky Water Resources Research Institute. Additional research, service, and technology transfer activities were funded through a variety of other sponsors. Memoranda of Agreement projects with the Kentucky Division of Water included Total Maximum Daily Load development for several Kentucky streams. The Kentucky River Authority supported watershed management services in the Kentucky River basin and a small local grant program to fund local grassroots organizations. The National Institute of Environmental Health Sciences supported research translation activities through the Superfund Public Outreach Program. The Kentucky Department for Environmental Protection supported 2 students through an Environmental Protection Scholarship Program coordinated by the Institute. The Division for Compliance Assistance funded support for stormwater BMP demonstration projects in 3 Phase II communities and provided registration support for all MS4 communities to send a representative to the annual meeting of the Kentucky Stormwater Association. The Institute also participated in several projects supported by the Department for Hometown Security.

The Kentucky Consortium for Energy and Environment, established by Lindell Ormsbee (Director of KWRRI), continued a collaborative program integrating faculty and students from Kentucky universities. The consortium was funded through the US Department of Energy to assist with efforts supporting a variety of environmental assessment and cleanup activities at the Paducah Gaseous Diffusion Plant. The main project coordinated by the KWRRI during 2011 involved gathering input from stakeholders regarding potential future uses of the property associated with the plant after the plant is shut down. During 2011, administration of the Consortium was transferred to the Center for Applied Energy Research at the University of Kentucky.

Eleven student research enhancement projects were initially selected for support through 104(b) FY2011 funding. One project (2011KY166B) was not initiated because the principal investigator (Rebecca Evans Kelley) resigned from Northern Kentucky University between the time the application was originally submitted and funding was ultimately made available through the program. An additional project (2011KY167B) was revised and resubmitted with a new investigator team through a different lead institution in early 2012. Due to the extremely late start of this revised effort, no significant progress can be reported and so the project proposal is provided as a part of this annual report rather than a status report (identified as 2011KY213B).

Projects were conducted at the University of Kentucky (6), Morehead State University (1) in collaboration with Asbury University, Kentucky State University (1), Murray State University (1), and Western Kentucky University (1). Projects represented a variety of discipline areas including civil engineering (2), biology (3), plant and soil science (2), geology (1), and chemistry (2). The goal of this approach is to support a number of student-based efforts representing a variety of discipline areas at numerous educational institutions throughout the state to develop broad research capacity related to water resources. Many state agencies are experiencing a significant loss of personnel through retirement and agency staff reductions and it is critical that undergraduate and graduate students are well trained and available to help fill these voids.

Reports for the 10 student research enhancement projects follow. Due to delayed initiation of funding, all projects have been provided with extensions through 12/31/2012. Progress on most of the projects was not sufficient to allow for presentations at the 2012 Kentucky Water Resources Annual Symposium, but all should be completed in time to provide project reports at the 2013 symposium.

Effects of streambed sediments on the fate of selenium in eastern Kentucky watersheds contaminated with surface coal mining operations

Basic Information

Title:	Effects of streambed sediments on the fate of selenium in eastern Kentucky watersheds contaminated with surface coal mining operations
Project Number:	2011KY165B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Sediments, Water Quality
Descriptors:	pollutant transport, geochemistry, adsorption, transport
Principal Investigators:	Yi-Tin Wang

Publications

There are no publications.

Effects of Streambed Sediments on the Fate of Selenium in Eastern Kentucky Watersheds Contaminated with Surface Coal Mining Operations

Problem and Research Objectives

Selenium is found naturally in the earth's crust. The main natural sources of selenium in the environment are the weathering of rocks, soils, and minerals, as well as volcanic activity. Anthropogenic sources of selenium are widespread including uses in electronics and photography, glass manufacturing, pigments, additives for metal processing, copper refining operations, fossil fuel combustion, petroleum refining, and agricultural drainage waters in the western United States. In eastern Kentucky, surface-mining operations in Appalachian coal regions have been identified as the major cause of selenium contamination in water and fish, due to the occurrence of selenium in some overburden soils exposed during mining activities.

Exposure to very high levels of selenium can cause dizziness, fatigue, irritation, collection of fluid in the lungs, and severe bronchitis. The U.S.EPA has set the maximum contaminant level for selenium in drinking water at 0.05 mg/L to protect the public.

The overall objective of the study is to obtain a better understanding of the interactions between streambed sediments and selenium in contaminated watersheds. The specific aims are:

1. To investigate the potential for microbial communities found in the streambed sediments to reduce selenium in watersheds contaminated by coal mining spoil deposits.
2. To isolate and identify selenium-reducing bacteria from streambed sediments.
3. To determine environmental factors affecting microbial reduction of selenium.
4. To evaluate abiotic mechanisms affecting the interactions between streambed sediments and selenium in the liquid phase.

Methodology

Streambed sediment (50 g) collected from the coal field is suspended in 150 mL of river water in the laboratory in 250-mL Erlenmeyer flasks containing nutrient broth (0.5 percent) and glucose (0.1 percent) to stimulate microbial growth. After selenium compounds (5 mg/L sodium selenate and sodium selenite) are added, the flasks are sealed with sterile rubber stoppers and incubated statically at 30° C for at least a week (or when a red color of Se (0) appears). The headspace gases of the flasks are analyzed for volatile selenium compounds by a gas chromatograph equipped with a flame-ionization detector. Selenate and selenite are determined using a colorimetric method according to Section 3500D of the Standard Methods for the Examination of Water and Wastewater (APHA, 1995). The colorimetric method is very sensitive, less costly than hydride generation

atomic absorption spectrometry, and can analyze both selenite (IV) and selenate (VI) in the sample. Autoclaved biological controls are run to account for any losses through abiotic mechanisms such as adsorption or chemical oxidation/precipitation.

The enrichment procedure is conducted using 50-mL Erlenmeyer flasks containing 35 mL nutrient broth and 5 g of sediment. The flasks are spiked with sterile sodium selenate or sodium selenite to a final concentration of 50 mg/L, capped with sterile rubber stoppers and incubated statically at 30° C for 5 days (or when a red color of Se(0) appears). The contents of the flask with red color are then serially diluted and spread on Se(VI) and Se(IV)-containing tryptic soy agar (TSA) plates respectively and incubated at 30° C for 5 days (or when red Se(0) precipitates are observed). The single colonies are restreaked on TSA plates with and without Se (VI) or Se (IV) to ensure that the red color is the result of microbial reduction of Se. The Se-reducing strain is identified using a modified method of Lane (1991) by a partial sequencing of 16S rDNA. The Se (-II)-forming cultures are obtained using the same procedure except that methylated selenides in the headspace of the flasks are monitored.

Batch culture is used to evaluate the effect of the following variables on relative rates of Se(VI) and Se(IV) reduction: concentrations of growth substrate, Se(VI), Se(IV), dissolved oxygen, temperature, pH, and bacterial species concentration. The growth substrate is selected based on the cultures identified. Se(VI) and Se(IV) concentrations are evaluated from 2 µg/L to 10 mg/L and temperature investigated from 5°C to 40° C with pH varied from 4 to 9. Dissolved oxygen levels are investigated from 0 to 9 mg/L.

Principal Findings and Significance

Field sampling of surface water sediments was completed throughout various locations in Perry County, Kentucky. A total of nine soil sediments were collected and stored on ice on June 22 and June 23, 2011 with two of the strains isolated producing apparent selenium reducing results as indicated by the red color in anaerobic batch reactors. The sampling pool consisted of one sediment sample from the Frasure Creek Mining Site on Starfire Haul Road taken approximately 100 ft down gradient from a strip mine reclamation area. The second sample was taken further down gradient on the Frasure Creek Mining Site in a small drainage pond containing coal slurry run off. The bacteria strain (FC-2) isolated from this site produced the best ability to reduce aqueous selenium, thus laboratory studies are focusing primarily on this strain. Figure 1 shows the preliminary results of anaerobic selenium reduction by this yet to be identified bacterium.

Two more sediment samples were taken from a creek near the Knott-Perry County line on Deadline Branch Rd. Another two sediment samples were taken from an Enterprise Mining Site approximately 600 yards down gradient from a sediment pond near Sassafras Creek just off of road 1088. The final three sediment grab samples were taken in the Robinson Forest Valley Fill (only one resulted in the isolation of a selenium reducing bacterial strain).

The project has been extended through December 2012. There are six major tasks as shown below. Task 1 has been completed and Task 2 is about to be completed (a selenium-reducing bacterium (FC-2) has been isolated but not yet identified. For the remainder of the project period, we will identify the selenium-reducing bacterium (Task 2), determine environmental factors affecting its reduction of selenium in batch reactors (Task 3) and evaluate abiotic mechanisms affecting the interactions between streambed sediments and selenium in the liquid phase (Task 4).

Task:

1. Investigate the potential for microbial communities found in the streambed sediments to reduce selenium in watersheds contaminated by coal deposits.
2. Isolate and identify selenium-reducing bacteria from streambed sediments.
3. Determine environment factors affecting microbial reduction of selenium.
4. Evaluate abiotic mechanisms affecting the interactions between streambed sediments and selenium in the liquid phase.
5. Data analysis
6. Final report

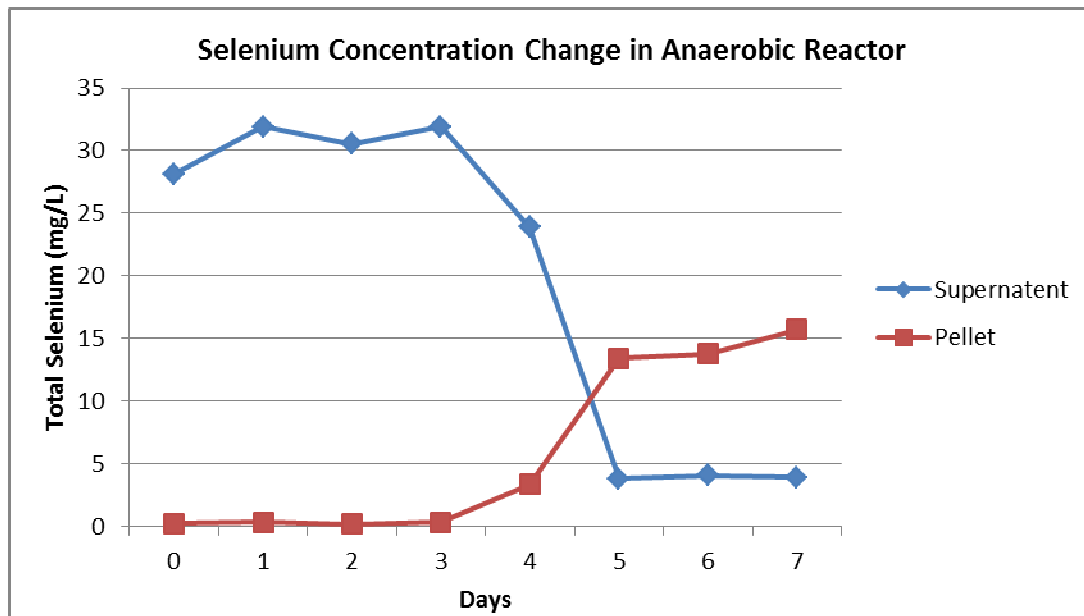


Figure 1: Selenium reduction in an anaerobic batch reactor by bacterium FC-2.

Sewage to fertilizer: a proposed solution to water pollution

Basic Information

Title:	Sewage to fertilizer: a proposed solution to water pollution
Project Number:	2011KY166B
Start Date:	3/1/2011
End Date:	2/29/2012
Funding Source:	104B
Congressional District:	KY 4th
Research Category:	Water Quality
Focus Category:	Nutrients, Nitrate Contamination, Surface Water
Descriptors:	
Principal Investigators:	Rebecca Kelley

Publications

There are no publications.

Sewage to Fertilizer: A Proposed Solution to Water Pollution

Project 2011KY166B was not initiated because the principal investigator (Dr. Rebecca Evans Kelley) resigned from the Department of Biological Sciences at Northern Kentucky University between the time that the application was originally submitted and when funding was eventually made available through the program to conduct the project. As a result, the project was cancelled, no research was conducted, and no report for this project is provided.

Using the stability of soil moisture distribution to understand soil system modularity and complexity at the landscape scale

Basic Information

Title:	Using the stability of soil moisture distribution to understand soil system modularity and complexity at the landscape scale
Project Number:	2011KY168B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Ground-water Flow and Transport
Focus Category:	Groundwater, Hydrology, Water Quantity
Descriptors:	geophysics, electrical resistivity, ER, vadose zone
Principal Investigators:	Ole Wendroth, Junfeng Zhu

Publications

There are no publications.

Using the Stability of Soil Moisture Distribution to Understand Soil System Modularity and Complexity at the Landscape Scale

Problem and Research Objective

Detailing the spatio-temporal organization of soil moisture can provide insight into the underlying properties and processes controlling soil moisture variability (Lin, 2006). The spatial organization of soil moisture can exhibit temporal persistence, where certain locations are characteristically higher, lower, or equal to the field average (Lin, 2006; Martinez-Fernandez et al., 2003; Vachaud, et al., 1985; Zhao, et al. 2010; Zhou, et al., 2007). Temporal stability analysis was first introduced by Vachaud et al. (1985) as a statistical tool to characterize the time invariant association of a sampling location relative to the field average or relative rank. This measure can identify sample locations that maintain their statistical relevance through time. These locations require less ambitious sampling schemes and thereby save time, labor, and cost associated with field efforts.

Accurately capturing and mapping the spatio-temporal distribution of soil moisture is challenging. Point observations, such as soil coring and *in situ* monitoring, are commonly employed to measure the spatio-temporal variability of soil properties. Due to their invasiveness, labor requirements, and cost limitations, these methods are not well suited for providing a comprehensive measure of soil variability. Therefore, the measured variability is often a consequence of sampling coverage (i.e. grid spacing and number of samples). For these reasons, auxiliary methods of capturing and mapping the spatio-temporal variability of soil moisture are rapidly expanding within the soil science community (Robinson et al., 2012). Most prominent of these methods is geophysics. Geophysical technologies are attractive because they are sensitive to soil moisture and are relatively economic, non-invasive, and perform rapid, reiterative, high resolution data acquisition (Amidu, et al., 2008; Robinson et al., 2012).

The objective of this research is to assess the temporal stability characteristics of soil water content and geoelectrical resistivity as an integrative soil state variable at five different landscape positions located along approximately a 400 meter hillslope transect. The results of electrical resistivity measurements are presented in this status report.

Methodology

The area investigated is a 410 meter hillslope transect located at Spindletop Farm, Fayette County, Lexington KY (38.116030 N, 84.491093 W). The area is dominated by forage grasses and underlain by Ordovician phosphatic limestones, calcareous shales, and interbedded limestone and shales. The soils are non-saline and typified by an argillic subsurface horizon. Soil depths vary according to landscape position ranging from 40 – 200+ cm.

Electrical resistivity (ER) soundings were completed using a SuperSting R8/IP (Advanced Geosciences Inc., USA). Five locations along the 410m transect were

investigated. Table 1 documents the relative landscape position of each ER survey. Each ER survey consisted of 84 collinear electrodes spaced .5 meter apart spanning a total of 41.5 meters. A dipole-dipole array was used to measure the apparent resistivity of the shallow subsurface (effective depth of penetration was ~ 9 meters). Resistivity sampling was scheduled every two weeks starting late September and ending mid-November; a total of four sampling events were completed during this duration. Apparent ER values were inverted using RES2DINV software. Inverted ER values were utilized for subsequent statistical analysis.

Capacitance readings were collected in tandem with ER soundings to assess volumetric water content in 10 cm intervals spanning from the soil surface to a 1 meter depth. Electrical resistivity lines were offset approximately 1 meter from the capacitance probe access tubes. Future work will entail performing correlation analysis between inverted ER readings and Diviner readings.

A first order soil survey was performed in fall 2011 with the Kentucky NRCS and Dr. Karathanasis (University of Kentucky, Soil Morphology Course) along the hillslope transect investigated. A total of 12 soil cores were collected using a Giddings hydraulic auger. Tile probing was used to determine soil depth. The soil cores were characterized to establish soil mapping boundaries and representative pedons along the transect studied.

Time stability analysis has been used extensively to characterize the temporal behavior of soil moisture distribution patterns (Lin, 2006; Martinez-Fernandez, et al. 2003; Vachaud, et al., 1985; Zhou et al., 2007). Over a given observation domain, certain areas will exhibit persistent patterns of wetness or dryness in comparison to the average moisture content across the observation domain (Lin, 2006). This persistence can be assessed using time stability (Vachaud, et al., 1984). The first step is to calculate the relative difference of sampled values [Eq. 1] (Vachaud et al., 1984):

$$\delta_{ij} = \frac{\Delta_{ij}}{\bar{S}_j} \quad [\text{Eq. 1}]$$

where Δ_{ij} is the difference between an individual measurement (S_{ij}) at location i and time j and the field mean at the same time (\bar{S}_j) [Eqs. 2 & 3] (Vachaud et al., 1985):

$$\Delta_{ij} = S_{ij} - \bar{S}_j \quad [\text{Eq. 2}]$$

and

$$\bar{S}_j = \frac{1}{n} \sum_{i=1}^n S_{ij} \quad [\text{Eq. 3}]$$

where n is the number of sampling locations. The mean relative difference for each sampling location is defined by [Eq. 4] (Vachaud et al., 1985):

$$\bar{\delta}_i = \frac{1}{m} \sum_{j=1}^m \delta_{ij} \quad [\text{Eq. 4}]$$

where m is the number of days sampled.

The mean relative difference (MRD) was calculated for each inversion depth (0, 0.13, 0.39, 0.67, 0.99, 1.3, 1.7, 2.1, and 2.6 meters) along the entire transect studied and the values were subsequently kriged in ArcMap 10 for visual analysis.

Principal Findings and Significance

The inverted resistivity data provided useful information in detailing soil morphological characteristics. There is a strong association between soil mapping descriptions and the inverted ER data in identifying the A and A/B (i.e. transition) horizons, argillic subsurface horizon, and soil depth. Results show these features are unique according to landscape position.

Figure 1 illustrates preliminary results for the mean relative difference of the nine inversion depths observed. The sign preceding the MRD values indicates locations that are persistently above or below the average for each respective depth. Extreme positive (above 1) MRD values exhibited deeper in the profile are anticipated to be relics of underlying parent material and/or bedrock. Preliminary analysis suggests that Sites 1 & 2 provide the best representation of the field average. Regions with negative MRDs are anticipated to have higher clay and soil moisture contents. Regions with positive MRDs are anticipated to be well drained and have lower bulk density (i.e. less compacted). Onsite investigations concur with this assessment, particularly for the surface soils at Site 3. The time stability results presented herein are largely related to soil properties related to electrical resistivity including soil texture, bulk density, degree of saturation, salinity and temperature. These combined properties tend to mask the effects specific to soil moisture. Therefore, future data manipulation will attempt to distill time stability characteristics specific to soil moisture using time-differencing.

Preliminary findings support the use of geoelectric techniques as a site reconnaissance tool to obtain general information about the heterogeneity of subsurface soil properties. This provides beneficial insight into soil properties that ultimately influence the spatial distribution of soil moisture and therefore allow field investigators to devise more representative sampling schemes to study soil hydrologic phenomena. The significance of the preliminary time stability analysis suggests that different landscape positions are unique with respect to their spatial behavior and cannot necessarily be regarded equally according to landscape position.

Future work will entail a more detailed discussion and presentation of inverted electrical resistivity data for each landscape position. Future temporal stability analysis will be performed on time-differenced inverted ER data to detail time stability characteristics of non-static soil properties (i.e. soil moisture) sensitive to electrical resistivity measures. Standard deviations of the mean relative differences will be included. Correlation analysis will be performed to reveal the association between capacitance and inverted resistivity values.

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Zhao, Y., S. Peth, X.Y. Wang, H. Lin and R. Horn (2010). Controls of Surface Soil Moisture Spatial Patterns and their Temporal Stability in a Semi-Arid Steppe. Water Resource Management **24**: 2247-2266.

Zhou, X., H. Lin, and Q. Zhu (2007). Temporal Stability of Soil Moisture Spatial Variability at Two Scales and Its Implications for Optimal Field Monitoring. Hydrology and Earth System Sciences Discussions **4**: 1185-1214.

TABLE 1	
Location	Landscape Position
Site 1	Shoulder (Upland), Convex, 2-6% Slopes
Site 2	Sideslope (Headslope) transitions into Drainageway (Depression), Concave, 6-12% Slopes
Site 3	Sideslope (Noseslope), Convex, 2-6% Slopes
Site 4	Sideslope (Noseslope), Convex, 2-6% Slopes

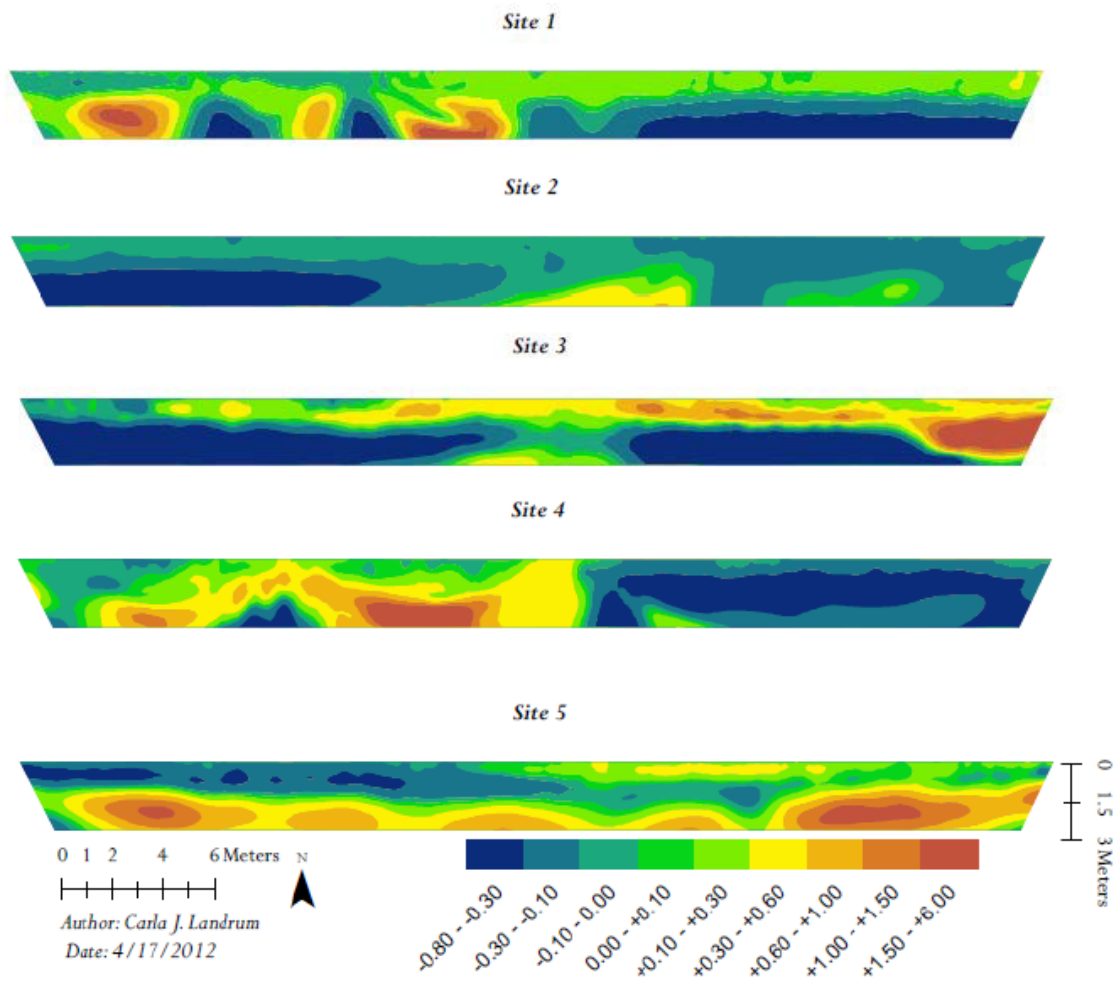


Figure 1. Mean Relative Difference (MRD) of ER soundings for each landscape position.

Delineating solute inputs to the headwaters portion of the Cane Run/Royal Spring basin

Basic Information

Title:	Delineating solute inputs to the headwaters portion of the Cane Run/Royal Spring basin
Project Number:	2011KY169B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Ground-water Flow and Transport
Focus Category:	Groundwater, Solute Transport, Water Quality
Descriptors:	karst, spring, NPS pollution, nutrients
Principal Investigators:	Alan Fryar, James Dinger

Publications

1. Skees, Catherine F. and Alan E. Fryar, 2012, Delineation of solute inputs to the headwaters portion of the Cane Run/Royal Spring basin, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, University of Kentucky, Lexington, KY, p. 83.
2. Skees, Catherine F. and Alan E. Fryar, 2012, Using geochemical analyses to delineate solute inputs to the headwaters portion of the Cane Run/Royal Spring basin of north central Kentucky, Geological Society of America Abstracts with Programs, Boulder, CO, vol. 44, no. 5.

Delineating Solute Inputs to the Headwaters Portion of the Cane Run/Royal Spring Basin

Problem and Research Objectives

The Cane Run watershed and underlying karst aquifer, which discharges to Royal Spring, serve as the primary source of drinking water for the city of Georgetown, Kentucky. The headwaters of this basin are located in north Lexington, an urban area with mixed industrial and residential land uses. The water quality of the basin has been compromised by non-point-source pollutants, including bacteria, sediment, and nutrients. The Lexington-Fayette Urban County Government (LFUCG) signed a consent decree with the U.S. Environmental Protection Agency to address combined sanitary-storm sewer overflows in 2008. Other potential sources of pollution include the University of Kentucky (UK) Spindletop Farm, the Kentucky Horse Park, private farms, and a trailer park. Current monitoring is limited to bacteria at several sites, plus analyses of Royal Spring water by the Georgetown Municipal Water and Sewer Service for constituents regulated under the Safe Drinking Water Act. General variations in groundwater quality throughout the basin have not been thoroughly investigated. Delineating solute inputs would aid in the development of remedial and future water-quality monitoring efforts.

During one year, we are collecting water samples for analyses of major anions, nutrients (nitrate, nitrite, and ammonium), stable isotopes, and field parameters (temperature, specific conductance, pH, and dissolved oxygen) within the urbanized headwaters. Such data can theoretically help identify areas of ground water/ surface water interaction and solute inputs, including nutrient hotspots. Understanding the processes affecting water quality within the Cane Run headwaters will enhance community and governmental efforts to protect this and similar karst aquifers.

Methodology

Nutrients, other anions, and stable isotopes of water are being monitored biweekly at seven sampling sites. These include two springs (the Citation Estavelle and Highland Spring) and five sites along surface drainages (CR01, CR04, CR05, CR06, and CR14) (Fig. 1). We originally proposed to monitor three springs (Boardwalk Spring, which could not be located, in addition to those mentioned above) and four surface-water sites (CR02 was eliminated and CR06 and CR14 were added). We proposed to monitor two sites during two storms, tentatively at hourly intervals in March and September, using ISCO automated samplers. However, because the ISCO samplers are not currently operational, we plan to collect grab samples at least at one surface water site during storms in spring and fall 2012. Temperature, specific conductance, and pH are measured in the field using a YSI multiparameter probe. Dissolved oxygen is measured by Winkler titration within 8 hours of sampling. Samples for lab analyses are passed through 0.45-micron pore-size disposable filters. Samples have been analyzed for sulfate, chloride, and phosphate by ion chromatography in the UK Environmental Research and Training Laboratory, and for nitrate and ammonium in the UK Department of Forestry. Oxygen-18

and hydrogen-2 (deuterium) are being analyzed in the UK Department of Earth and Environmental Sciences stable isotope laboratory using a gas-source, continuous flow, isotope-ratio mass spectrometer.

Networked computers and software, including ArcGIS and MATLAB, are being used for data analysis and interpretation. Data are being analyzed by standard graphical techniques such as bivariate plots. Analytical results are being entered into a GIS coverage of the basin. Statistical approaches such as principal component or hierarchical cluster analyses will also be employed. Spatial and temporal trends in solute and isotope concentrations will be sought, as well as relationships between concentrations and hydrologic parameters (e.g., precipitation and stream stage, which are monitored at several sites in the basin).

We expect that field parameters, solutes, and stable isotopes can provide supplemental evidence of sources of anthropogenic recharge. In particular, chloride and stable isotopes are considered to be conservative groundwater tracers, i.e., not affected by chemical reactions (other than dissolution, in the case of chloride). Stable isotopes are differentially fractionated by evaporation, and thus should indicate industrial cooling-water inputs.

Principal Findings and Significance

Samples have been collected and field water quality measurements have been made at each site biweekly since September 7, 2011. Results have been plotted using MATLAB to infer trends and anomalies for the different sampling locales under normal/baseflow and wet conditions. A comprehensive GIS database has been compiled for the basin. Layers include soils, lithology, watershed boundaries, streams, active USGS gages, roads, counties, sinkholes, and aerial imagery, as well as digital elevation model and land cover designations.

Field water quality data show significant variation between wet conditions (antecedent rainfall > 1.9 cm 72 hours prior to sample collection) and normal/baseflow conditions (antecedent rainfall < 1.9 cm) as well as seasonal variations. Field parameters are least variable among the sites that are considered to be dominantly groundwater, Citation Estavelle (CE) and Highland Spring (HS). In fact, CE does not appear to behave as an estavelle (an orifice that functions as a discharge point at low flow and a recharge sinkhole at high flow). CR1, located within the most industrialized area of the headwaters, exhibits the greatest variations in both field and lab analytes. Concentrations of chloride, sulfate and nutrients are elevated under normal/baseflow conditions relative to rain events (Fig. 2). The temperature variations of this site could be indicative of wastewater input from the surrounding industrial area. Isotopic data will be examined to further investigate this possibility and to determine the extent of ground water/surface water interaction at the CE and HS sites. Nutrient contamination is highest in the residential locations, including CR4 and HS. Under both wet and dry conditions, these sites have higher levels of phosphate and nitrate, especially in the winter months.

Leakage from sanitary sewers would be likely to result in elevated nutrient concentrations in baseflow conditions. Nutrient and field data for CR1 data indicate some degree of dilution following storms.

Biweekly monitoring of the seven sites will continue until September 2012. Two storm events will be monitored, tentatively in late April-early May and late September-early November. Sampling will occur at one surface-water site and, if possible, simultaneously at one spring at 30-minute to 1-hour intervals over a 24-hour period. Precipitation samples (collected since June 2011) will continue to be analyzed for stable isotopes. Isotopic data collected within the headwaters will be compared to a meteoric water line for the Fayette/Scott county area. Comparisons will be made between precipitation and Cane Run discharge based upon USGS gage data. Data on nutrient concentrations and field parameters from other locations in the basin during the 12-month sampling period will be sought for further comparison. Statistical analyses will be performed to determine the degree to which each site is related.

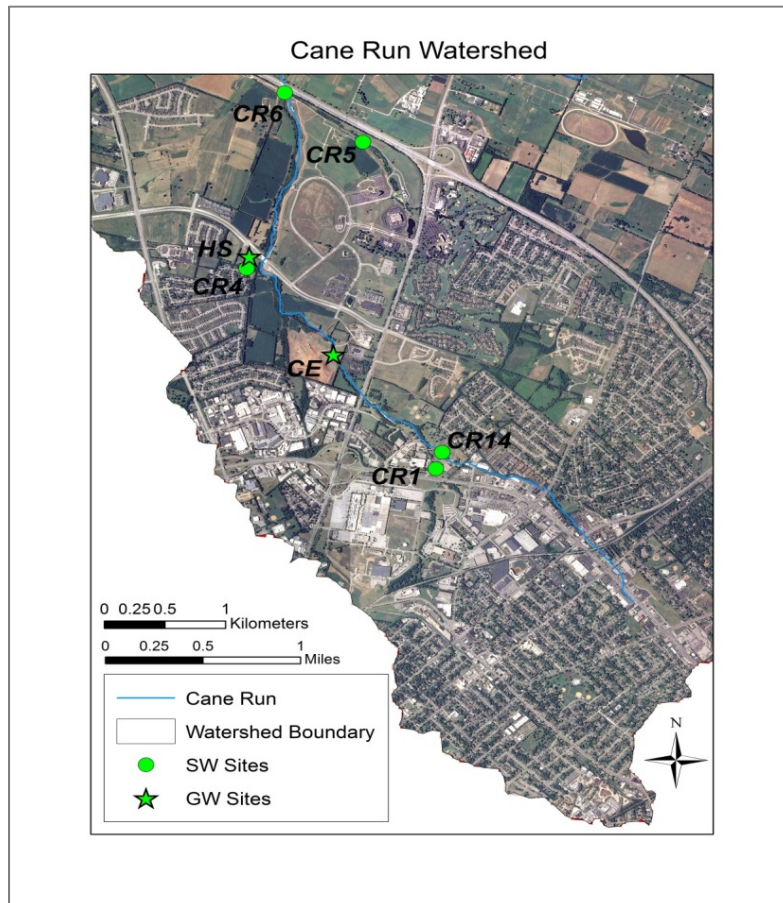


Fig. 1. Site locations; aerial imagery (USDA 2010).

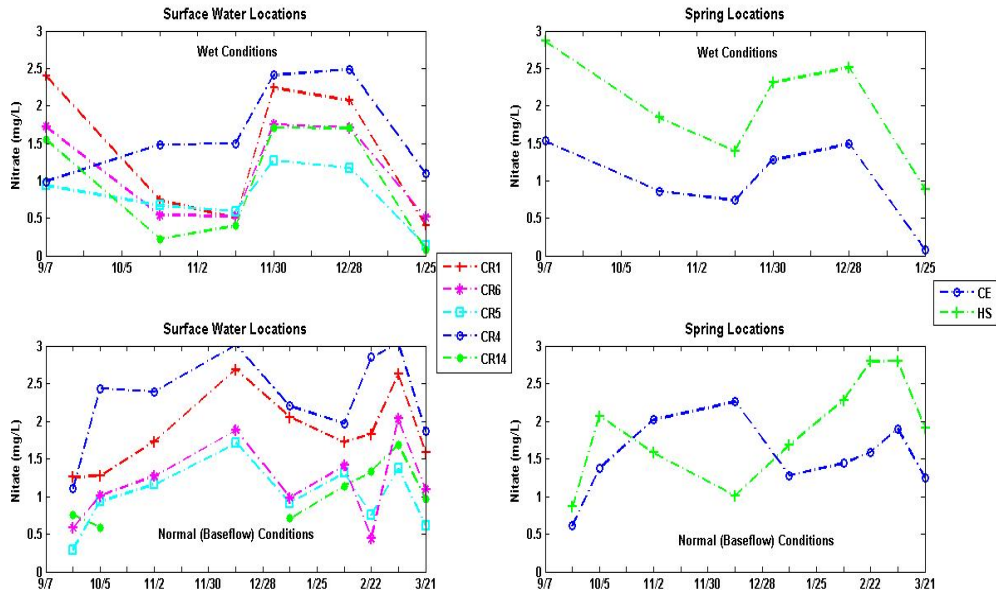


Fig. 2. Nitrate concentrations under wet and dry conditions at surface-water and spring locations.

Quasi-real time sediment discharge measurements using inexpensive experimental technology

Basic Information

Title:	Quasi-real time sediment discharge measurements using inexpensive experimental technology
Project Number:	2011KY170B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Engineering
Focus Category:	Sediments, Surface Water, Methods
Descriptors:	flow rate, sensor technology, light sensor, suspended sediment
Principal Investigators:	James F. Fox

Publication

1. Stewart, Robert, Jimmy Fox, Cindy Harnett, and Thomas Lawrence, 2012, Laboratory calibration of experimental velocity and sediment concentration sensors to monitor water and the environment, in Proceedings of the Kentucky Annual Water Resources Symposium, Kentucky Water Resources Research Institute, University of Kentucky, Lexington, KY, p. 81-82.

Quasi-Real Time Sediment Discharge Measurements Using Inexpensive Experimental Technology

Problem and Research Objectives

Sediment erosion and deposition in streams and rivers represents one of the predominant types of pollution impacting surface waters in Kentucky. Sediment pollution is harmful to aquatic communities and sediment can make filtering of water difficult for drinking purposes. Sediments act as a transport mechanism for pollutants such as heavy metals and pesticides. Carbon and other environmentally significant elements are also transported with sediment. Sediments can also fill reservoirs or river channels which can decrease the volume of water within the reservoir/channel thus reducing the amount of supply for drinking water and causing flooding in rivers. Temporal and spatial variability of suspended sediment in streams means that discrete monitoring is unlikely to characterize suspended sediment patterns accurately (Orwin and Smart, 2005). Continuous sediment discharge data, collected spatially across a watershed network is critical in developing erosion, sediment transport, and carbon flux models. High resolution data needs to be balanced with economic feasibility in a sediment monitoring plan. The use of an innovative, inexpensive sediment sensor network offers the potential to meet these needs.

The overall objective of this project is to determine the accuracy to which flow rate and sediment discharge can be measured using the inexpensive newly developed sensor technology. The effects of secondary variables on the calibrated sediment concentration relationship will be determined. A computer program will be developed that automatically calculates flow rate and sediment discharge from the data received from the newly developed sensors.

Methodology

The investigated sediment sensor technology may allow researchers to collect data that will assist in designing better watershed management plans and will increase the accuracy that researchers make on the estimates of pollutant and nutrient fluxes out of watersheds. The nature of this research is developing technology that can measure flow rate and sediment discharge in near real-time. Preliminary testing of the sensors in the laboratory show they are able to measure both velocity and sediment concentration accurately with high temporal resolution. Sediment discharge will be estimated using both highly accurate commercially available instruments as well as the new inexpensive real-time velocity and sediment concentration sensors developed by Dr. Cindy Harnett at the University of Louisville. The newly developed sensors will be calibrated in the laboratory and left in the field to measure sediment discharge where the results will be verified using highly accurate commercially available equipment and widely accepted techniques.

The newly developed sensors are comprised of inexpensive commercially available components that are being used in an innovative manner to measure sediment discharge. The velocity sensors use a strain gage that changes voltage as more force is applied to the sensor by the flow. The voltage read from the sensor is calibrated at known velocities so that a relationship can be developed. The sediment concentration sensors work in a similar fashion but are sensitive to light intensity, light intensity changes the voltage across the sensor. Percentage of light reaching the sensors will be measured at known depths and sediment concentrations to develop a relationship that relates depth and concentration to the relative amount of light reaching the sensor. A computer program will be used to optimize parameters of velocity and sediment concentration profile equations to the data collected by the sensors. Once flow velocity and sediment concentration profiles are determined they will be numerically integrated across the cross-section of the channel by the computer program. Flow rate times concentration will give sediment discharge with units of mass per time.

An in depth literature review showed that limitations exist for optical sensors as a measure of sediment concentration. (i) The relationship for concentration and light intensity can vary due to secondary variables (sediment size, color, shape, composition, and amount of organic material) (Connor and Visser, 1992; Sutherland et al., 2000). (ii) the relationship for concentration and light intensity needs to be developed for each watershed. (iii) the relationship may vary seasonally. (iv) the relationship may require repeated empirical calibrations (Connor and Visser, 1992)

In order to meet the stated objectives, the following scope has been outlined:

1. Further develop an in depth literature review of the flow rate and sediment concentration measurement on-going by other researchers;
2. Collect data in the lab using the newly developed sensors and compare with highly accurate commercially available sensors;
3. Collect data in rivers in Kentucky using both the newly developed and the commercially available sensors over a wide range of flow conditions;
4. Write a computer program that analyzes the data and calculates both flow rate and sediment discharge;
5. Verify accuracy of the data collected and the results of the program with data from other sources.
6. Provide a discussion of how the research results overcome the cited limitations identified in the literature.

Research is currently underway investigating sediment discharge studied by other researchers and the analysis techniques they used. From this review, general steps in the data collection methods and analysis have been identified and are being revised to better understand sediment discharge in rivers. Secondary variables have been identified for determining their effects on the optical sediment sensors.

Data will be collected in the laboratory for a range of sediment properties and concentrations expected in the rivers of Kentucky. The sediment concentration data from the newly developed sensors will be compared to a YSI turbidity probe and to direct measurements of sediment concentration. Sediment samples will be analyzed for particle size distribution and chemical composition. The new velocity sensors will be tested in a tow tank and compared to an acoustic Doppler velocimeter and a Gurley meter.

Data will be collected in the field to determine the sensors' effectiveness to be deployed for long term use and to verify that relationships developed in the laboratory remain valid in the field. All sensors will be used in the field, however, only the new velocity and sediment concentration sensors will be used in the very high flow conditions, due to safety concerns.

Since the sensors are expected to measure frequently (5 minute intervals) a computer program will be developed that is capable of calculating sediment discharge automatically from the data collected. The program will be either in Matlab or another suitable available computer language.

The data collected in the field will be verified with the acoustic Doppler velocimeter, Gurley meter, turbidity probe, direct sediment concentration measurements, and by comparing the results with a USGS gage station located near the sensor network deployment location.

A discussion of results will be provided that works to clarify the effects of secondary variables on measuring sediment concentration with the optical sediment sensors. The frequency and accuracy of data collected by the sensor network will be compared to methods used by other researchers.

Principal Findings and Significance

Laboratory data collection for Light Attenuation Sediment Sensors (LASS) and Velocity Bend Sensors (VBS) was performed and analysis is on-going. LASS findings show that experimental results agree well with existing theory for light attenuation and scattering caused by sediments. Macro-analysis of our results with other sediment-light attenuation studies in published water resources papers have allowed us to better understand our sensors and add some ideas to the literature. A semi-theoretical model for application of the LASS is still in its final development. We strive towards a more theoretical model but are reducing to semi-theoretical approach based on our available data/parameters.

VBS findings show the ability of the sensor to measure mean velocity. The VBS exhibit transition from rigid to elastic bending with increasing freestream velocity, which can be described using dimensionless fluid and beam bending properties. The relationship between stream velocity and voltage drop across the circuit is nonlinear. A semi-theoretical approach to estimate time-average streamwise velocity from the voltage drop

based on fluid drag, elastic member bending, and circuit principles is applied and shows good agreement with our experimentally derived calibration curves.

The VBS also shows the ability to measure turbulence in streams/rivers. The Triple Decomposition Theorem and spectral analysis are performed on VBS and acoustic Doppler Velocimeter (ADV) time-series. Results show that the VBS captures low frequency characteristics of macroturbulence present within the turbulent open channel flow but is unable to measure smaller-scale characteristics of eddy shedding for these hydraulic conditions. Turbulent intensity calculated using VBS data is 12% of that from the ADV attributed to the lack of detection of shedding sized eddies. The linear fit between turbulent intensity from the VBS and ADV suggest that the VBS can be used as a proxy for more detailed turbulent measurements when applied in streams.

Substantial progress has been made for Scope 1 and 2, including literature review and laboratory data collection and analysis. Scopes 3 through 6 will be focused upon during summer 2012.

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Metagenomic analysis of microbial urea transformation in soil

Basic Information

Title:	Metagenomic analysis of microbial urea transformation in soil
Project Number:	2011KY171B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Water Quality
Focus Category:	Nitrate Contamination, Agriculture, Non Point Pollution
Descriptors:	urease inhibitors, microbial processes, biotransformations
Principal Investigators:	Luke Moe, Mark Steven Coyne

Publications

There are no publications.

Metagenomic Analysis of Microbial Urea Transformation in Soil

Problem and Research Objective

Biological transformation of nitrogenous fertilizer has a significant impact on the speciation and load of nitrogen in runoff and leachate. The outcomes of this nitrogen loss from agricultural settings include eutrophication and alterations of ecosystem ecology in downstream bodies of water, often with disastrous effects. We seek to understand the role of microbial urease enzymes in biotransformation of agriculturally applied urea *in situ*. To this end, we seek to develop metagenomic methodologies to characterize the microbial urease populations in soils under varying agronomic regimes. Our objectives are to develop a functional metagenomic screen to identify microbial ureases from a soil metagenomic DNA library and to develop a gene-targeted metagenomic methodology that can be applied to characterize the diversity of urease encoding genes from a soil metagenomic DNA extract. Together, we aim to develop a metagenomic toolkit that will be used to correlate *in situ* urease soil assays with the content of the soil metagenome.

Methodology

The original proposal described a functional metagenomic approach for discovery and characterization of microbial ureases. Owing to complications associated with development of this methodology (described below), we have opted to incorporate an additional gene-targeted PCR-based metagenomic approach. Thus, we have focused on developing methods for identification of urease genes from the soil metagenome based on both a functional screen (as described in the original proposal) as well as a method based on PCR amplification and sequencing of DNA sequences that show identity to known microbial urease genes. The PCR-based method relies on development of degenerate PCR primers that will be used to specifically amplify DNA encoding urease genes. Briefly, the primers are developed based on comparison of microbial urease genes in genbank. Regions of high identity among urease genes are used to develop a forward and reverse primer set that will amplify a ~280 bp DNA region that shows sequence divergence that is sufficient to distinguish between closely related genes, yet conserved enough to be able to identify the sequence as a urease gene. We will use the PCR primers to amplify urease genes from the soils of interest then we will clone and sequence the genes in order to assess the diversity of urease genes from that environment. Additionally, we will assess soil urease activity from these soils using conventional soil urease assays. Ultimately, we plan to correlate soil urease activity with the abundance and diversity of urease genes from these environments.

Principal Findings and Significance

Initial work focused on developing a growth selection strategy to identify urease genes from soil metagenomic libraries. The metagenomic libraries were transformed into a laboratory *E. coli* strain and selected for growth on urea as the sole nitrogen source. Because this strain does not encode a urease, it cannot naturally liberate ammonia from

urea and therefore cannot use urea as a nitrogen source. After verification of this using growth studies, recombinant metagenomic libraries were constructed and screened but did not yield any growth among the recombinant clones. We obtained a vector with a functional urease gene cluster from Professor Harry Mobley (University of Michigan) to verify that, in the presence of a functional urease, the laboratory strain of *E. coli* would liberate ammonia from urea and grow. This vector, pMID1010, encoded a complete urease gene cluster from the bacterium *Proteus mirabilis*, and gave a weak growth phenotype under these selection conditions. We conducted a genomic screen from the urease positive bacterium *Helicobacter pylori* RPH 13487 and did not obtain a positive clone using this strategy. Ultimately, the growth selection strategy as originally designed did not prove to be an efficient method for identification of urease genes from a soil metagenomic library.

In addition to the growth selection strategy, we developed a functional metagenomic screen to detect the same urease reaction. The screen is based on the components of “urea segregation agar” that allow for a color change in the medium to be detected if a colony is exhibiting urease activity. This is a simple pH indicator method in which a red halo surrounding a colony on the indicator medium is indicative of liberation of ammonia from urea in the medium. The control vector pMID1010 gave a strong positive phenotype in *E. coli* in urea segregation agar, and the PCR amplified urease gene cluster from *H. pylori* also gave a strong phenotype when subcloned, but functional metagenomic screens on this medium did not yield any positive metagenomic clones from several soil metagenomic DNA libraries.

Because urease is a multiprotein complex that requires efficient expression of multiple genes to be functional (e.g. the urease operon of *H. pylori* is ~6Kbp), and because it also requires efficient uptake of nickel from the growth medium for the urease active site, we inferred that a more efficient method for functional screening would be to include a number of ancillary urease genes within the screening vector. To this end, we have constructed a cloning vector that incorporates a nearly complete urease operon—minus the gene encoding the main urease protein subunits. The vector includes the urease gene cluster from *H. pylori* as well as a gene encoding a nickel uptake mechanism (*nixA*), but it is missing the *ureA* and *ureB* genes that encode the protein components harboring the enzyme active site (therefore the vector is urease negative in *E. coli*). Our hypothesis is that with the remaining functional urease components, identification of *ureA/B* genes through a functional metagenomic screen may be more feasible than identification of complete operons through the screen. This vector requires addition of metagenomic DNA cloning sites at a prescribed location within the urease operon, thus we have removed the *ureA/B* genes and engineered novel restriction enzyme sites within the urease cluster. The vector is currently undergoing sequence verification before deployment in a functional screen.

Regarding the metagenomic PCR-targeted strategy, we have designed and tested a number of different PCR primer sets based on different primer design strategies. One set in particular, based on the Conserved Domain Database of NCBI, has proved to be

suitable for amplification of urease genes from positive control genomic DNA as well as from soil metagenomic DNA extracts. As a test, we amplified metagenomic DNA using the primers, subcloned the appropriately sized (~280 bp) fragments, and sequenced 16 of the subclones. Of the 16, 15 gave BLAST results with strong similarity to urease sequence indicating that the strategy was successful in selectively amplifying urease genes.

We were unable to get appropriate field samples during the summer months in 2011. We will obtain them this summer (2012). This may have worked out better ultimately as we had not yet optimized the PCR-based strategy last summer. With the PCR primer set now validated, we are set to perform experiments assessing the molecular diversity of microbial urease genes in soils that exhibit varying degrees of total urease activity. We have identified sites at the University of Kentucky research farm that are suitable for these purposes. We have materials for the soil urease assays and for metagenomic library construction, and plan to commence with sampling in early May 2012. We anticipate that the project will be finished by the end of the summer.

characteristics to investigate intraspecific differences of largemouth bass and paddlefish due to the construction of modern

Ordination of fish morphological characteristics to investigate intraspecific differences of largemouth bass and paddlefish due to the construction of modern navigation dams on the Ohio River, USA

Basic Information

Title:	Ordination of fish morphological characteristics to investigate intraspecific differences of largemouth bass and paddlefish due to the construction of modern navigation dams on the Ohio River, USA
Project Number:	2011KY172B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Biological Sciences
Focus Category:	Ecology, Surface Water, None
Descriptors:	trophic shifts, ordination analysis
Principal Investigators:	Tamara Sluss

Publications

There are no publications.

Ordination of Fish Morphological Characteristics to Investigate Intraspecific Differences of Large Mouth Bass and Paddlefish Due to the Construction of Modern Navigation Dams on the Ohio River, USA

Problem and Research Objectives

Damming is one method of regulating large rivers, typically resulting in a less variable system laterally and increased water residence times, similar to lacustrine systems. These habitat shifts may impact fish feeding morphology but such a study has been difficult to undertake due to the paucity of data from large rivers and the rapid construction of dams with little prior biological sampling.

The original objective of this research was to determine whether feeding morphology has changed within planktivorous (paddlefish) and a piscivorous (large mouth bass) fish species due to the construction of modern navigation dams on the mainstem Ohio River between Louisville, KY and Cincinnati, OH, USA. The planned nature and scope of the research involved studying fish specimens in archival collections that spanned the construction of the McAlpine and Markland dams in 1961 and 1964 respectively through the analysis of digital imagery and ordination.

Unfortunately, we were not able to locate actual specimens from before damming on the Ohio River. The collections in our query had records stating that they had the fish specimens, but we were able to locate only one fish from the pre-dam period in collections maintained by Thomas More College and the Cincinnati History Museum. Therefore, we were forced to shift our focus to investigate the differences in feeding morphology of bluegill and freshwater drum from specimens collected in alluvial pools compared to the morphology of specimens from constricted channel reaches of the Ohio River.

Methodology

The Ohio River is over 1,500 kilometers long with 20 navigational dams segmenting it into navigational pools. This study was conducted to determine the effects of navigational dams on feeding morphology for two predatory fish, freshwater drum, *A. grunniens* (n=155) and bluegill, *L. macrochirus* (n=45) in pools with differing geomorphology using specimens from the Cincinnati Natural History Museum. Feeding parameters (jaw length and gap width of the mouth) were ascertained using a Nikon D90 digital camera and SPOT Advance for each individual fish. Parameters were then analyzed with principle component analysis by PC-ORD.

Principle Findings and Significance

The *A. grunniens* morphological parameters are less similar for individuals from the semi-constricted glaciated valley than for individuals that inhabited the alluvial valley and upper Ohio River. The *L. macrochirus* feeding morphology is more similar between

individuals in the semi-constricted glaciated valley and the upper Ohio River. The influencing morphological measurement was jaw length; *A. grunniens* mean jaw length was 0.273cm and *L macrochirus* mean jaw length was 0.228cm.

Although, we had to be flexible for this study, we believe that the results contribute to the field of large river science - rarely studied due to their size, distance from replicates, expense, variability, and because they are considered open systems. The image software and statistical analyses can be used for future studies. This approach is easily transferrable to other species or experimental designs. Ordination techniques are especially valuable as they can be used to ask questions using suites of variables at multiple hierarchical levels (individuals to ecosystems).

The effects of prescribed fire on amphibian diversity

Basic Information

Title:	The effects of prescribed fire on amphibian diversity
Project Number:	2011KY173B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 1st
Research Category:	Biological Sciences
Focus Category:	Ecology, Surface Water, Water Quality
Descriptors:	freshwater ecology, community dynamics, species richness
Principal Investigators:	Howard H. Whiteman

Publication

1. Knopp, Robert and Howard Whiteman, 2012, The effects of prescribed fire on amphibian and reptile diversity in an oak-grassland restoration area, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, University of Kentucky, Lexington, KY, p. 21.

The Effects of Prescribed Fire on Amphibian Diversity

Problem and Research Objectives

Prescribed burning simulates the occurrence of natural wildfires with the intent of restoring the flora to its natural state prior to any fire suppression techniques (Garren 1943; Barden and Woods 1976). These controlled burns alter the ecosystem in many ways that may impact reptiles and amphibians. Leaf litter and coarse woody debris decreases after a burn event (Albrecht and Mattson 1977), which are primary micro-habitat sources for upland snakes and other herpetofauna, including amphibians (Owens *et al.* 2008). Inorganic nutrients often increase in soil and nearby water sources which result in more nutritious plants (Knight and Holt 2005), promoting macroinvertebrate community shifts (McGann 2006; Minshall *et al.* 2001; Minshall *et al.* 2001b). Community changes can lead to an increase (Knight and Holt 2005) or decrease (Johnson *et al.* 2008) in macroinvertebrates depending on the habitat, thus affecting many amphibians, smaller snakes and lizards (Moore and Sloan 2007). Canopy coverage of burned forests is less dense than a forest that hasn't been burned or thinned (Phillips *et al.* 2004), potentially causing lower moisture levels (Anderson 1965), which may affect amphibian abundance, a primary food source of upland snakes (Moore and Sloan 2007). The specifics of these causalities are complex, but it is clear that fire management has the potential to significantly affect amphibian and reptile populations.

Prescribed burns have been shown to affect multiple species in diverse ways, yet few studies have focused on reptiles or amphibians, particularly near water resources. Ford *et al.* (1999) found no significant changes in herpetofauna (amphibians and lizards) or small rodent numbers before and after burns; however, the study admits that collections were done early in the season before hibernation was at an end. A significant change in community structure of herpetofauna was reported in a similar study area: lizard species increased dramatically while other herpetofauna remained constant (pine and oak-hardwood forests in Maryland; McLeod and Gates 1998). Langford *et al.* (2007) discovered an increase in specific species (i.e. oak toad, southern cricket frog and eastern mud turtle) after a prescribed burn, although most species were not affected. Perry *et al.* performed a similar study in pine woodlands and found niche dependent results and no clear response from many species (2009). Clearly, there is still a great deal of uncertainty as to the results of prescribed burning upon reptiles and amphibians. This study contributes by assessing effects of prescribed burns on reptiles and amphibians that exist within an oak-grassland dominated ecosystem.

Given the wide range of results described in the referenced studies it is difficult to predict the outcome of this investigation; change is anticipated, but what form that change will take is highly dependent upon the composition of species present within a specific habitat. From a competition perspective, it is likely that species well adapted to drier warmer environments will excel in restored oak-grasslands, outcompeting and filling niches at a higher rate as compared to water dependent species, due to higher light levels and lower leaf litter. For instance, organisms that are resistant to desiccation such as snakes and lizards are expected to be found in high numbers. Shallow fossorial

dwellers (many salamanders, anurans and small snakes) may experience reduced habitat because of fewer substrates to live beneath. The decrease in land micro-habitat, such as leaf litter and coarse woody debris (CWD), may directly affect the abundance of semi-aquatic species, such as salamanders, anurans, turtles and aquatic snakes by reducing places to lay or brood eggs, reducing cover used as predator-avoidance, and reducing prey habitat. These potential results will be reflected through changes in species abundance, composition, richness and diversity.

Methodology

This project is taking place in Land Between The Lakes National Recreation Area of northwestern Tennessee and southwestern Kentucky. The specific study area encompasses approximately 8,630 acres (3492.44 ha) in the Oak-Grassland Restoration Area (OGRA; Fig. 1), which has been managed via prescribed burning since 2006.

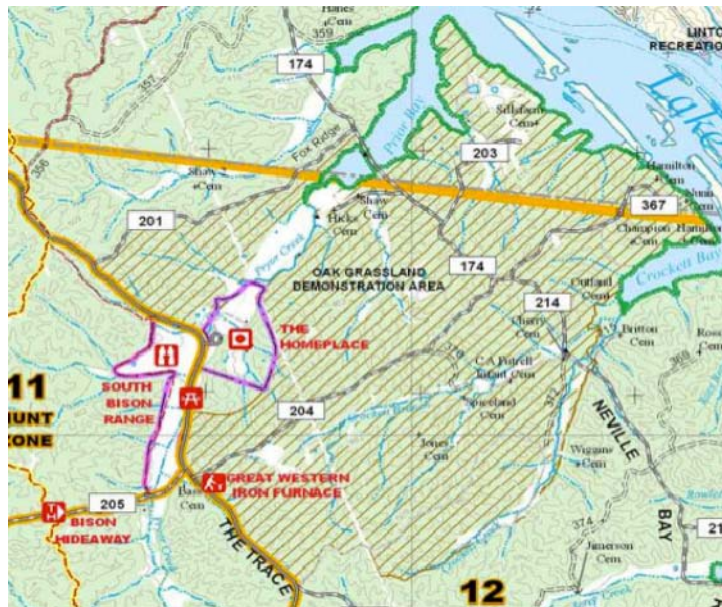


Fig. 1: Oak-Grassland Demonstration Area (OGRA) at the Land Between the Lakes National Recreation Area.

Sample sites focus on forested areas within and outside of the OGRA that have been subjected to different prescribed burning practices (= treatments: no burning, and burned within 5-6 years). In May, 2012, we will determine tree species composition, canopy cover, leaf litter depth, percent ground cover and density of coarse woody debris (CWD) for each of the chosen areas, using methods proposed by Condit (1998) and Harmon and Sexton (1996). Values for canopy cover, leaf litter depth, and CWD (= >10cm) will be calculated as an average of the total sum for each category found along each 100m transect associated with each pond.

Canopy cover will be calculated using a Cajanus tube. Leaf litter will be measured in centimeters above the ground using a simple metal rod. CWD found along each transect will be measured at both ends and in the middle to determine volume using calipers and logger's tapes.

Pond sites are wildlife ponds found within and outside of the OGRA that have been subjected to the different prescribed burning practices, with eight ponds within each burning frequency treatment and eight control ponds. Sites will be matched for size, slope, permanence and tree species diversity to minimize differences both within and between treatments. We are collecting data on canopy cover and water quality (e.g., temperature, pH, DO, nutrient levels) for each of the chosen ponds. Water quality has been checked on a biweekly basis, using a portable YSI meter (Lynch and Shapiro 1981), during sampling efforts. Ponds were sampled using dip nets on a bi-weekly basis. Six random dips were taken while standing at the shoreline of each pond. Each dip net effort was approximately 1.5 m in length. We performed a total of 10 dip-netting days during June, July and August 2011. Additionally, minnow traps were employed for five consecutive days during June, July and August. All herpetofauna captured were identified to species and released back in the pond, along with any other organisms captured. Reptiles were captured and/or sight identified at each pond site, using hand methods in conjunction with the methods described above.

We used a modified version of a drift fence design used by Burgdorf et al. (2005) (Fig. 2). Fences were made of aluminum construction flashing measuring 46 cm in height. Drift fences were installed adjacent to each pond on the southwestern side. The long axis (10 m) of each drift fence was oriented northwest to southeast, while the shortest side (5 m) was directed southwest. The long and short legs intersected at a 90° angle. At the point of intersection, a .6m by .6m funnel trap made of wire mesh was used to capture large snakes. The fences and box traps were buried 7cm to 12cm beneath the soil's surface. Each box trap was supplied with a water dish. Twelve 3.79 L pitfall traps were buried against the drift fence legs; two pitfall traps were located on each side of the legs at opposing ends. Water and filtration holes were provide in each pitfall. All traps were provided with a water source. Fences were installed in late summer and activated on September 5, 2011. All fences were checked daily through October 20, 2011. Organisms found at drift fences were noted and classified to species.

Drift fences were reopened on January 22, 2012 and closed May 5, 2012. The data for this sampling period has not been compiled at this time. Therefore this sampling period is not included in the preliminary results.

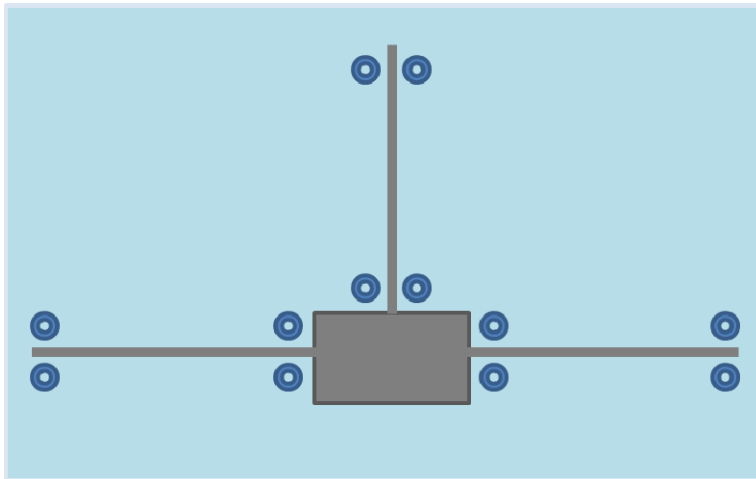


Fig 2: Drift fence design used in this study.

Shannon's diversity indices were used to calculate and compare diversity between the control area (non-burn) and the experimental area (burn). These values were then tested using the bootstrapping method (Efron and Tibshirani 1993). A percentile bootstrapping test was used each H' for the three sampling methods (dip-netting, minnow traps and drift fences) fell within a bootstrap 95% C.I. using 1,000 iterations of the test. We used a Bootstrapped t-test to determine significance. After all data are compiled, we will analyze differences in these variables as well as terrestrial habitat variables using a permutation multivariate analysis of variance (PERMANOVA).

Principal Findings and Significance

Sampling for herpetofauna has been completed successfully, and has led to over four thousand individual captures. We are currently working to analyze the species data and determine its full meaning and implications for the effects of prescribed fire upon herpetofauna. Currently, we know that dip-netting allowed us to document nine species in the control and eight species in the burn area, and Shannon Diversity values were not significantly different from one another (Fig. 3). However, minnow trapping and drift fences both resulted in statically higher diversity in the burn area (Figs. 4 and 5). Thus, two out the three methods suggest that prescribed fire in our study benefits amphibians and reptiles. However, there is still a great deal of data to analyze before our findings become fully conclusive. Considerable data collected after January 2012 will be added to our analysis. Additionally, we are currently working to finish vegetation transects so that these control variables (e.g., canopy coverage) may aid in our understanding of the interaction between the physical environment and the heptofauna.

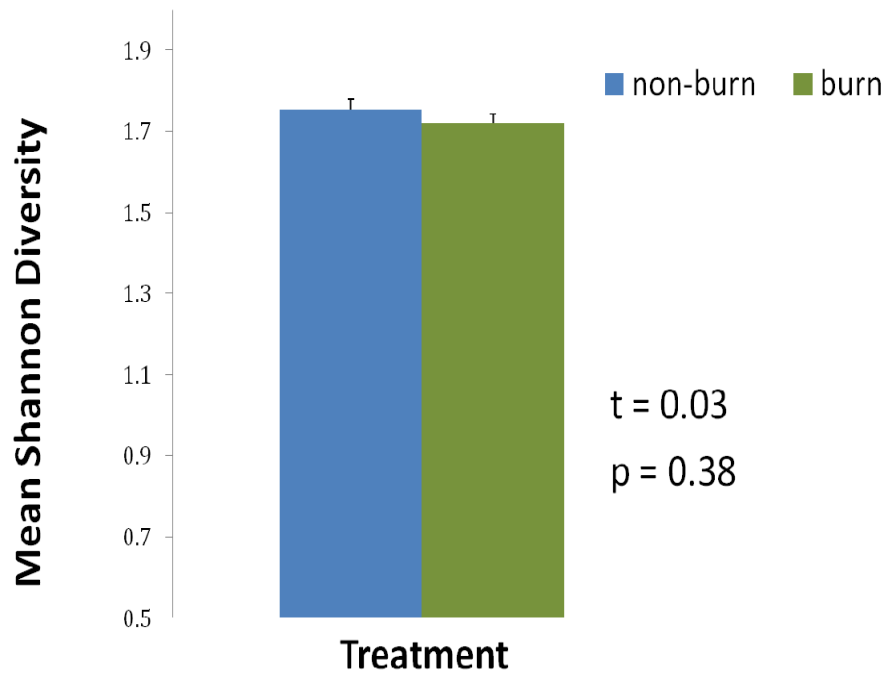


Figure 3. Mean Shannon diversity values after bootstrapping for dip-netting. Nine species were located in the control and eight species in the experimental area.

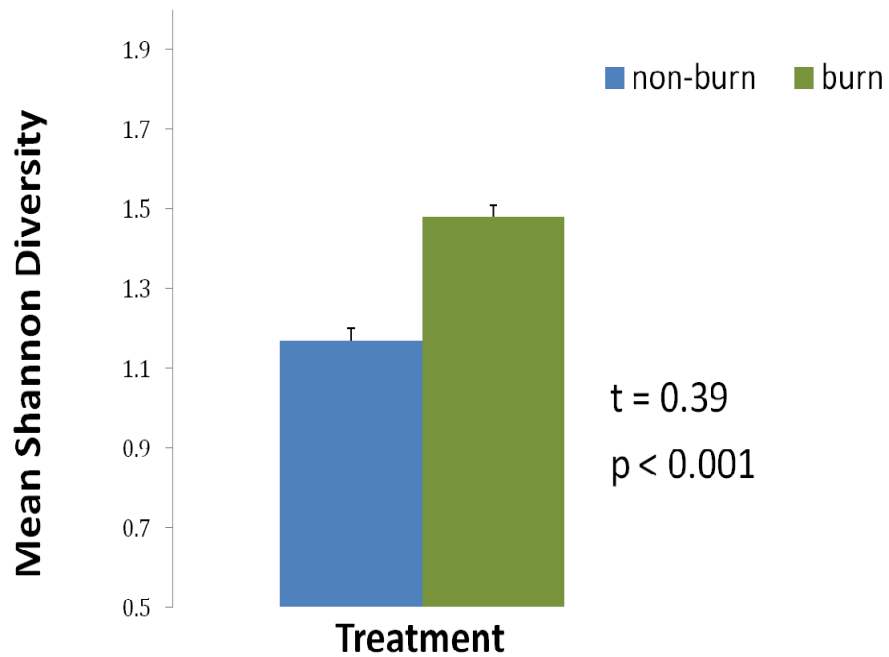


Figure 4. Mean Shannon diversity values after bootstrapping for minnow traps. Ten species were located in the control and twelve species in the experimental area.

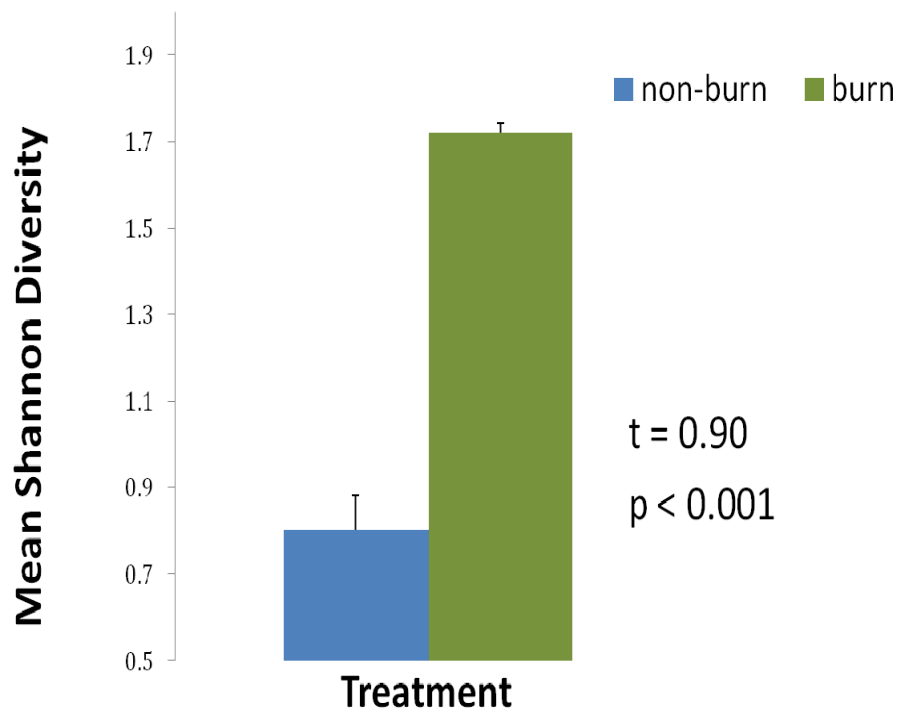


Figure 5. Mean Shannon diversity values after bootstrapping for drift fences. Twelve species were located in the control and fifteen species in the experimental area.

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By-proxy monitoring of aqueous nitrate photolysis and the effect of hydroxyl radical

Basic Information

Title:	By-proxy monitoring of aqueous nitrate photolysis and the effect of hydroxyl radical
Project Number:	2011KY174B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 2nd
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Nitrate Contamination, Methods
Descriptors:	photochemical reactions, infrared vibrational spectroscopy
Principal Investigators:	Matthew Nee

Publication

1. Wyatt, Jonathan and Matthew Nee, 2012, By-proxy monitoring of aqueous nitrate photolysis and the effect of hydroxyl radical, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, University of Kentucky, Lexington, KY, p. 87.

By-Proxy Monitoring of Aqueous Nitrate Photolysis and the Effect of Hydroxyl Radical

Problem and Research Objectives

The photolysis of aqueous nitrate ion (NO_3^-) in the natural environment has been shown to produce a variety of compounds which are considered either human health hazards or are potentially environmentally dangerous. These include the nitrogen oxides ($\text{NO}_x = \text{NO} + \text{NO}_2$) and ozone. To properly model the impact that nitrate photolysis has on climate change and atmospheric composition, a full set of kinetic data (rate constants, mechanisms, and branching ratios) must be known for the complex network of reactions which terminate in the production of NO_x and O_3 .

Some of these reactions are shown in Figure 1. The laboratory experiments performed here represent one aspect of the information which is necessary for proper modeling of the Earth's atmosphere. The original project objective was to use the broken symmetry in the infrared spectrum of nitrate ion as a proxy for the reaction kinetics occurring surrounding the chromophore. We have shown that, while this approach may be successful using Raman spectroscopy, no change is indicated by our experiments in the infrared spectrum. However, in the process of reaching this conclusion, we have found that we are able to observe a temperature dependence to the split peaks in the infrared spectrum of nitrate ion. This information gives fundamental insight into the energetic origin of the two peaks, and allows us to explore symmetry breaking in nitrate ion as we search for other means of exploring the kinetics following photolysis.

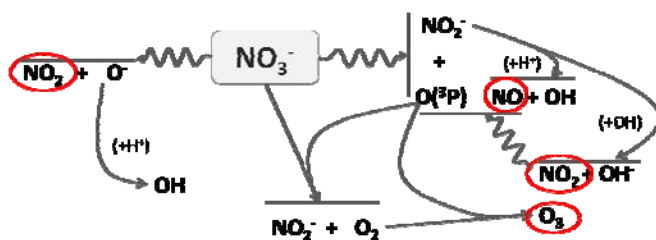


Figure 1. A sample of reactions occurring following the UV photolysis of aqueous nitrate ion. Wavy lines indicate light-triggered reactions, while those compounds circled are greenhouse gases NO_x ($\text{NO}+\text{NO}_2$) and O_3 .

Methodology

Infrared spectra of nitrate ion were collected every 30 seconds for a total of three and a half hours following exposure to ultraviolet radiation (photolysis). A standard mercury lamp source was used as the photolysis radiation. The infrared spectrometer used was a HP Spectrum One Fourier transform infrared (FTIR) spectrometer. Samples were sealed between two sapphire windows in a custom-built temperature-controlled sample cell holder. Sapphire is needed because it is resistant to degradation by water and also passes ultraviolet radiation. Normally, aqueous spectra cannot be well monitored with infrared spectroscopy because of the large signals due to water absorption. However, NO_3^- has its primary infrared signal (asymmetric stretching mode) in the range from 1200 to 1500 cm^{-1} , which is completely clear of any water absorption. It is well established that the spectral line shape is well described by two Gaussian peaks corresponding to different molecular geometries of nitrate which exist in solution. Because these two geometries

arise as a result of the electrostatic forces between nitrate ion and water molecules, our original hypothesis was that, as the photolysis reaction proceeded and the concentrations of different components changed, the relative amounts of the two geometries would change. This would manifest as a change in the relative areas of the Gaussians used to fit the infrared spectrum.

Originally, we had intended to perform these experiments in the presence of benzoic acid, which has been shown to serve as a proxy for the presence of hydroxyl radical, one of the main reaction components seen in Figure 1. Although we were not able to see any changes to the nitrate ion signal, those experiments did reveal for the first time that the infrared spectrum may be sufficient to indicate the presence of hydroxyl radical in solution using the benzoic acid proxy.

Principal Findings and Significance

Figure 2 shows the complete spectra for several representative times following photolysis. Both show strong water signals which are saturated (despite background subtraction). These are the “noisy” sections of the spectrum from 500 to 1000 cm^{-1} , from 1500 to 1650 cm^{-1} , and from 3000 to 3650 cm^{-1} . Also prominent in both is the nitrate ion asymmetric stretch (1200 to 1500 cm^{-1}), which will be discussed in greater detail below. The primary difference seen between these two spectra lies in the region from 2000 to 2400 cm^{-1} . In this region, circled in Figure 2, the spectra evolve slowly over time for the spectra in which benzoic acid has been added as a proxy for the detection of hydroxyl radical. Thus, the amount of hydroxyl radical present can be assumed to evolve with time following photolysis. Further experiments are underway to assess this effect more quantitatively.

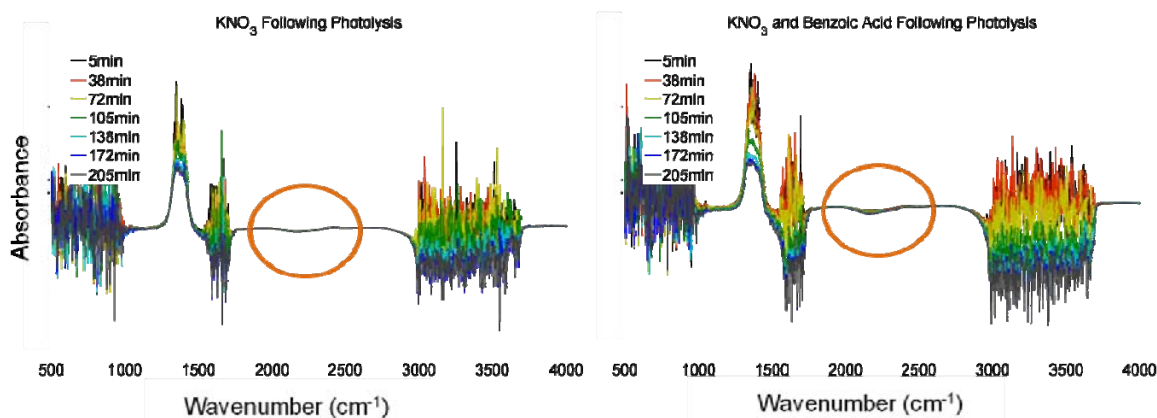


Figure 2. Representative FTIR spectra of solutions of KNO_3 with and without the addition of benzoic acid as a proxy for the generation of hydroxyl radical. On the right, changes to the intensity of the broad feature from 2000 to 2400 cm^{-1} are interpreted to indicate changes in the presence of hydroxyl radical.

The results of our experiments with and without benzoic acid are shown in greater detail for the asymmetric stretching region in Figure 3. These results show a representative sample of spectra collected at various times following five minutes of UV

exposure. In the top panel, several spectra are shown. Although an overall total decrease in nitrate ion signal appears to occur during the course of the experiment, the relative intensities of the peaks does not appear to change. This is shown in the lower panels, which show the peak areas fitted from all data collected. To support that the peak fitting algorithm used is valid, the centers of the fitted peaks are also shown. It is clear that neither the areas nor the centers of the peaks change during the course of the reaction. Thus, while we are able to monitor the presence of hydroxyl radical by adding benzoic acid, we are not able to correlate this to a change in the nitrate peak areas as we had originally hoped.

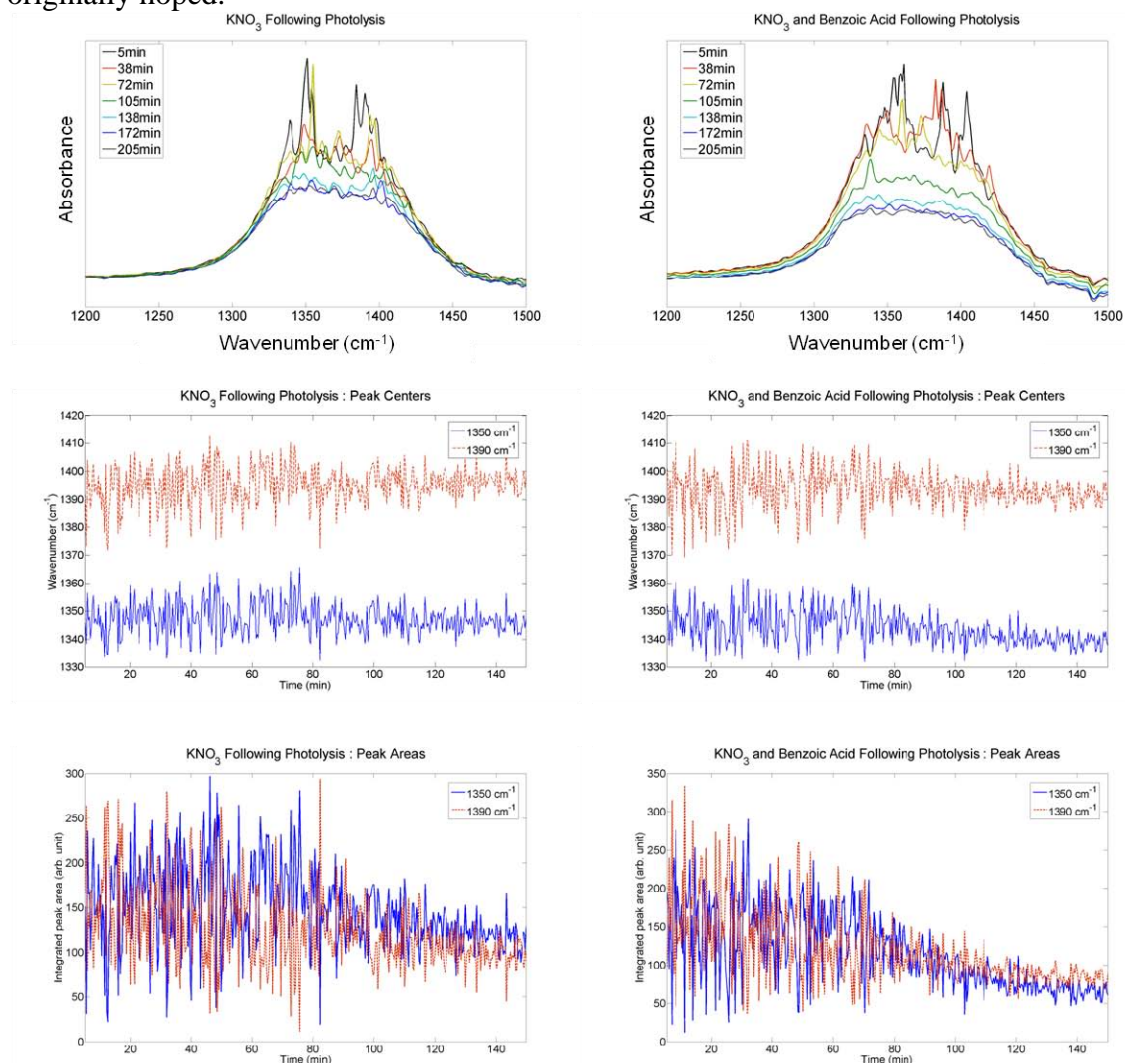


Figure 3. Details of the asymmetric stretch region following photolysis. Solutions with (right) and without (left) benzoic acid behaved indistinguishably. However, contrary to our hypothesis, no significant changes in the relative intensities of the two peaks occurred, indicating that solvation geometries are not affected by photolysis.

Beyond our original intentions, we have shown that the relative intensities of the different solvent geometries does change as a function of temperature, in the absence of photolysis. This behavior is shown in Figure 4. As the temperature increases, the peak at high frequency increases as the peak at low frequency decreases in area. This implies

that, as more thermal energy is available to the system, the solvation geometry with the higher frequency becomes accessible. More data points (over a broad range of temperatures) will be able to provide the relative enthalpies and entropies of the two states.

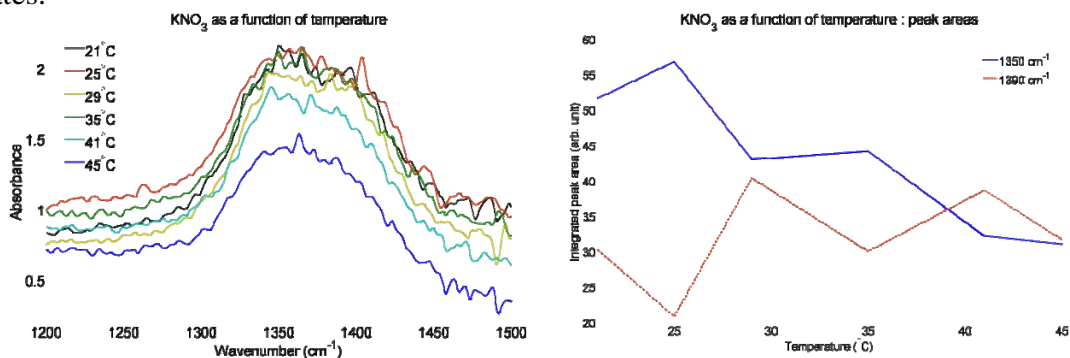


Figure 4. Changes to the relative intensities of the two solvation geometries of aqueous nitrate ion as a function of the temperature of the solution. These data indicate that the thermodynamic parameters for conversion between the two geometries can be determined experimentally, which provides fundamental information on the nature of the solvation environment.

This project has made good progress in spite of some minor hurdles regarding formal staffing. The original student Co-PI left our program, but an undergraduate student was able to continue the project during January and February 2012. We completed milestone (1) from our original plan. This has allowed us to conclude that, while our original hypothesis was not correct, FTIR spectroscopy can be used to monitor hydroxyl radical by proxy in the presence of benzoic acid. This will prove useful to future studies which need the sensitivity and selectivity that FTIR provides to monitor other reactions in aqueous solution. All of this work will continue to improve our understanding of the production of atmospheric contaminants such as NO_x and O₃. Proceeding according to the original plan is no longer prudent. However, more quantitative data regarding the detection of hydroxyl radical in solution will be undertaken with the remainder of the funding.

From a more fundamental standpoint, we are poised to explore a completely different aspect: the origin of the two peaks seen in the nitrate ion asymmetric stretch spectrum. The preliminary data shown in Figure 4 indicates that we should be able to collect enough data for a peer-reviewed journal article.

During summer 2012, a student enrolled in our combined BS/MS program will complete the data collection for the temperature-dependent studies. In June, he will collect higher-quality temperature dependent data which will allow us to determine the thermodynamic parameters relevant to the different solvation geometries in aqueous nitrate. He has already begun computational chemistry calculations which will provide a theoretical basis for the behavior seen. In July, he will expand the study to include multiple counterions (K⁺, Na⁺, Ca²⁺, and NH₄⁺), and will test the effects of concentration and ionic strength on the thermodynamics.

A second student who has just completed her master's thesis will be employed over the summer to expand her expertise to interpret the benzoic acid data. She will test a variety of different solutions (with and without nitrate; with and without hydrogen peroxide as a hydroxyl radical source) for their benzoic acid response in the 2000 to 2400 cm^{-1} region of the spectrum.

Arsenic removal from water with functionalized magnetic nanoparticles

Basic Information

Title:	Arsenic removal from water with functionalized magnetic nanoparticles
Project Number:	2011KY175B
Start Date:	3/1/2011
End Date:	2/29/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Water Quality
Focus Category:	Toxic Substances, Water Quality, None
Descriptors:	water treatment, filtration
Principal Investigators:	David A. Atwood

Publication

1. Atwood, David, Partha Jana, R. Douglass, and Robert DeJaco, 2011, Arsenic (III) removal from water with new thiol-containing filtration materials, in Proceedings 67th Southwest Regional Meeting of the American Chemical Society, Austin, TX, #1.

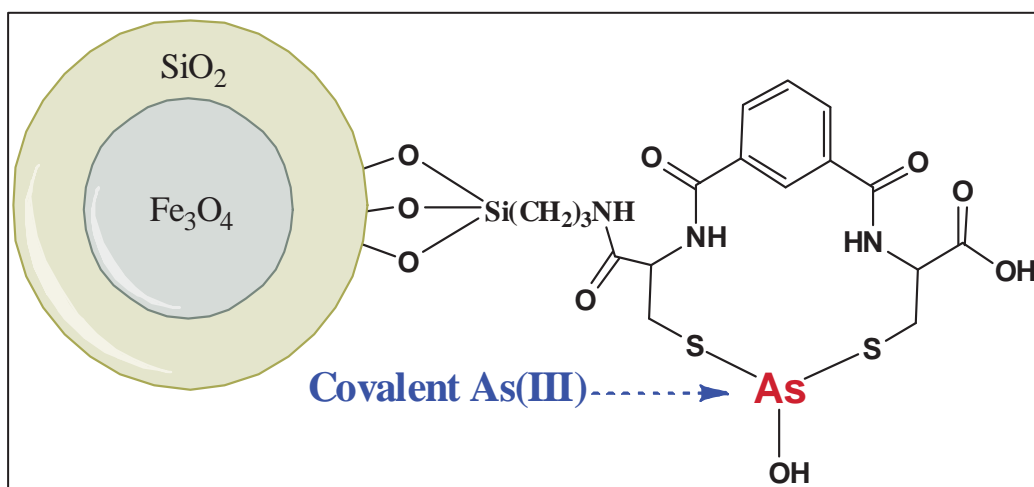
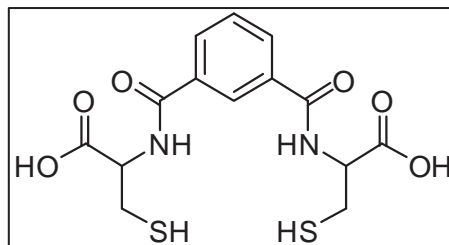
Arsenic Removal from Water with Functionalized Magnetic Nanoparticles

Problem and Research Objectives

Magnetic nanoparticles (MNPs) have been studied extensively for various applications such as magnetic resonance imaging, bimolecular sensors, bioseparation, and as sorbent materials that can be readily removed from a solution by the application of an external magnet. This project will utilize this property of MNPs to create a nanocomposite material that can capture and remove arsenic(III) from water.

The principle of the separation process is to utilize MNPs coated with a silica core shell in order to adsorb functional compounds. In this project the functional compound is based on compounds known to covalently capture arsenic(III). After arsenic(III) capture, the MNP will be magnetically separated from the aqueous solution being treated.

N,N'-bis(2 mercaptoethyl) isophthalamide) abbreviated, BDTH₂, known commonly as "B9" has a high affinity for "soft" heavy metals and metalloids, such as arsenic. B9 is capable of removing arsenic from contaminated solutions through covalent bonding. The carboxylate derivative of B9, BDTH₂-COOH, known commonly as "AB9" (shown in the adjacent figure) was prepared by graduate student, Partha Jana in a previous project (2010KY148B). The current project involves attaching AB9 to silica-coated MNPs in order to prepare a solid reagent with high capacity for arsenic binding and ease of isolation from water. The objective of the project is the creation of the composite MNP-Si-AB9-AsOH as shown in the figure below.



Methodology

The first step in the procedure was the synthesis of MNPs with specific size and homologous morphology. MNPs were prepared by coprecipitation of iron(II) and iron(III) ions in basic solution. The MNPs were then coated with a silica layer by a sol-gel reaction. The MNPs were then to be treated with poly-(vinylpyrrolidone) (PVP) and mixed with tetraethoxysilane (TEOS) under basic conditions to form the silica shell around the MNPs. The PVP coating described in the original proposal was found to be unnecessary and the MNPs were successfully coated with TEOS. The alternate approach using poly(oxyethylene) nonylphenyl ether as the nonionic surfactant was also not necessary.

The second step, also already achieved, was to functionalize the Si-MNPs with the “linker” (3-aminopropyl)triethoxysilane through reflux in toluene. The presence of the free amino group produced a covalent attachment site to the Si-MNPs. The concentration of free amine attached to Si-MNPs will be determined by titration. Following the silanation, the condensation of AB9 will result in the formation of the desired target structure shown in the figure above. Characterization of the final product will be carried out by infrared spectroscopy (IR) and thermogravimetric analysis (TGA). Elemental analysis will be used to show the relative amounts of both nitrogen and sulfur on the Si-MNPs before and after functionalization. Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES) will be used to quantify the amount of sulfur on the particle, and that information will provide the amount of compound on the surface (in mmol/g).

The final step will be to quantify the ability of the AB9-Si-MNPs to covalently capture As(III) from water. Batch and column studies will be performed on arsenic-containing water samples. The arsenic-removal ability of the material will be measured by ICP-OES (for sulfur) and graphite furnace atomic absorption (GFAA, for arsenic). Batch studies will be done to determine the leaching of AB9 from the Si-MNP and then to determine the arsenic capacity of the material. The silica-coated surface will be characterized post-metal encapsulation to investigate the possible interaction between the silica surface and arsenic. Optimization of the silica surface, amount of free amine capable of bonding to AB9, and the nature of arsenic bonding will be studied. Finally, AB9-Si-MNPs will be used to remove arsenic from water in a laboratory test, then separated magnetically from the treated water.

Principal Findings and Significance

The new AB9-Si-MNP composite material will be a potential replacement for conventional silica-based processes for the design of water filtration materials. The removal of arsenic(III) from contaminated water with subsequent magnetic separation will provide new possibilities for the treatment of water, particularly when large quantities of water need to be treated quickly and with no residual treatment agent present in the cleaned water.

The MNPs have been prepared, coated with silica, and derivatized with the “linker” that will connect the MNP to the AB9 arsenic-capture agent. The remaining goals to be achieved include: 1) attachment of AB9 to the MNPs, 2) demonstration of complete arsenic(III) removal through magnetic separation, and 3) complete characterization of the ultimate MNP-Si-AB9-AsOH composite.

Step 1) described above is currently being conducted and the research should be completed by July 1, 2012. Step 2) should be completed two weeks thereafter, with the remainder of the summer occupied with obtaining the characterization of the MNP-Si-AB9-AsOH composite. Project activities are anticipated to be complete by August 15, 2012.

ssion in longear sunfish (*Lepomis megalotis*) and green sunfish (*Lepomis cyanellus*) as a biomarker of polychlorinated biphenyl and metal exposure

Use of gene expression in longear sunfish (*Lepomis megalotis*) and green sunfish (*Lepomis cyanellus*) as a biomarker of polychlorinated biphenyl and metal exposure

Basic Information

Title:	Use of gene expression in longear sunfish (<i>Lepomis megalotis</i>) and green sunfish (<i>Lepomis cyanellus</i>) as a biomarker of polychlorinated biphenyl and metal exposure
Project Number:	2011KY213B
Start Date:	1/1/2012
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 5th and 6th
Research Category:	Biological Sciences
Focus Category:	Toxic Substances, Water Quality, Surface Water
Descriptors:	None
Principal Investigators:	David Peyton, Ben Brammell

Publications

There are no publications.

Use of Gene Expression in Longear Sunfish (*Lepomis megalotis*) and Green Sunfish (*Lepomis cyanellus*) as a Biomarker of Polychlorinated Biphenyl and Metal Exposure

Project 2011KY213B was delayed in starting. No significant progress was completed during FY 2011. As a result, the project report will be included in the FY 2012 Annual Technical Report. The abstract of the project follows:

Abstract: Recent studies have described widespread incidences of anatomical and physiological alterations in fish linked to waterborne pollutants. Observed alterations include occurrence of intersex fish, abnormal hormone levels, reduction in reproductive success, the presence of tumors, and abnormal liver to body weight ratios. Although such studies provide great insight into the nature of contaminant effects, they are expensive, time consuming, and require specialized techniques. Monitoring the expression of biomarkers provides an alternative mechanism of contaminant detection that offers a number of advantages. This study will evaluate the use of eight biomarkers at the level of mRNA expression in two species of fish widely distributed throughout North America. The project will obtain complete sequences for eight biomarker genes (cytochrome P4501A, superoxide dismutase, metallothionein, glutathione, peroxidase, catalase, glutathione S-transferase, aromatase CYP19, and vitellogenin) from both longear and green sunfish. These sequences will be submitted to GENbank as soon as they are obtained and should be globally available to all researchers even before the completion of the analysis of expression and the preparation of the final project report.

Information Transfer Program Introduction

Information transfer activities are an important part of the overall program of the Kentucky Water Resources Research Institute. There are two main components, an annual symposium and the institute web sites. The institute also participates in and supports numerous other technology and information transfer activities throughout the year.

Kentucky information transfer activities

Basic Information

Title:	Kentucky information transfer activities
Project Number:	2011KY176B
Start Date:	3/1/2011
End Date:	12/31/2012
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Not Applicable
Focus Category:	None, None, None
Descriptors:	annual symposium, web site
Principal Investigators:	Lindell Ormsbee, Anna Goodman Hoover, Jim Kipp

Publications

1. Proceedings 2012 Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, 102 p.
2. Proceedings 56th Annual Midwest Ground Water Conference, Kentucky Water Resources Research Institute, Lexington, Kentucky, 30 p.

Kentucky Information Transfer Activities (2011KY176B)

Problem and Objectives

The Water Resources Research Act requires that Institutes or Centers shall:

- (1) plan, conduct, or otherwise arrange for competent applied and peer reviewed research that fosters –
 - (A) improvements in water supply reliability;
 - (B) the exploration of new ideas that –
 - (i) address water problems; or
 - (ii) expand understanding of water and water-related phenomena;
 - (C) the entry of new research scientists, engineers, and technicians into water resources fields; and
 - (D) the dissemination of research results to water managers and the public.
- (2) cooperate closely with other colleges and universities in the State that have demonstrated capabilities 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.

Each institute shall also cooperate closely with other institutes and other organizations in the region to increase the effectiveness of the institutes and for the purpose of promoting regional coordination.

Kentucky information transfer activities are conducted in support of these objectives.

Methodology

Information transfer activities are an important part of the overall program of the Kentucky Water Resources Research Institute. There are two main components, an annual symposium and the institute web site (including the electronic newsletter). The Institute also participates in and supports other technology and information transfer activities throughout the year.

The Associate Director develops the program for the Annual Water Resources Symposium. Presentations in both platform and poster format allow for researchers and practitioners to share progress on planned, ongoing, and completed water-related activities throughout the Commonwealth each year.

The Information Specialist Senior assists with creating program announcements and the proceedings volume for the symposium. She also prepares information for the electronic newsletter. She develops and maintains content for several web sites including the main Institute page at: ww.uky.edu/WaterResources/. Links for additional sites describing projects and activities (for example volunteer sampling results and watershed

pages for the Kentucky River basin) are provided on the main web site. Research translation to make results accessible for a variety of audiences is a major goal for all of the technology transfer activities of the unit.

The Institute cooperates closely with other groups and agencies in planning additional technology transfer activities in the Commonwealth. These efforts included support for seminar/lectures, other web sites, an open house during Earth Science Week, and a weeklong summer camp for high school sophomores from eastern Kentucky counties. Institute staff members serve a variety of support roles on technical committees and advisory panels for agencies and volunteer organizations to help disseminate relevant information about ongoing activities and research results;

Principal Accomplishments and Activities

The Kentucky Water Resources Research Institute at the University of Kentucky was designated as a Center of Excellence for Watershed Management by Region IV of the U.S. Environmental Protection Agency on March 21, 2011. With this designation, KWRRRI became the first such Center of Excellence in Kentucky and the seventh in the Southeast. These centers help communities identify watershed-based problems and assist with the development and implementation of locally sustainable solutions. To become a Center of Excellence, an institution must demonstrate technical expertise in identifying and addressing watershed needs; involvement of students, staff, and faculty in watershed research; the capability to involve the full suite of disciplines needed for all aspects of watershed management; and a willingness to partner with other institutions.

Kentucky Water Awareness Month is an educational program of the University of Kentucky Cooperative Extension Service, Environmental and Natural Resources Issues Task Force (the Associate Director of KWRRRI is a member). The program promotes overall water awareness for citizens of Kentucky during May each year. Materials are developed by a committee at the state level and distributed to all of the 120 county extension offices in the state. Individual county agents are encouraged to tailor the program to fit their county's specific needs and to use the materials to enhance their program efforts. The materials remain available throughout the year for use by classroom teachers, 4-H volunteers, and others interested in water issues through the ENRI internet site: www.ca.uky.edu/enri/

The Robinson Scholars Program serves first generation college-bound students from 29 eastern Kentucky counties who have demonstrated the potential to succeed, but who might encounter social, economic, cultural, or institutional impediments to completion of a four-year college degree. The program provides general support, leadership development opportunities, and a University of Kentucky scholarship upon graduation from high school. The Water Pioneers Water Quality Initiative was developed by KWRRRI for rising high school sophomores in the program. It is held for 5 days in June and immerses the teens in activities designed to open their eyes to the importance of healthy watersheds using a diverse curriculum designed to show nature's

interconnectivity. Following the camp, the students use knowledge that they gain to partner with educators, volunteers, and other interested groups in their home counties to increase awareness of best management practices for water quality through a community service/outreach project of their own design.

KWRRI co-sponsored the 56th Midwest Ground Water Conference (and two associated field trips) with the Kentucky Geological Survey and the Kentucky Division of Water on September 19-21, 2011 in Lexington, Kentucky. The Associate Director of the Institute coordinated the submission of all abstracts for the program, arranged session moderators for oral presentations, and created the proceedings volume for the meeting:

<http://kgs.uky.edu/kgsweb/MWGW/MWGWProceedings.pdf>

The Kentucky Stormwater Association Annual Conference was held September 27-29, 2011 in Frankfort, Kentucky. Through a grant obtained from the Kentucky Division of Compliance Assistance of the Department for Environmental Protection, the KWRRI was able to underwrite the registration fee for one representative from each Municipal Separate Storm Sewer System (MS4) community in Kentucky to attend the annual KSA conference.

The Kentucky Water Resources Research Institute and the University of Kentucky Department of Earth and Environmental Sciences co-sponsored the National Ground Water Association Henry Darcy Distinguished Lecture “Development of Reliable Hydrologic Data Sets in Difficult Environments: Case Studies from Benin, West Africa” by Dr. Stephen Silliman on September 29, 2011.

An open house was held on Wednesday evening 10/5/2011 prior to Earth Science Week. This event was co-sponsored with the Kentucky Geological Survey. KWRRI staffed a water exhibit for the elementary, middle school, and high school students and their parents who attended the event (approximately 200 people).

KWRRI served as a co-host for the University of Kentucky Environmental Seminar Speaker Series on the evening of November 3, 2011. Peter H. Gleick, Co-Founder and President of the Pacific Institute presented “New Thinking for Water in the 21st Century.” Additional sponsors included the University of Kentucky College of Agriculture, Cooperative Extension Service Environmental & Natural Resources Initiative, the Tracy Farmer Institute for Sustainability & the Environment, and the UK Office of the Vice President for Research. This presentation was free and open to the public. Earlier in the day, faculty and research staff members were invited to Environmental & Sustainability Summits including “Perspectives on Sustainability” and “Sustainable Cities.”

The Virginia Water Resources Research Center, the West Virginia Water Research Institute, and the Kentucky Water Resources Research Institute co-hosted a 2011 Water Research Symposium “Coal & Water in Central Appalachia: The Challenge to Balance” in Blacksburg, Virginia on November 15, 2011. The KWRRI Associate

Director served as moderator for the panel discussion session at the conclusion of the conference.

Cyberseminars provided through the Consortium for the Advancement of Hydrologic Sciences, Inc. were made available by KWRRI on the University of Kentucky campus for interested faculty, staff, students, and local professionals.

The Kentucky Water Resources Annual Symposium was held on March 23, 2012. Although the date of the symposium fell outside of FY2011, most of the planning and preparation occurred during the fiscal year. The conference opened with a plenary session on “Water Research Needs in Kentucky” with perspectives provided by representatives from the Kentucky Division of Water, the USGS Kentucky Water Science Center, the KWRRI, and the University of Kentucky Department of Earth and Environmental Sciences. Two concurrent sessions provided time slots for 28 additional oral platform presentations. Twenty-two posters were also presented during a separate poster session. Approximately 130 people attended the meeting. Abstracts for all of the presentations were distributed to participants on the day of the meeting: Proceedings of the Kentucky Water Resources Annual Symposium, 2012, Kentucky Water Resources Research Institute, Lexington, Kentucky, 102 p. The full proceedings document is also available free of charge online through a link on the institute web site. The document includes contact information for all authors and presenters and an abstract for each presentation. Symposium participants also receive a list of attendees providing basic contact information for each individual who pre-registered for the symposium. Attendees include researchers, personnel from local, state, and federal agencies, undergraduate and graduate students, participants from volunteer groups and NGOs, and members of the general public. Conference registration fees are kept low through partial subsidy of symposium expenses (using 104(b) technology transfer and matching funds) to ensure accessibility to individuals from all potential audiences.

Maintenance of the institute web site provides open access for those interested in the activities of the Institute as well as providing links to related sites and information maintained by others. Creation and maintenance of the web site are ongoing throughout the year. Links on the site provide direct access to the Association of State Dam Safety Officials, the Kentucky Research Consortium for Energy and the Environment, the Kentucky River Watershed Watch Sampling Database, the National Institutes for Water Resources, PRIDE, the UK Superfund Basic Research Program Research Translation Core and the Kentucky River Watershed page.

USGS Summer Intern Program

None.

Student Support					
Category	Section 104 Base Grant	Section 104 NCGP Award	NIWR-USGS Internship	Supplemental Awards	Total
Undergraduate	2	0	0	0	2
Masters	7	0	0	0	7
Ph.D.	2	0	0	0	2
Post-Doc.	0	0	0	0	0
Total	11	0	0	0	11

Notable Awards and Achievements

2011KY174B - A Western Kentucky University FUSE grant was awarded to support the computational efforts associated with bringing this work to peer-reviewed publication. An NSF CAREER proposal will also be submitted this summer based upon the preliminary data acquired as a direct result of the funding of this project.

2011KY169B - A University of Kentucky, Department of Earth and Environmental Sciences Fern Research Grant (\$600) was received July 2011 to support this effort. A Geological Society of America, Southeastern Section, Graduate Research Grant (\$400) was also received to support this research effort.

Publications from Prior Years

1. 2010KY148B ("Filtration column for the covalent removal of arsenic from water") - Dissertations - Jana, Partha, 2012, Solid supported reagents for the removal of arsenic (III) from water, PhD Dissertation, Department of Chemistry, College of Arts and Sciences, University of Kentucky, Lexington, KY, 256 p.