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Kentucky Water Resources Research Institute Annual Technical Report FY 2010

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Kentucky Water Resources Research Institute, University of Kentucky

Kentucky Water Resources Research Institute
Annual Technical Report
FY 2010

Introduction

The 2010 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.

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. The National Institute of Environmental Health Sciences supported research translation activities through the Superfund Public Outreach Program. The Kentucky Department for Environmental Protection supported 3 students through an Environmental Protection Scholarship Program coordinated by the Institute. The Division of Compliance Assistance funded support for Phase II stormwater communities.

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 undertaken by KWRRI during 2010-2011 involved gathering input from stakeholders regarding potential future uses of the property associated with the plant.

Twelve student research enhancement projects were selected for support through 104(b) FY2010 funding. Projects were conducted at the University of Kentucky (7), Northern Kentucky University (2), Eastern Kentucky University (2), and Western Kentucky University (1). Projects represented a variety of discipline areas including civil engineering (3), chemistry (3), biology (3), geology (2), and plant and soil science (1). 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. Many state agencies are experiencing a significant loss of personnel through retirement and it is critical that undergraduate and graduate students are well trained and available to help fill this void. Project completion synopses for the 12 student research enhancement projects follow. All projects reported their results at the Kentucky Water Resources Annual Symposium on March 21, 2011. An additional project initiated with FY 2009 funds to assess volunteer sampling data from the Kentucky River basin was also completed and a synopsis of that project is also included in this annual report.

Ten year assessment of the Kentucky River Watershed Watch Program

Basic Information

Title:	Ten year assessment of the Kentucky River Watershed Watch Program
Project Number:	2009KY133B
Start Date:	3/1/2009
End Date:	6/30/2010
Funding Source:	104B
Congressional District:	Kentucky 5th and 6th
Research Category:	Water Quality
Focus Category:	Surface Water, Hydrogeochemistry, Water Quality
Descriptors:	
Principal Investigators:	Jim Kipp

Publications

1. Akasapu, Madhu and Lindell Ormsbee, 2010, Relatonship Between Fecal Coliform and E. coli Values within the Kentucky River Basin, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 89-90.
2. McAlister, Malissa, Madhu Akasapu, Ben Albritton, Lindell Ormsbee, and Dan Carey, 2010, Kentucky River Watershed Watch: A Summary of Volunteer Water Quality Sampling Efforts in the Kentucky River Basin from 1999 to 2009, Kentucky Water Resources Research Institute, Lexington, Kentucky, 42 p. http://www.uky.edu/OtherOrgs/KRWW/KRWW_Ten_Year_Report_Final_Version.pdf

Ten-Year Assessment of the Kentucky River Watershed Watch Program

Problem and Research Objectives

The Kentucky River Watershed Watch (KRWW) organization was formed in 1997 to enable citizen volunteers to sample local waterways within the Kentucky River Basin and learn how to improve and protect water quality. KRWW has grown to include approximately 250 volunteers living throughout the basin, which extends over much of the central and eastern portions of the state and is home to over 710,000 Kentuckians. The watershed includes all or parts of 42 counties and drains over 7,000 square miles, with a tributary network of 15,000 miles.

Since the inception of KRWW, the Kentucky Water Resources Research Institute has assisted its volunteers with data analysis by producing a yearly summary report of water quality sampling results. A five-year analysis was completed in 2004. With the accumulation of over 10 years of sampling data, a longer term analysis and summary of KRWW sampling results through 2009 was possible to assist the organization and its volunteers by providing further interpretation. Additionally, a survey of KRWW leadership and volunteers and their resulting recommendations helped strengthen the organization and helped participants better achieve their overall mission and goals.

This project compiled and statistically analyzed KRWW's water quality data collected between 1999 and 2009 (NOTE: Data for 11 years). The resulting summary report, titled *Kentucky River Watershed Watch: A Summary of Volunteer Water Quality Sampling Efforts in the Kentucky River Basin from 1999 to 2009,* was distributed to KRWW volunteers via face-to-face meetings and through electronic methods (e-mail and online posting).

http://www.uky.edu/OtherOrgs/KRWW/KRWW_Ten_Year_Report_Final_Version.pdf

The report includes 1) a short history of the KRWW organization; 2) an overview of sampling efforts; 3) an analysis of the annual spring, summer and fall sampling events; 4) field chemistry results; 5) a list of sampling sites and watersheds of concern; 6) how volunteers can apply their data; and 7) an assessment of the current status of the KRWW organization. The report also includes appendices describing individual water quality parameters and instructions for using the interactive, online KRWW database.

Methodology

A Microsoft Access database and Microsoft Excel were used to compute statistical analyses of KRWW water quality data. For most parameters, geometric means were calculated to determine averages for the assessment period when at least three separate results were available. In order to interpret these results, graphs were created that compared the geometric means to specific water quality standards established by the state of Kentucky to protect waterways for aquatic life and human uses.

The summer pathogen samples were analyzed for fecal coliform from 1999-2007 and for E. coli from 2008 to 2009. In order to compare the sampling results over the entire period, a statistical analysis was conducted to determine a relationship between the two pathogen indicators. Once this relationship was established between fecal coliform and E. coli, a t-test was used to determine if a sampling site showed improvement (decreased pathogen levels) by comparing the 1999-2003 five-year time period with the 2004-2009 six-year time period. To

perform this test, available data were converted to natural logarithms and the respective t-test statistic was examined to evaluate if changes were statistically significant. Unfortunately, several sampling sites lacked sufficient data for such a comparative analysis.

A KRWW volunteer survey was conducted in 2009 using an online service. This survey assessed opinions on the importance of specific KRWW functions (general coordination, monitoring, training, volunteer events, subwatershed projects, and advocacy) and how well they were being performed. The survey results were compiled in an Excel spreadsheet and summarized in narrative format.

Overall data interpretations were summarized in a narrative final report, along with appropriate tables, graphs and maps. Arcview GIS applications were used to produce the maps for the report. The report, as well as the full KRWW database, is posted on the KRWW website at www.krww.org for public access. The report is also available by a link from the KWRRI web site.

Principal Findings and Significance

In 1999, Kentucky River Watershed Watch volunteers sampled at 87 sites throughout the basin. After reaching a high of 248 sites in 2006, the number of sites decreased to 171 in 2009. Sampling has become increasingly more concentrated in the central region of the Kentucky River Basin, which includes the more densely populated, urbanized areas. Thus, there are currently more KRWW data results for this central region than for the upper basin areas in southeastern Kentucky or the lower basin in northern Kentucky. Figure 1 illustrates the changes in sampling site distribution from 1999 to 2009. In 2009, there were no sites located in the South and Middle Forks of the Kentucky River and only a few in the northern portion of the Kentucky River Basin.

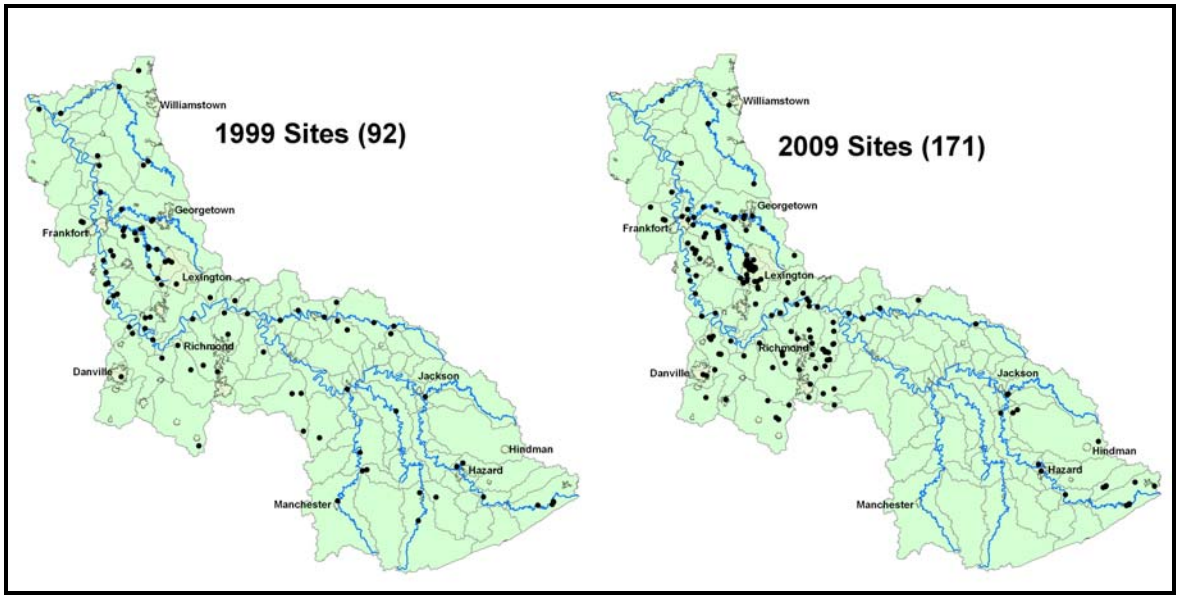

Figure 1. Comparison of 1999 and 2009 KRWW Sampling Locations

KRWW volunteers typically sampled the water quality at their chosen sites three to four times each year. The sampling events included herbicides and an insecticide (spring), synoptic

and follow-up pathogens (twice in summer), and metals, chemistry, and nutrients (fall). Pesticides were analyzed during the spring sampling event due to the increased likelihood of recent crop applications. Pathogens were assessed during the summer months, when people were more likely to be coming in direct contact with waterways through various recreational activities. The nutrient, chemical and metal parameters were analyzed during the fall water sampling event because of the typically lower flows observed during this time of year and the associated potential for increased concentration. In addition, samplers analyzed dissolved oxygen, pH, temperature, conductivity, and habitat condition in the field during sample collection.

Figure 2 illustrates the number of samples collected during each major sampling event from 1999 through 2009. The number of herbicide and insecticide samples that were collected were relatively low, because they were generally only assessed at newly sampled sites. Follow-up pathogen sites included only those sites where pathogen concentrations exceeded the safe swimming standard during the initial synoptic sampling event. Metals were only assessed for specific sites during the fall, low-flow sampling events.

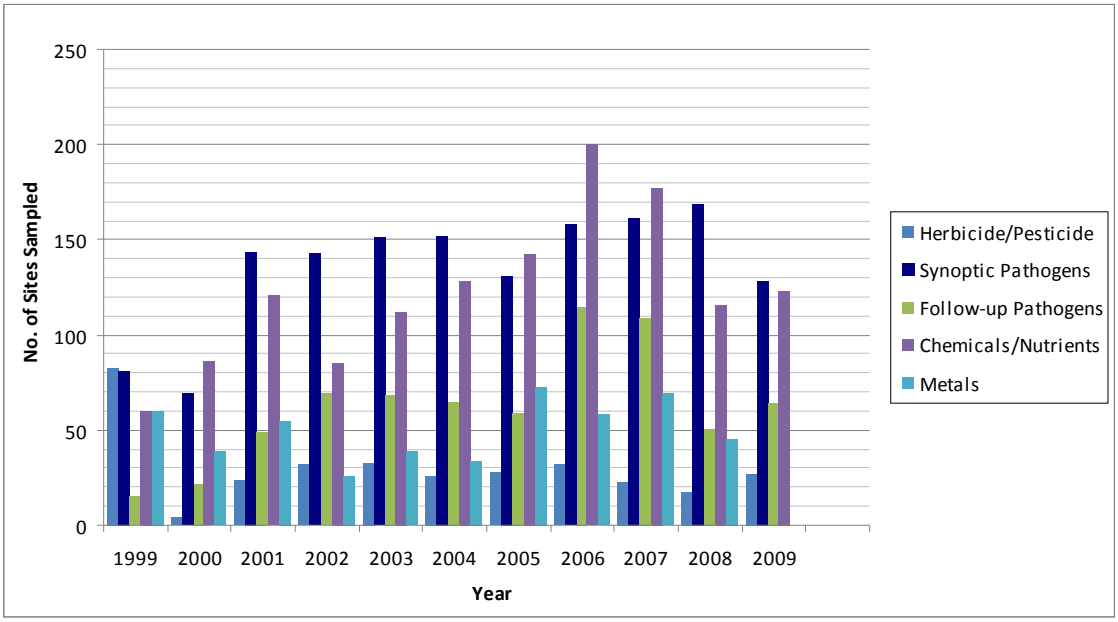

Figure 2. Number of Sites Sampled During Regular Sampling Events (1999-2009)
NOTE: No metals were assessed during 2009

Water quality results collected during each of the annual sampling events were entered into a Microsoft Access database and assessed. This assessment was based upon averaging sampling results for sites with data from at least three of the eleven sampling years for comparison with available water quality standards. The following water quality parameters were assessed: herbicides (Metolachlor, Atrazine, 2,4-D), insecticide (Chlorpyrifos), pathogens (fecal coliform and E coli), nutrients (nitrate nitrogen, total phosphorus, sulfate), metals (antimony, arsenic, barium, beryllium, cadmium, chromium, copper, iron, lead, nickel, selenium, silver, thallium, zinc), dissolved oxygen, pH, and temperature.

This analysis of eleven years of KRWW sampling data from 1999 through 2009 provided evidence of water quality concerns throughout the Kentucky River Basin. A list of sampling

sites and associated watersheds of concern was produced, along with a map showing their locations within the Basin.

In the future, this type of analysis could be strengthened by the availability of more continuous data for the individual sampling sites. By sampling regularly year-to-year, volunteers will be more likely to gain valuable insights to the current status and changes of water quality at their chosen sites. For many of the sampling parameters, water quality issues were most evident in the central region of the Kentucky River Basin. Although these findings are instructive, it should be noted that a disproportionate share of the KRWW sampling sites are located in this region.

This analysis of KRWW water quality results was intended to provide KRWW volunteers and organizers with information to guide continued sampling, focused sampling, and water quality improvement efforts. Additional sampling data from future sampling years will be useful to build on this analysis and strengthen its conclusions.

Identification of heavy metal sources in Wilgreen Lake, Madison County, Kentucky

Basic Information

Title:	Identification of heavy metal sources in Wilgreen Lake, Madison County, Kentucky
Project Number:	2010KY136B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Hydrogeochemistry, Toxic Substances
Descriptors:	limnology, contamination, lake sediments
Principal Investigators:	Walter S. Borowski

Publications

1. McMaine, Clint, and Walter Borowski, 2011, Patterns of Heavy Metal Concentration in Core Sediments, Wilgreen Lake, Madison County, Kentucky, in Proceedings of Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 81.
2. Von Gruenigen, Chad, and Walter Borowski, 2011, Concentration of Heavy Metals in the Waters and Surface Sediments of Wilgreen Lake, Madison County, Kentucky, in Proceedings of Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 83.

Identification of Heavy Metal Sources
In Wilgreen Lake, Madison County, Kentucky

Problem and Research Objectives

Wilgreen Lake was formed in the mid-1960's by damming Taylor Fork (Figure 1), which ultimately feeds Silver Creek and the Kentucky River. A cursory sampling of the lake in 2007 found elevated levels of cadmium, copper, lead, and nickel in surface and deeper waters that exceed standards for natural waters (401 KAR 10:031; EPA, 2003). Heavy metals are known to produce severe health problems in humans and other organisms (EPA, 2006a; see individual fact sheets) and are also known for their bioaccumulation properties (e.g., Eby, 2004), thus compounding their potentially damaging effects. Therefore it is crucial to document and understand the source and behavior of heavy metals within Wilgreen Lake.

If lake sediments are indeed the source of heavy metals, the lake and its biota should be monitored at regular intervals. Moreover, the sediments of the lake should remain undisturbed and unoccupied after the end of the lake's life in order to prevent remobilization of these sequestered heavy metals.

Methodology

We were able to take cores at two locations within Wilgreen Lake as planned, that is, from the levee systems of Old Town Branch and Taylor Fork (Fig 1). Both cores were measured and sectioned into pieces according to lithology, resulting in 31 and 34 sediment samples from Old Town Branch and Taylor Fork cores, respectively. These samples were digested according to established protocols (Method 3030, Eaton et al., 2005) that move trace metals into solution. Samples were then sent to Activation Laboratories for analysis of antimony, arsenic, cadmium, chromium, copper, cobalt, lead, selenium, silver, thallium, and thorium via using ICP/OES (Method 3120, Eaton et al., 2005).

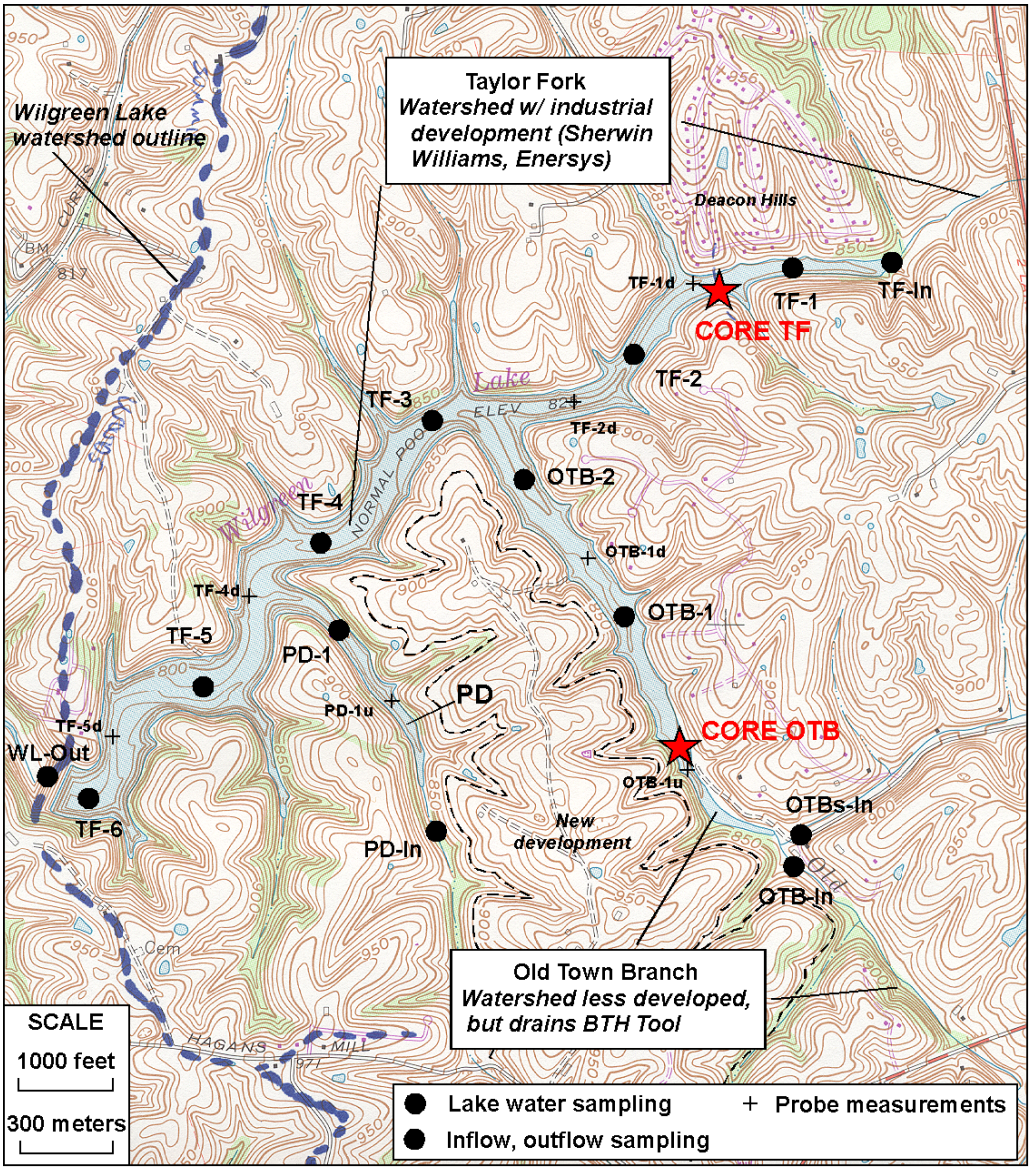

Figure 1. Map of Wilgreen lake with locations of coring sites and water sampling stations. Base map: Richmond South 7.5-minute topographic quadrangle map.

Principal Findings and Significance

(1) Core sediments are comprised of mud with or without plant material. There are also zones of black mud and zones with reddish-brown globules in core sediments (Figure 2).

(2) Most trace elements show no pattern with depth and, surprisingly, also seem unrelated to sediment lithology.

(3) Sb and Cd have concentrations equivalent with the method blank (Figure 3).

(4) Ar, Cr, Ag, Se, Tl, and Th show little change with core depth and no difference in baseline concentration between Taylor Fork and Old Town Branch (Figure 3).

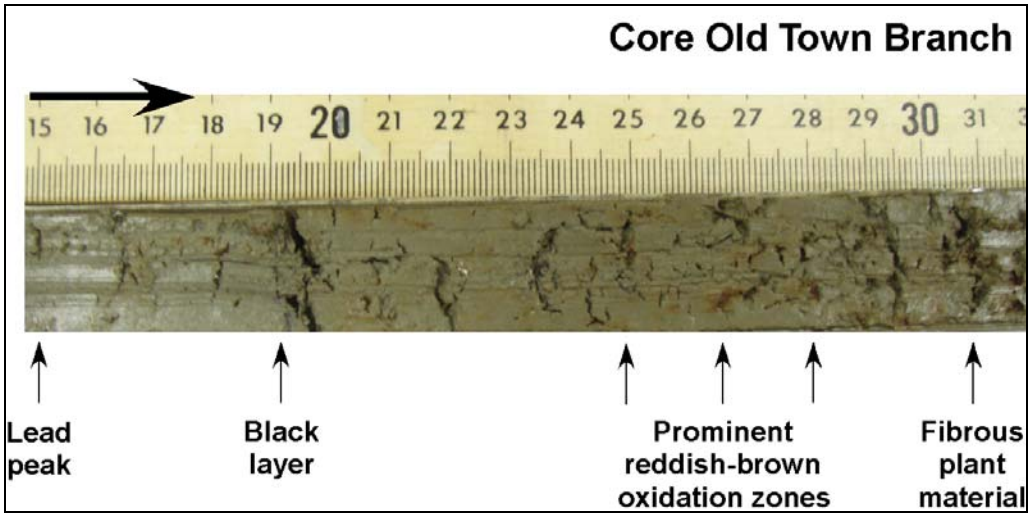

Figure 2. Photograph of a portion or Core OTB showing overall mud lithology, black zones, and reddish-brown oxidation zones.

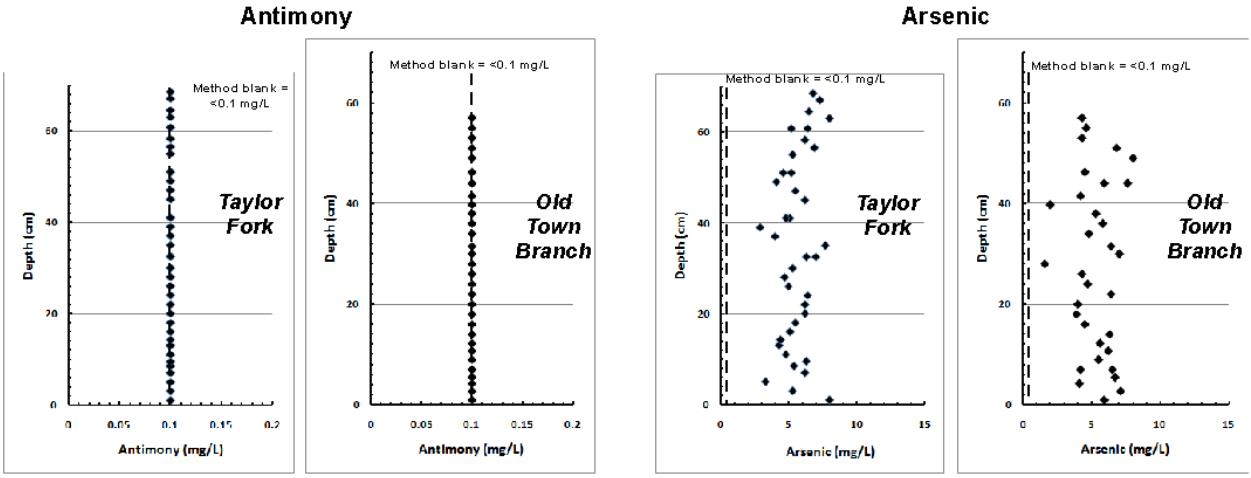

Figure 3. Trace metal concentration profiles of antimony and arsenic from sediments of both cores. Note the antimony values track the method blank, whereas arsenic values are well above the method blank but show no little variation at either site.

(5) Cu also shows little change with depth but baseline concentration at Taylor Fork is about 25% higher (Figure 4).

(6) The baselines for Ni and Cr are similar for each core locality, but values are higher in the upper third of the core at Taylor Fork, which also occurs for Cu and Pb values.

(7) The Pb concentration baseline is ~40% higher within Taylor Fork sediments (Figure 4).

(8) Higher baseline concentrations of copper and lead in the Taylor Fork core suggest a metal source in its watershed, or inputs from the residential area adjacent to the coring site.

(9) Trace metal increases in Co, Cr, and Pb observed in upper core sediments are consistent with increases in surface sediment levels at nearby stations M2 and TF-1, but Ni is not elevated in surface sediments and As is not elevated in core sediments.

(10) Patterns of trace elements within Wilgreen sediments at depth likely reflect diagenetic processes as well as deposition of trace elements. Wilgreen sediments serve as a reservoir for trace elements as evidenced by higher sediment versus water concentrations. Some metals (Co, Pb, Th) demonstrably enter anoxic lake waters from sediments, but other elements seem locked within sediments. Increases in Cr, Cu, Ni, and Pb at the top of the Taylor Fork core perhaps represent mobilization of metals from deep in the sediments within anoxic pore waters and upward movement to shallower depths. There, redox conditions promote precipitation of metals forming the peaks.

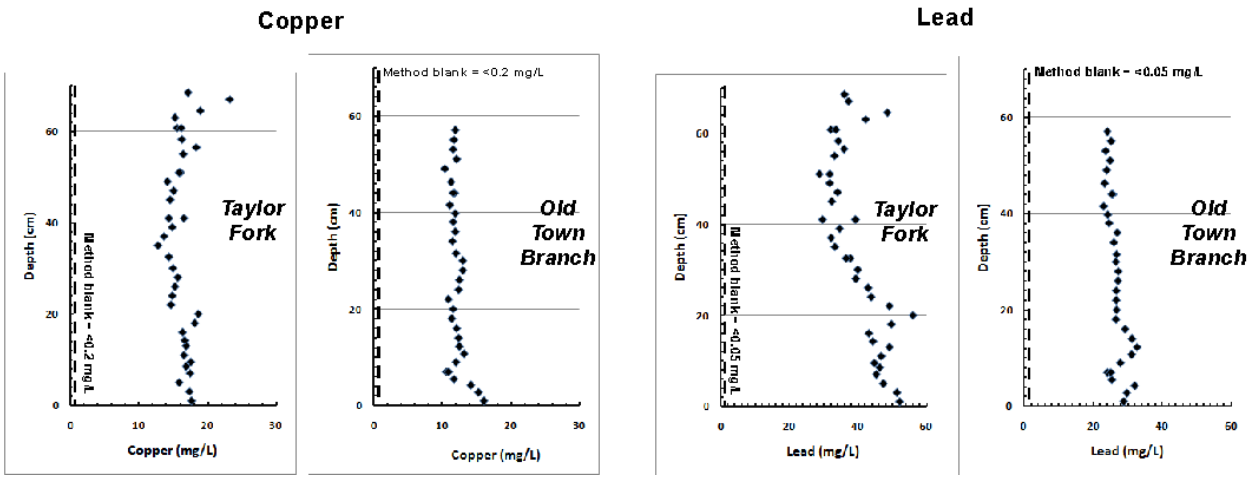

Figure 4. Trace metal concentration profiles of copper and lead from sediments of both cores. For copper, note the higher baseline value in the Taylor Fork core. Lead also shows a higher baseline concentration value in Taylor Fork sediments and also shows concentration peaks in the upper part of the core that may be due to diagenetic mobilization of lead from deeper in the core and precipitation higher in the core due to changing redox conditions.

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Method 3120, Metals by plasma emission spectroscopy. *In:* Eaton, A.D., L.S. Clesceri, E.W. Rice, A.E. Greenberg, (Eds.), 2005, Standard Methods for the Examination of Water and Wastewater (21st ed.). American Public Health Association, American Water Works Association, Water Environment Foundation. Baltimore: Port City Press, pp 3-38 to 3-44.

USGS 7.5 minute quadrangle topographic map series, Richmond South (37084-F3-TF-024), 1965. Photorevised in 1987.

A coupled hydrologic and biogeochemical modeling approach to understand if in-stream depositional zones are a carbon source or sink to the atmosphere

Basic Information

Title:	A coupled hydrologic and biogeochemical modeling approach to understand if in-stream depositional zones are a carbon source or sink to the atmosphere
Project Number:	2010KY137B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Climate and Hydrologic Processes
Focus Category:	Geochemical Processes, Hydrogeochemistry, Water Quality
Descriptors:	biogeochemistry, watershed, suspended sediment
Principal Investigators:	James F. Fox

Publications

1. Martin, Darren, and Jimmy Fox, 2011, Development of a Comprehensive Sediment Transport Method in First Order Watersheds with Contour Coal Mining, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 23-24.
2. Adams, Nathaniel, Darren Martin, and Jimmy Fox, 2011, Variability of Particulate Organic Carbon Flux with Mining in Small Appalachian Watersheds, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 93-94.
3. Acton, Peter, and Jimmy Fox, 2011, Long Term Estimates of Soil Organic Carbon and Nutrient Dynamics in Reclaimed Appalachian Mine Soil, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 95.
4. Martin, D., N. Adams, and J.F. Fox, 2011, Development of a Comprehensive Sediment Transport Method in First Order Watersheds with Contour Coal Mining, in Proceedings World Environmental & Water Resources Congress 2011: Bearing Knowledge for Sustainability, Palm Springs, California, May 22-26, 2011.
5. Martin, Darren, 2011, Development of a Sediment Transport Method for First-Order Watersheds with Mountaintop Coal Mining Disturbances, MS Thesis, Department of Civil Engineering, College of Engineering, University of Kentucky, Lexington, Kentucky.

A Coupled Hydrologic and Biogeochemical Modeling Approach to Understand if In-Stream Depositional Zones are a Carbon Source or Sink to the Atmosphere

Problem and Research Objectives

Recent global-scale carbon assessment of inland freshwater ecosystems have concluded that roughly half of the carbon that enters streams, rivers and lakes is not exported to estuaries and oceans, and one of the largest unknowns is the fate of carbon within small streams and watersheds (Cole et al., 2007). Kentucky's streams, rivers, and lakes have large amounts of sediment deposition where in-stream biogeochemical processes occur, which is a missing component of the carbon budget. For example in the Bluegrass region of central Kentucky it has been estimated that streambeds temporarily store 30.1 kg m^{-2} sediment, and associated with this sediment is 0.97 kg m^{-2} C, which is a substantial increase relative to the amount of carbon in watershed soils (0.485 kg m^{-2} C). The fate of the accumulated carbon is relatively unknown.

Improvement of coupled watershed models is needed because recent studies have significantly contributed to resolving the fate of suspended particulate organic matter (SPOM) carbon pools in large river systems, but much less can be extrapolated with regards to: (i) the flux of carbon associated with SPOM in small watersheds; (ii) the decomposition and outgassing of CO_2 within the streambeds of small streams; and (iii) the flux of carbon associated with the development and turnover of biomass within the small stream and watershed ecosystems.

The overarching goal of this research is to study the erosion processes that occur on the reclaimed coal mine surface soils, forest surface soils, and streambank soils and the transport of fine sediments from two watersheds that differ in mining disturbance histories in the Appalachian Forest Region of Southeastern Kentucky through the application of a refined sediment fingerprinting method using $\delta^{13}C$ and $\delta^{15}N$ and an erosion and sediment transport model. The specific objectives of the research were to formulate an accurate coupled model that accounts erosion and sediment flux; to collect field data that can be used as calibration tools for the model, and to apply the model to a set of watersheds to answer the questions surrounding hydrologic sediment transport rates, carbon and nitrogen decomposition rates, and growth rates.

Methodology

This study consisted of three primary components: (1) field sampling, (2) laboratory analysis, and (3) computer modeling and numerical analysis. Field sampling was completed for three summers at the field sites in Southeastern Kentucky. This sampling included the collection of source soil samples and sediment samples from four neighboring watersheds with differing disturbances. Sediment samples were collected weekly for the entire sampling routine. Laboratory analysis for tracer identification was completed in the Hydrosystems Laboratory at UK and the Environmental Research and Training Laboratory (ERTL) at UK. Computer modeling and numerical analysis was completed in the UK Student Computing Services Laboratories.

Principal Findings and Significance

A comprehensive sediment transport method was developed that provides sediment yields (tons/yr) from multiple erosion sources in 1st order watersheds with surface coal mining through the use of sediment fingerprinting and a sediment transport model. We found that $\delta^{13}C$ and $\delta^{15}N$ of sediment reflect the amount of disturbance and erosion sources for the watersheds, which is consistent with the literature. As carbon and nitrogen are cycled through plants and the soil, an organic "fingerprint" becomes bound to the soil particles due to isotopic fractionation. Figure 1 depicts the shift from 2007 to 2009/10 in the average carbon and nitrogen isotopic and elemental values. It was recommended that researchers use the $\delta^{13}C$ and $\delta^{15}N$ tracers to determine if the land-use is disturbed, allowing for a better understanding of the erosion processes occurring on the site. We found that $\delta^{13}C$ and $\delta^{15}N$ of sediment also reflect natural disturbance in a watershed, in our case the erosion of forest soils due to tree throw caused by severe ice and rain storms, which in turn increased forest surface soil erosion. This development will be useful when studying the effects of natural disasters that occur in a watershed.

Our advanced dual-isotope un-mixing method accounted for C and N un-mixing using $\delta^{13}C$ and $\delta^{15}N$, and it is recommended for future use. Sensitivity for hypothetical scenarios showed that C and N enrichment during transport and tracer non-conservativeness can vary sources percentages by ±10%. The sediment fingerprinting results were used to calibrate the sediment transport model, for the first time to our knowledge. Without the results from the dual-isotope un-mixing method, calibration of the sediment transport model would not be possible because the sediment yield from each sediment source was unknown. The dual-isotope un-mixing method provides a means to calibrate sediment transport models that do not have erosion measurements for the different sediment sources.

Figure 2 shows that the sediment yield from reclaimed surface mining sites decreased over time including 4.4, 2.1, and 0.8 t yr^{-1} ha^{-1} for 1½, 4 and 6½ years after mining, which is attributed to the reclamation practices that occur on the mined sites. Over time, vegetation growth decreases the effects of raindrop impact. The severe ice and rainstorms increased forest sediment production by 3.5 times from 2007 to 2009/10, and sediment yield increased from 0.08 to 0.29 t yr^{-1} ha^{-1} because the large root mass dislodges soil upon tree tip over. Change in the streambank sediment yield over time was not pronounced. Whitaker Branch bank sediment production was 5 times higher than Island Branch, which was due to the larger susceptible bank area and logging trucks driving across the stream.

Reference

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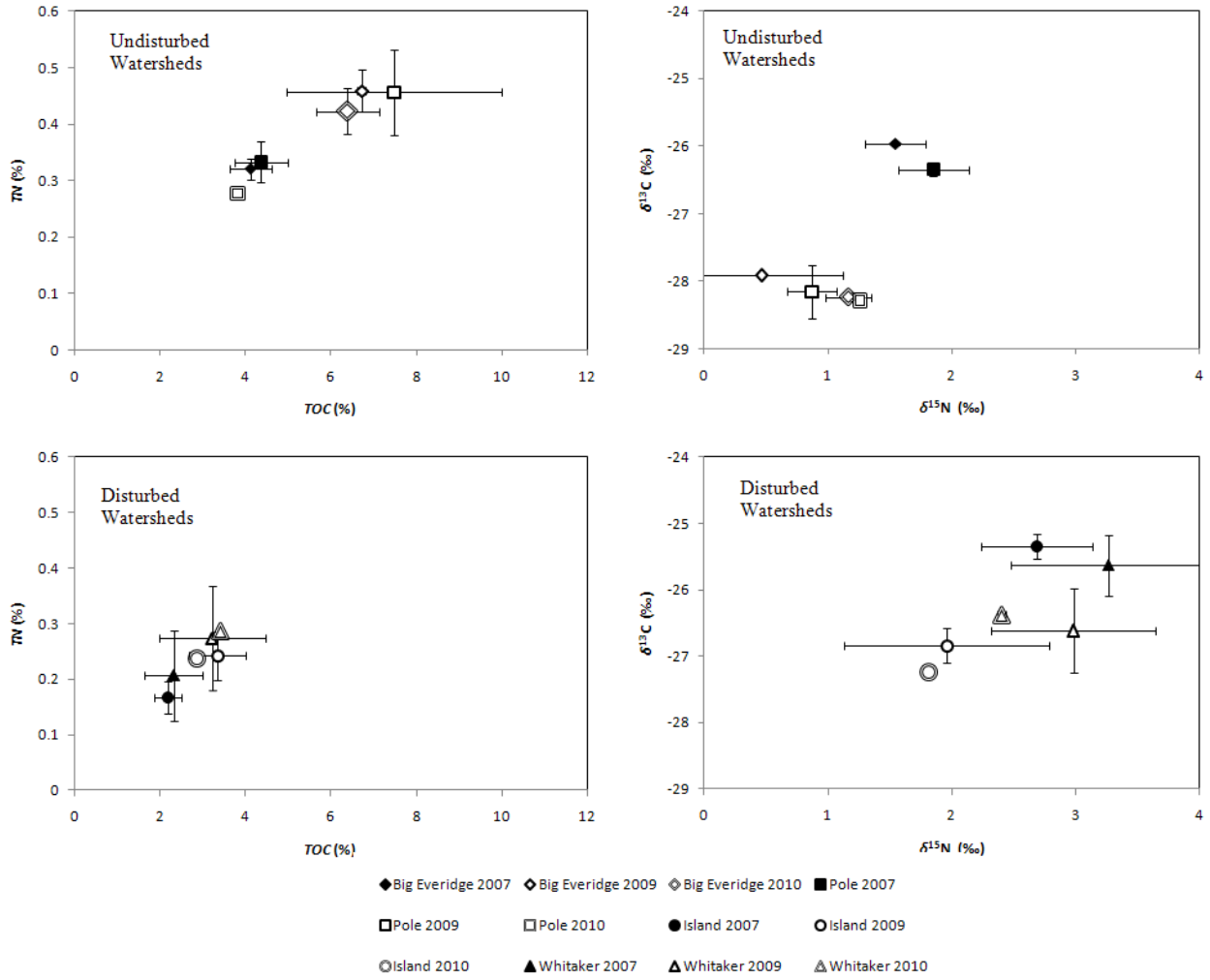

Figure 1: Average Carbon and Nitrogen Isotopic and Elemental Values of the Transported Sediments

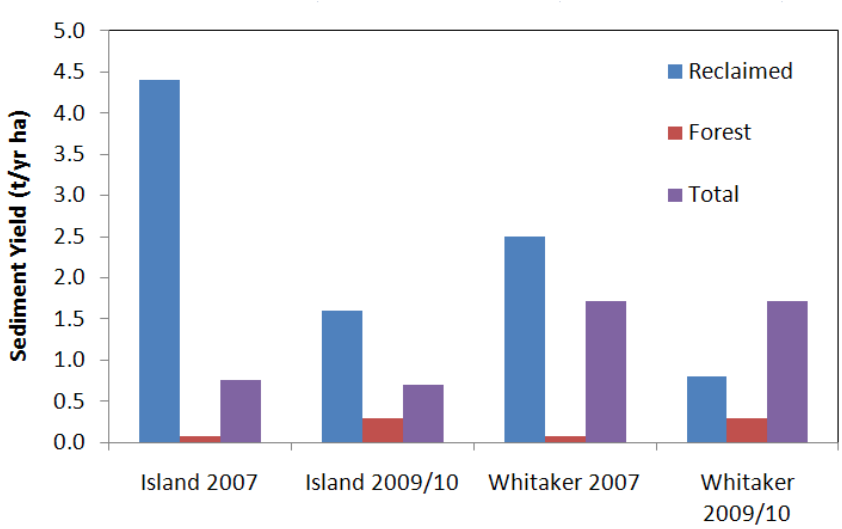

Figure 2: Annual Sediment Yield per Area for each Watershed (first bar reclaimed, second bar forest, third bar total)

Improving water quality: river channel versus riparian edge

Basic Information

Title:	Improving water quality: river channel versus riparian edge
Project Number:	2010KY138B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Fourth
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Nutrients, Wetlands
Descriptors:	nitrogen, aquatic systems, denitrification, sediment
Principal Investigators:	Kristine Hopfensperger

Publications

1. Kirtman, E.R., and K.N. Hopfensperger, 2010, Improving Water Quality: River Channel versus Riparian Edge, in Proceedings Ohio River Basin Consortium on Research and Education (ORBCRE) Annual Conference, Highland Heights, Kentucky.
2. Kirtman, E.R., and K.N. Hopfensperger, 2011, Improving Water Quality: River Channel versus Riparian Edge, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 69.

Improving Water Quality: River Channel versus Riparian Edge

Problem and Research Objectives

Our Nation's waterways and wetlands receive increased amounts of nutrients each year from agricultural and residential surface water runoff. Land managers and policy regulators have implemented a variety of best management practices; however, excess nutrients continue to facilitate the formation of hypoxic dead zones in water bodies every summer (Kemp et al. 2005). These low-oxygen zones kill many ecologically and economically important plant and animal species. One natural solution to excess nitrogen (N) in water is denitrification, the microbial conversion of water soluble nitrate to forms of N gas. Denitrification has been studied in a variety of aquatic systems, yet a direct comparison between river channel and riparian edge denitrification rates has yet to be completed. Understanding where denitrification rates are greatest can help managers maximize N removal from water before it moves downstream; thereby, minimizing hypoxic areas in the U.S.

Riparian wetlands sit at the interface between the sources of N-polluted surface runoff (i.e., farms and lawns) and waterways. In this scenario, riparian wetlands become the integral kidney of the landscape, removing excess nutrients through multiple mechanisms (including denitrification) before they inundate coastal waters. Therefore, as nitrate-rich water passes over wetland sediments, the nitrate concentration typically decreases. Several studies have shown the ability of wetlands to remove nitrogen including a study in an urban tidal freshwater marsh along the Potomac River (Hopfensperger et al. 2009). Denitrification rates suggested that N removal at the site could be substantial – approximately 14,600 kg of N over an annual timescale assuming no large seasonal variations (however rates may be much higher in spring and summer; Hopfensperger et al. 2009).

Investigations of N removal in river channel (Laursen and Seitzinger 2002) found lower denitrification rates in the river sediment than has been found in riparian soil studies. One would expect higher denitrification rates in the riparian wetlands because they contain the ideal conditions for denitrification including anoxia and high nitrate and organic carbon concentrations (Robertson and Groffman 2007). The variability of wetland surface elevation also allows for both oxic and anoxic zones in the sediment (McManus 1998). This allows for a tight coupling between the aerobic microbial process of nitrification, which converts ammonium to nitrate, and anaerobic denitrification, which releases the converted nitrate back to the atmosphere as N gas. On the other hand, sediment from the main river channel would have lower denitrification rates owing to the constant water inundation creating highly anoxic soils with no nitrification activity. In addition, riparian wetlands receive high organic carbon inputs from land-surface runoff and high plant productivity in the wetland that fuel the microbial processes, while channel sediment can lack such carbon inputs. Further work demonstrating the importance of nutrient removal in riparian wetlands along rivers is critical and will provide support for the preservation and restoration of these disappearing ecosystems. Indeed, Kaushal et al. (2008) found that restoration of floodplain wetlands near streams significantly increased denitrification rates.

The Licking River has its headwaters in the mountains of southeastern Kentucky and flows through rural areas towards its confluence with the Ohio River. This confluence is adjacent to both downtown Cincinnati, OH and Newport/Covington, KY. Urban areas have been known to influence water quality through increased suspended sediment and nutrient concentrations, which can greatly alter microbial activity and subsequent process rates (e.g., denitrification). In this study, we investigated the difference between channel and riparian denitrification rate along a rural to urban gradient on the Licking River, KY. We hypothesized that denitrification rates (i.e., nitrogen removal) would be higher at the riparian sites due to greater organic matter content in the sediment and greater variability in soil moisture. We also hypothesized that denitrification rates would be higher in rural areas due to greater organic matter and nitrate concentrations in the surface water runoff entering the system.

Methodology

Four paired sampling sites (sampling from the main river channel and the adjacent riparian area at each site) were located along the Licking River starting near the confluence with the Ohio River and continuing upstream into Pendleton County to capture the rural to urban gradient (n=8 sampling locations). Sampling sites were selected in coordination with the Licking River Watershed Watch (Marc Hult) and the Campbell and Kenton Counties Conservation Districts (Mary Kathryn Dickerson). We ended up with sampling sites in Newport, KY (a few hundred yards from the confluence), Visalia (downstream of a series of three very small communities), Morning View (Meyer; a rural, partially agricultural area), and Butler (an area with agriculture and few residences) (Figure 1). This sampling design allowed us to see how N concentrations and denitrification rates change in the riparian areas and river as the water makes its way from a rural area to a major metropolitan area.

Sediment samples were collected from each of the four paired sampling locations once per month from June 2010 through February 2011 (plan to continue sampling through May 2011). Three replicate sediment cores (5 cm diameter by 10 cm deep) were extracted from each location during each sampling event. We accessed the main channel from the river's edge and used a 10-foot long clay soil auger to obtain the sediment samples. In the field, the sediment cores were placed in a cooler and then returned to the laboratory and stored in a 4^{o}C cooler until processing. Soil cores from the riparian soil were taken with a 0.75-inch diameter soil probe.

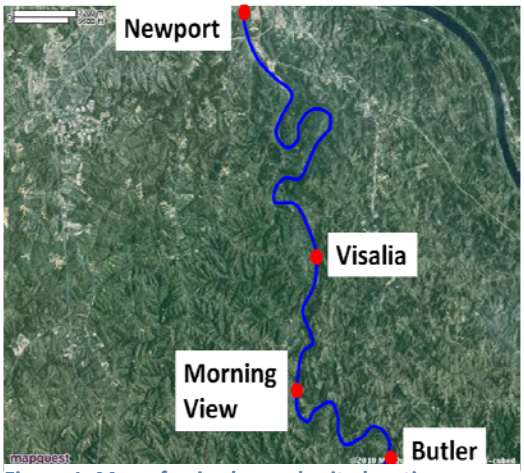

Figure 1. Map of paired-sample site locations

Sediment cores from each sample location were homogenized, processed, and analyzed for denitrification rate, soil organic matter content, and soil moisture in the laboratory. The denitrification enzyme assay (DEA) technique measures potential rates of denitrification (Smith and Tiedje, 1979) and was used to evaluate differences in denitrification rates between riparian and main channel sites. The technique allows for numerous samples to be analyzed at one collection time and followed the protocol of Groffman et al. (1999). The soil organic matter content for each site was measured using the loss on ignition

technique (Nelson and Sommers 1996). Soil moisture was measured following the protocol of Jarrell et al. (1999). Soil nitrate and ammonium concentrations were also measured from each sampling event by extracting the available inorganic N from the samples and analyzing the extracts on a microplate reader (Sims et al. 1995, Sims 2006). Two replicate water samples were collected from each paired sampling location during every sampling event. The water samples were analyzed for nitrate concentration using a microplate reader, so we could assess any influence of sediment nitrate removal on the water quality. We also wanted to see if any trends existed along a rural to urban gradient. Data was checked for normality and analyzed with repeated measures analysis of variance (RMANOVA) and linear regression using SPSS and SAS.

Principle Findings and Significance

Overall, inorganic N concentration did not differ between riparian soil and river sediment; however, when we looked at this comparison for each location, we found significant differences (Figure 2). Soil ammonium concentration differed between riparian soil and channel sediment ($F=14.3$, $p<0.001$); however, as with nitrate, significant interactions existed with sampling location and month. Channel sediment may have had higher ammonium due to water inundation inhibiting nitrification, the conversion of ammonium to nitrate. In fact, we found soil ammonium concentration increased significantly with soil moisture ($R^2=0.22$, $p<0.001$). At the same time, the channel sediment locations have less plant biomass taking up ammonium compared to riparian soil. Indeed, we found riparian soil to have greater soil organic matter concentrations than channel sediment in July (i.e., peak biomass time). The differences found between riparian and sediment samples among the locations provide confidence that we will find differences in denitrification rates.

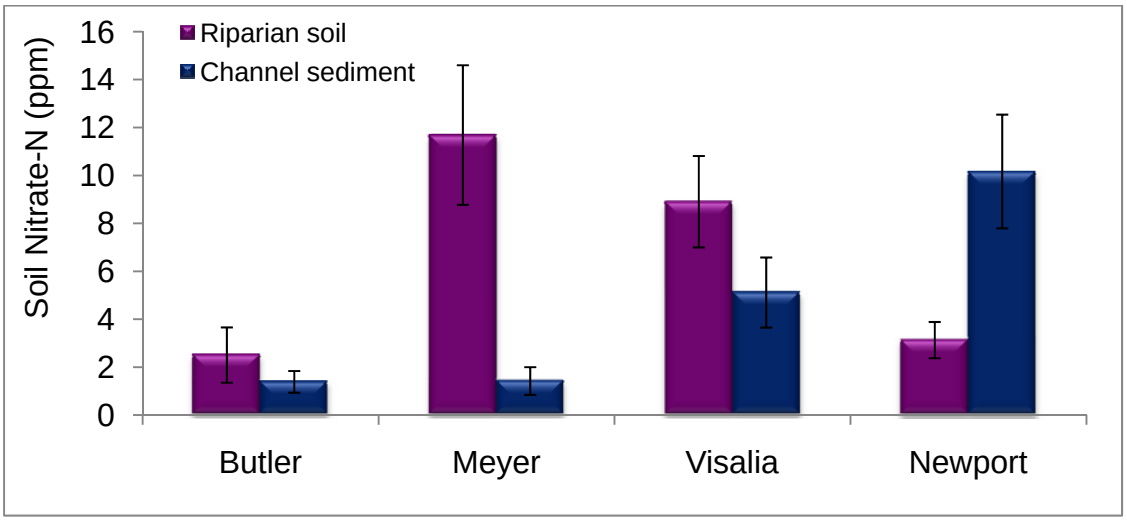

Figure 2. Soil nitrate concentration differed between riparian soil and channel sediment, but was dependent on location.

When combining riparian and channel nitrogen data and examining the rural to urban gradient, we found significant differences in soil N concentrations among sites during specific months (p=0.029; Figure 3).

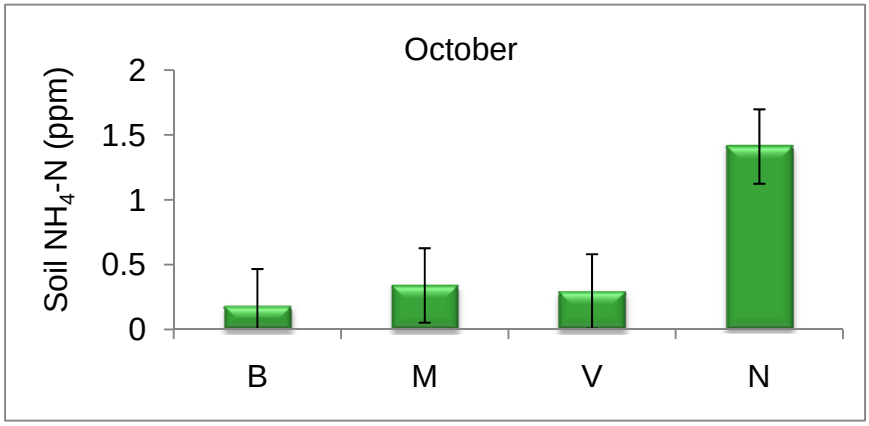

Figure 3. Soil ammonium concentration (riparian soil and channel sediment data combined) differed among sampling sites in October.

Water N-concentrations changed dramatically among sites and sample months. High concentrations were found more than once at the urban site (Newport) and sporadically at the most rural sites (Butler and Meyer). We found water nitrate and ammonium concentrations from June were greatest in rural areas, likely due to fertilizer runoff from agriculture. Further data analysis can be done to explore relationships between precipitation events, fertilizer application, and water N-concentrations. We also found water nitrate concentrations increased with soil nitrate concentrations during some sample months (November p=0.02), which lends support for the river water to be contributing nitrogen to the sediment. At the time of writing this report, we still plan two additional months of sampling, and have yet to finish analyzing the gas samples from the DEA technique. Our study will conclude in May and then we will evaluate all of the chemical and physical soil variables with the denitrification rates. The nutrient data presented here leads us to believe that we will have some interesting findings with the denitrification data. Understanding N-dynamics in riparian and in-channel areas will allow for better management to maximize N-removal from our water ways and reduce hypoxic zones downstream.

Acknowledgements

Thanks to Mary-Kathryn Dickerson from the Boone, Kenton, Campbell Counties Soil and Water Conservation District and Marc Hult for helping locate our sites. We would also like to give our thanks to the Thaxton's Canoe (the Butler site), Mr. Meyer (the Morning View site), and the Newport Rowing Club for access to the river.

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The carboxylic acid-bound iodine layer - towards an anti-fouling coating for water sensors and water treatment facilities

Basic Information

Title:	The carboxylic acid-bound iodine layer - towards an anti-fouling coating for water sensors and water treatment facilities
Project Number:	2010KY139B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Methods, Water Quality, None
Descriptors:	algal growth inhibitor
Principal Investigators:	Yuguang Cai

Publication

1. Lu, Lingbo, and Yuguang Cai, 2011, The Carboxylic Acid-Bound Layer - Towards an Anti-Fouling Coating for Water Sensors and Water Treatment Facilities, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 25.

The Carboxylic Acid-Bound Iodine Layer - Towards an Anti-Fouling Coating for Water Sensors and Water Treatment Facilities

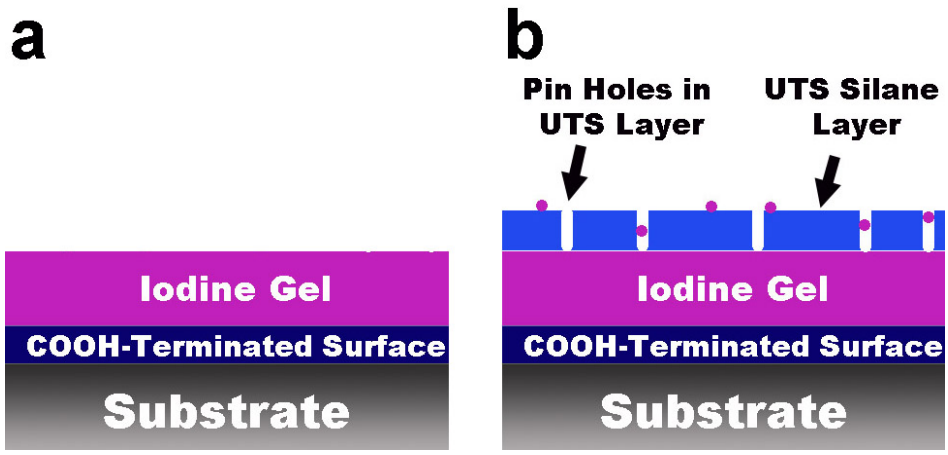
Problem and Research Objectives

Carboxylic acid (-COOH)-terminated group can hold iodine tightly on surfaces. Since iodine is an antiseptic agent, we proposed to use the iodine bound surface as an anti-fouling surface to prevent the growth of organisms (such as algae and bacteria). The iodine coating can protect pipe, sensors and other water treatment equipment.

Two objectives were proposed:

1) to characterize and adjust the iodine leaching of the COOH-bound iodine layer in water; and
2) to demonstrate the actual anti-fouling function of the iodine-bound surface.

Methodology



Scheme 1. Two proposed structures for the iodine-loaded surfaces

We proposed fabricating carboxylic acid-terminated silane surfaces and loading the surfaces with iodine. Then we characterized the iodine-loaded surface with infrared imaging (IR) and atomic force microscopy (AFM). Two types of surfaces were fabricated, as illustrated in Scheme 1. In the first type, iodine was directly loaded on the carboxylic acid-terminated surface (UTSoxI2, Scheme 1a). In the second type, an additional silane was coated on the iodine layer loaded on the carboxylic acid-terminated surface (UTSoxI2OTS, Scheme 1b). The iodine layer is sandwiched between the two silane layers. In this structure, the iodine is released to the environment through the pin-hole defects in the top silane layer. Therefore, the iodine leaching in the second type surface was expected to be much slower than the first type surface.

Next we used the iodine electrode to measure the amount of iodine leaching when the iodine-loaded surface is incubated in water over a period of time. For objective 2, we compared the actual algae growth and the *e. coli* growth on the iodine-loaded surfaces

with the iodine-free surface in order to test whether the iodine-loaded surface has an anti-fouling function.

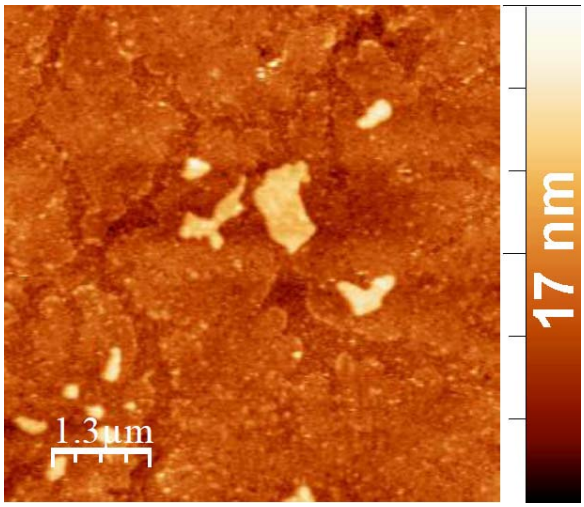


Figure 1. The AFM topography image of iodine-loaded surface. (UTSoxI2)

Principal Findings and Significance

1. Two types of designed iodine-loaded surfaces (UTSoxI2 and UTSoxI2OTS) have been fabricated. The Infra-red spectra for UTSoxI2 and UTSoxI2OITS have been obtained. The AFM surface microscopic images for these two surfaces have been acquired. Fig. 1. shows a representative surface. Now the surface structures of these two surfaces before and after iodine loading are known.

2. We measured the iodine leaching of the iodine-loaded surface. (objective 1). We

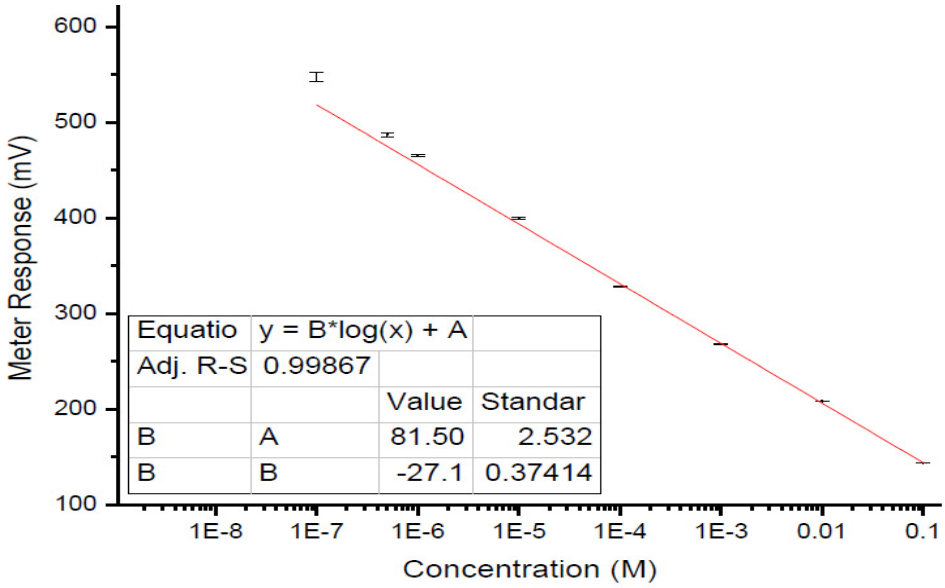


Figure 2. The obtained standard curve for the soluble iodine concentration measured by the Orion ion-selective electrode.

purchased an Orion iodide ion selective electrode, in order to measure the iodine concentration in the solution. The standard aqueous iodine concentration curve and electrode voltage was measured (Fig. 2). After incubating the iodine-loaded surface in water for a given amount of time, the iodine in the water was quantitatively converted to iodide. Then, we used the iodide electrode to measure the iodine concentration to quantify the iodine leaching of the iodine-loaded surface. We found that upon dipping the iodine-loaded surface into water, the amount of iodine released into water was below the

detection limit of the Orion iodide ion selective electrode. (leaching < 10^{-8} M/inch^2 surface for 3 day of incubation in water.)

3. We have accomplished objective 2, demonstrating the iodine-loaded surface indeed has an anti-fouling function. The anti-fouling function of these two iodine-loaded surfaces has been confirmed.

Here are our main findings:

a. We put a UTSoxI2 coated silicon wafer and a control silicon wafer (just UTSox) into a fish tank for 3 weeks. After rinsing, these two samples were examined using optical microscopy. For the control sample, the green algae cell density is 38±7/mm^2. In contrast, on the UTSoxI2 surface, the green algae cell density is 2±1/mm^2. The representative optical images are shown in Fig. 3. The iodine-loaded surface suppresses the growth of green algae on the surface.

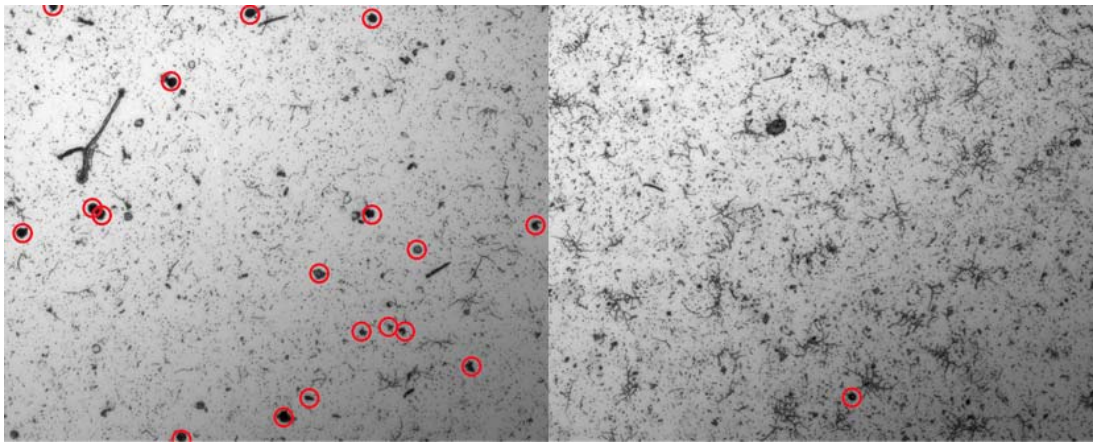
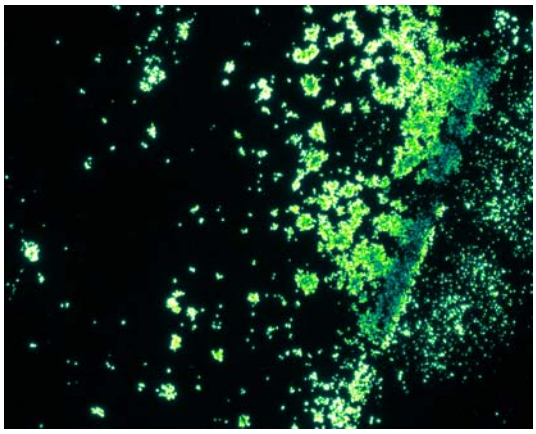
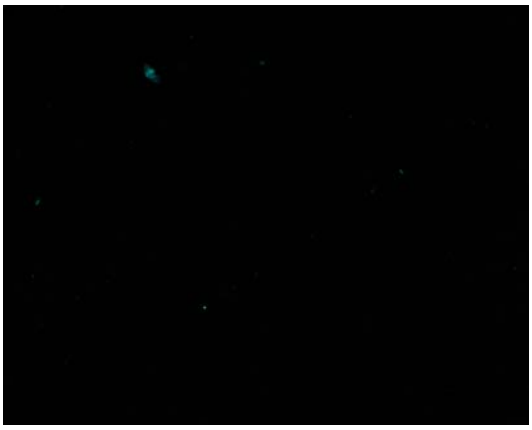


Figure 3. Left: The green algae (within red circles) on an iodine loaded sample. Right: The green algae on an iodine-free sample. Both samples were incubated in a fish tank for 3 weeks.

b. A UTSoxI2OTS wafer and a control OTS coated wafer were incubated with green fluorescent protein (GFP) labeled *e. coli*. for 72 hours at 37°C. The GFP is in the cytoplasm of the *e. coli.* After rinsing, both samples were examined by bright light optical microscopy and fluorescent microscopy. On the control OTS surface, the *e. coli* grew into micron-sized colonies, which is the precursor of bio-film (Fig. 4a). In contrast, on the iodine-loaded surface, *e. coli* is discretely distributed. Each *e.coli* cell exists as a single entity. No agglomeration of *e.coli* cells was found. Fluorescent microscopy reveals that the *e.coli* cells on the OTS surface are motile, which emit bright green light. If a cell is dead, the cell wall is broken. Thus the GFP in the cytoplasm flowed out and was rinsed off the surface. As a result, dead cells have very low fluorescence. The fluorescent microscopy on the UTSoxI2OTS surface reveals that >98% *e.coli* cells are dead (Fig. 4b).



a. Control Sample: Iodine-free OTS surface incubated in the GFP-labeled *e.coli* for 72 hours.



b. Iodine-loaded surface: UTSoxI2OTS surface incubated in the GFP-labeled *e.coli* for 72 hours.

Figure 4. a) and b) were both acquired with 300 ms exposure time. 500× optical image, 160×130 μm^2. Each green dot represents a motile *e.coli* cell.

Assessing short-term changes to headwater stream structure and function following alternative forest harvesting

Basic Information

Title:	Assessing short-term changes to headwater stream structure and function following alternative forest harvesting
Project Number:	2010KY140B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Fifth
Research Category:	Biological Sciences
Focus Category:	Surface Water, Water Quality, Ecology
Descriptors:	habitat, stream-side management zones, aquatic macroinvertebrates
Principal Investigators:	Scott Grubbs

Publication

1. Bergenson, Chad, and Scott Grubbs, 2011, Assessing Short-Term Changes to Headwater Stream Structure and Function Following Alternative Forest Harvesting Practices in a Cumberland Plateau Watershed, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 85-86.

Assessing Short-Term Changes to Headwater Stream Structure and Function Following Alternative Forest Harvesting

Problem and Research Objectives

Forested headwater stream systems are coupled tightly to the adjacent riparian forest (Grubbs and Cummins 1996) and downstream ecological processes (Vannote et al. 1980), and provide important habitat needs for a diverse suite of macroinvertebrate taxa (Lowe and Likens 2005, Grubbs 2011). Headwater systems are typically small, abundant globally, and comprise the majority of total stream length in most watersheds (Sidle et al. 2000, Richardson and Danehy 2007), yet are more prone to anthropogenic disturbance than larger systems and paradoxically can be inadequately managed (Gomi et al. 2002, Meyer et al. 2007).

Riparian zones, or stream-side management zones (SMZs), act as buffers and stabilizers to anthropogenic disturbance (Keller and Swanson 1979, Naiman and Decamps 1997) and regulators of energy and nutrient flow into headwater streams (Naiman et al. 1988). Riparian zones structure stream food webs and contribute to the productivity of streams, particularly in the form of leaf abscission (Cummins et al. 1989, Vannote et al. 1980, Wallace et al. 1999) in forested watersheds globally.

Forest harvesting can negatively influence the functionality of riparian areas. Stream response to harvesting varies regionally, and is affected by aspect, elevation, latitude, and the intensity and size of the disturbance. Harvesting has been shown to decrease leaf litter inputs (Hetrick et al. 1998, Kreutzweiser et al. 2004). This decrease can impart strong bottom-up effects and result in lower in-stream aquatic macroinvertebrate productivity (Wallace et al. 1999). Shredders, aquatic macroinvertebrates that feed directly on decaying leaf materials (Merritt and Cummins 2006), are directly affected by reductions in litter inputs (Stone and Wallace 1998). Productivity may decline (Gurtz and Wallace 1984, Wallace et al. 1999) or increase (Stout et al. 1993, Stone and Wallace 1998) following forest harvesting practices. The negative effects of harvesting can be either minimized or eliminated, however, if partial or total SMZ protection can be provided.

The purpose of this study was to assess macroinvertebrate community structure (taxa present) and functional feed group context between pre-harvesting (2004 and 2005) and short-term (< 18 mo.) post-harvest conditions (2010) of three distinct SMZ harvesting practices in a Cumberland Plateau watershed. Recent research within this watershed revealed diverse community structure and subtle upstream-downstream shifts (Grubbs 2011), providing an excellent pre-harvesting template for evaluating the short-term influence of SMZ harvesting.

Methodology

This project was performed in Clemons Fork, a 3rd-order Cumberland Plateau watershed located in the Kentucky River Basin and part of a series of Robinson Forest tracts (Fig. 1). Eight study subwatersheds draining into Clemons Fork were established as *a priori* replicates. Subwatersheds were similar according to drainage area (23.7–108.7 ha), drainage density (0.0032–0.0052 m/m^2), and elevation gradient (304–481 m). Streams draining each subwatershed were divided into intermittent and perennial channels based on prior work by Svec et al. (2003). Each channel was further subdivided into midstream and downstream reaches that were 50 m in length. Hence, four sets of reaches per stream were employed in this study. Stream substrates are mainly sandstone with interspersed coal seams and shale outcrops.

Stream macroinvertebrates were collected during spring (April) both prior to, and following, SMZ harvesting. Preharvest sampling occurred in 2004 and 2005. Starting in May 2008, SMZ harvesting treatments were applied across six experimental subwatersheds (Table 1). Treatments were applied differently adjacent to stream channels defined as intermittent (I) and perennial (P). Two subwatersheds served as controls and treatments were applied in replicate pairs. Post-harvesting sampling was carried out in to 2010.

All samples were taken from coarse substrate riffles with a Surber sampler (0.09 m^2 sampling area; 500-µm mesh net). Five replicate samples were obtained per reach. Riffles were preferred over bedrock runs and small pools because this habitat supports the greatest taxonomic richness and abundance of benthic macroinvertebrates (Brown and Brussock 1991). In the laboratory, each sample was sorted under a dissecting microscope. All identifications were taken to the lowest practical level, mainly to genus and to species when possible. To date, the only post-harvest samples that have been processed in full and identified pertain to the most comprehensive SMZ treatment in the Booker Fork and Shelly Fork North subwatersheds (Table 1). Identification of samples in the remaining subwatersheds is ongoing.

Midstream and downstream reach data of the intermittent and perennial channels were combined prior to analyses. Macroinvertebrate data were condensed to richness (number of unique taxa) and density (no./m^2) measures of seven variables: total community, the EPT index (Ephemeroptera [= mayflies], Plecoptera [=stoneflies], and Trichoptera [= caddisflies]), and five functional feeding group measures (filtering-collectors, gathering-collectors, scrapers, shredders, and predators). EPT taxa are indicative of healthy water quality conditions and are typically prone to anthropogenic disturbance, making this index a simple and broadly applicable measure of biotic integrity (e.g., Wallace et al. 1996, Barbour et al. 1999). Functional groups were assigned according to Barbour et al. (1999) and Merritt et al. (2008). The functional classification scheme permits whole assemblage assessment of the relative importance of specific classes of food resources (Merritt and Cummins 2006) and linkage to the adjacent

terrestrial landscape (e.g. degree of canopy openness). Three functional feeding group ratios were also calculated (Merritt and Cummins; Table 2). These were used to assess the relative importance of autotrophy or heterotrophy to stream energetics and the amounts of coarse- (> 1 mm) and fine- (< 1mm) sized organic detritus available to macroinvertebrates.

Principal Findings and Significance

In total, 95 macroinvertebrate taxa in total were collected between 2004 and 2010 from the two study streams draining the two subwatersheds. Two taxa that were not obtained in 2004 and 2005, the mayfly *Siphlonurus* sp. and the caddisfly *Cheumatopsyche* sp., were surprisingly collected from the Shelly Fork North intermittent stream channel in 2010. Following SMZ harvesting, overall total taxa richness was lower in the intermittent and perennial channels draining both subwatersheds (Table 3). The largest overall relative declines were in the perennial channels (Booker Fork: 60 to 36, = 40% lower; Shelly Fork North: 63 to 37, = 41% lower).

Mean taxa richness, expressed per channel, was lower following SMZ harvesting in both subwatersheds (Table 4), but the response was more pronounced in the Shelly Fork North subwatershed. Between 2004 and 2010, mean richness in the intermittent stream channels was reduced from 23.1 to 13.4 in the Booker Fork subwatershed and 28.5 to 11.0 in the Shelly Fork North subwatershed. Mean richness in the perennial channels decreased from 27.4 to 15.7 and 25.7 to 12.7 in the Booker Fork and Shelly Fork North subwatersheds, respectively, from 2004 to 2010. Similarly, EPT richness declined from 2004 to 2010 in both Booker Fork subwatershed channels (intermittent: 16.2 to 10.2; perennial: 17.2 to 11.4) and more prominently in the Shelly Fork North subwatershed (intermittent: 28.5 to 11.0; perennial: 25.7 to 11.7) (Table 4). Mean richness for each functional group in the intermittent and perennial channels of both streams was also lower in 2010, and particularly for filtering-collectors, scrapers, and shredders (Table 4). Gathering-collector declines were less marked.

There was an inconsistent between-subwatershed pattern of declining macroinvertebrate density following SMZ harvest (Table 5). Density values for the total community, EPT taxa, and four functional groups (excluding predators) decreased between 2004 and 2010 in both intermittent and perennial channels draining the Shelly Fork North subwatershed. The least-pronounced decline was for shredders in the perennial channel (540.3 to 457.5). In contrast, in the Booker Fork subwatershed only filtering-collectors (intermittent: 45.2 to 15.1; perennial: 74.3 to 2.4) and scrapers (intermittent: 172.2 to 47.4; perennial: 225.0 to 104.1) exhibited decreased densities in 2010 compared to 2004 (Table 5). Total density decreased by ca. 25% in the intermittent channel (2171.1 to 1621.0) yet increased markedly in the perennial channel (1685.6 to 2783.1). Similarly, both EPT and gathering-collector densities were lower in 2010 in the intermittent channels (EPT: 1570.5 to 1356.3; GC: 1114.1 to 539.3) but ca. 2-fold higher in 2010 in the perennial channels (EPT: 1146.4 to 2226.9; GC: 692.1 to 1390.9) (Table 5).

Shredders were the sole functional group with higher density values in 2010 in both Shelly Fork North channels (intermittent: 620.0 to 938.6; perennial: 430.6 to 1187.6) (Table 5).

Gathering-collectors were the dominant functional group obtained from both intermittent and perennial stream channels (Table 6), followed by shredders and predators. Scrapers and filtering collectors were the least abundant. The relative abundance of scrapers in pre-harvest conditions ranged from 6.6 to 13.3% and declined to a range of 2.3 to 6.9% in 2010 (Table 6), implying a community food web structure that is heavily dependent on riparian leaf fall and not on in-stream productivity. The ratio of scrapers to shredders + total collectors was < 0.19 during preharvest conditions (Table 7), similarly implying a heterotrophic system, and declined to < 0.08. With one exception, the shredders to total collector ratio values were > 0.25 (Table 7). This also implies a community food web structure that is heavily dependent on the carbon derived from riparian leaf fall during autumn. Filtering-collector to gathering-collector ratios ranged from 0.02 to 0.11 during 2004 and 2005 and decreased slightly to a range of < 0.01 to 0.03 in 2010 (Table 7). This ratio, combined with the low overall proportion of filtering-collectors (Table 6), strongly suggests that each of the study channels have only minimal levels of fine detritus in transport.

There were few individual taxa that were markedly more abundant in 2010 compared to 2004, and none displaying this pattern in both subwatersheds (Table 8). The stonefly genus *Leuctra* sp., in particular, exhibited 2.2- (intermittent) and 3.2-fold (perennial) higher density values in 2010 in the Booker Fork subwatershed. This increase was largely responsible for the higher Booker Fork EPT index value (perennial channel only) and shredder densities (both channels) in 2010 (Table 5). The two other taxa with higher density values in 2010 were the mayfly *Epeorus* sp. (scraper) and stonefly *Isoperla* sp. (predator). The sole taxon displaying higher values in 2010 in both channels in the Shelly Fork North subwatershed was the predatory dipteran *Hemerodromia*, yet this taxon comprised only 0.1% of the total number of macroinvertebrates collected and imparted only a negligible influence on the between-year difference.

In contrast, many taxa displayed consistently lower density values in 2010 (Table 8). Several common EPT taxa exhibited marked decreases in the Shelly Fork North Channels. The gathering-collector mayflies *Paraleptophlebia* sp. (5^{th}-most abundant) exhibited 3+-fold decreases in both channels, *Ephemerella* sp. (6^{th}-most abundant) values were ca. 50- (intermittent) and 3-fold (perennial) lower, *Ameletus* sp. (9^{th}-most abundant) densities were ca. 3.5- (intermittent) and 20-fold (perennial) lower, and *Eurylophella* sp. (26^{th}-most abundant) was entirely absent following harvesting (Table 8). Although densities of the mayfly scraper *Epeorus* sp. were much lower compared to Booker Fork both pre- and post-harvest, this taxon was absent from the Shelly Fork North intermittent channels in 2010 and only one individual was collected from the perennial channels. A second scraping mayfly, *Maccaffertium meririvulanum* (27^{th}-most abundant), was nearly absent after harvesting (Table 8). Mayfly taxa that displayed lower

densities in both Booker Fork channels in 2010 included *Ameletus* sp., *Eurylophella* sp., *M. meririvulanum,* and *Drunella* sp.(gathering-collector; 28th-most abundant) (Table 8). The less-common scraping mayfly *Cinygmula subaequalis* (39th) was not obtained in 2010. Similarly, several common stonefly taxa displayed lower densities across both channel types in both subwatersheds in 2010 (Table 8), namely *Sweltsa* sp. (predator; 8th-most abundant), *Peltoperla arcuata* (shredder; 17th-most abundant) and *Yugus kirchneri* (predator; 20th-most abundant). The predators *Eccoptura xanthenes* (30th-most abundant) and *Acroneuria* sp. (43rd-most abundant), in near identical manner to *C. subaequalis*, were either near-absent or completely absent post-harvest. Several abundant caddisflies were found in much lower densities in 2010 (Table 8). Prominent examples included the shredder *Lepidostoma* sp. (10th-most abundant), scrapers *Agapetus* sp. (11th-most abundant) and *Neophylax* sp. (24th-most abundant), the filtering-collector *Diplectrona modesta* (12th-most abundant), and predators *Rhyacophila carolina* (19th-most abundant), *Polycentropus* sp. (21st-most abundant), and *R. nigrita* (37th-most abundant).

Non-EPT taxa that also had reduced densities across both subwatersheds in 2010 compared to 2004 included several coleopterans (beetles) and dipterans (true flies). Notable examples include the elmid beetle genera *Stenelmis* sp. (14th-most abundant), *Optioservus* sp. (18th-most abundant), and *Oulimnius latusculus* (38th-most abundant), and the psephenid *Ectopria nervosa* (22nd-most abundant) (Table 8). All four beetle taxa are scrapers. Prominent examples of declining post-harvest dipteran densities include the predaceous tipulids *Cryptolabis* sp. (7th-most abundant), *Hexatoma* sp. (15th-most abundant) and *Limnophila* sp. (25th-most abundant), and the filtering-collector simuliid *Simulium* sp. (23rd-most abundant) (Table 8).

In summary, there were three clear trends emanating from this preliminary comparison of headwater stream macroinvertebrate communities draining two subwatersheds that received the most comprehensive SMZ forest harvesting treatment between 2004 and 2010 at Robinson Forest. First, overall taxa, EPT, and individual functional group richness decreased. Second, the majority of the most abundant taxa experienced marked declines in density values. Third, the relative contribution of each functional group, however, wasn't consistently different enough following SMZ harvesting to imply a shift from heterotrophy to autotrophy (i.e., higher proportions of scrapers). Functionally, the study streams draining each subwatershed have similar attributes compared to pre-harvest conditions. Identification of macroinvertebrate samples from study streams draining the remaining six subwatersheds, particularly in the two control subwatersheds, are needed in order to more comprehensively frame between-year community differences due to random changes compared to the forest harvesting treatments.

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Table 1. Robinson Forest SMZ harvesting treatments categorized by channel type. Streams in **boldface** refer to data analyzed in this report.

Stream	Stream channel type Intermittent	Perennial
Falling Rock	control	Control
Little Millseat		
Booker Fork (BF)	**[1]7.6 m, [2]0%**	**16.8 m, 50%**
Shelly Fork North (SFN)		
Goff Hollow	7.6 m, 25%	16.8 m, 100%
Shelly Fork South		
Shelly Fork West	15.2 m, 25%	33.5 m, 100%
Wet Fork		

[1]First value listed: SMZ width; [2]Second value listed: % trees left standing in SMZ

Table 2. Functional feeding group (FFG) ratios as indicators of stream ecosystem energetic properties. FC = filtering-collectors, GC = gathering-collectors, SCR = scrapers, SHR = shredders, TC = total collectors. Modified from Merritt and Cummins (2008).

Ecosystem parameter	FFG ratio	Criteria
Autotrophy to heterotrophy	SCR / SHR + TC	> 0.75: autotrophic system
Coarse detritus to fine detritus	SHR / TC	> 0.25: diverse and robust shredder community
Fine detritus in transport to fine detritus deposited on stream bottom	FC / GC	> 0.50: fine detritus load in transport high

Table 3. Comparison of total macroinvertebrate taxa richness between 2004 and 2010. BF = Booker Fork, SFN = Shelly Fork North, nd = no data.

Year	BF - Intermittent	BF – Perennial
2004	53	60
2005	49	50
2010	39	36
	SFN- Intermittent	SFN – Perennial
2004	56	63
2005	nd	nd
2010	36	37

Table 4. Comparison of mean stream macroinvertebrate taxa richness values for the entire community and each functional group between 2004-2005 (pre-harvesting) and 2010 (post-harvesting). BF = Booker Fork, SFN = Shelly Fork North, FC = filtering-collectors, GC = gathering-collectors, SCR = scrapers, SHR = shredders. nd = no data.

	BF - Intermittent						BF – Perennial					
Year	Total	EPT	FC	GC	SCR	SHR	Total	EPT	FC	GC	SCR	SHR
2004	23.1	16.2	1.9	6.4	5.1	3.9	27.4	17.2	1.9	6.9	6.9	3.0
2005	16.7	11.5	1.4	4.7	2.4	2.5	21.3	14.1	1.9	6.1	3.8	2.2
2010	13.4	10.2	0.7	5.0	1.3	2.5	15.7	11.4	0.2	5.4	2.7	2.1
	SFN- Intermittent						SFN – Perennial					
Year	Total	EPT	FC	GC	SCR	SHR	Total	EPT	FC	GC	SCR	SHR
2004	28.5	17.6	3.2	7.1	5.2	4.5	25.7	16.3	1.8	7.1	5.0	4.2
2005	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
2010	11.0	7.3	1.0	4.9	1.5	2.0	12.7	7.9	0.7	3.9	2.8	2.1

Table 5. Comparison of mean stream macroinvertebrate density (no./m^2) values for the entire community and each functional group between 2004-2005 (pre-harvesting) and 2010 (post-harvesting). BF = Booker Fork, SFN = Shelly Fork North, FC = filtering-collectors, GC = gathering-collectors, SCR = scrapers, SHR = shredders. nd = no data.

	BF - Intermittent						BF – Perennial					
Year	Total	EPT	FC	GC	SCR	SHR	Total	EPT	FC	GC	SCR	SHR
2004	2171.1	1570.5	45.2	1114.1	172.2	620.0	1685.6	1146.4	74.3	692.1	225.0	430.6
2005	1975.2	782.5	29.1	1290.6	48.4	396.1	2960.1	1137.7	180.8	1595.2	125.9	474.7
2010	1621.0	1356.3	15.1	539.3	47.4	938.6	2783.1	2226.9	2.4	1390.9	104.1	1187.6
	SFN- Intermittent						SFN – Perennial					
Year	Total	EPT	FC	GC	SCR	SHR	Total	EPT	FC	GC	SCR	SHR
2004	2384.2	1665.2	80.7	1443.4	158.2	447.8	2290.6	1519.9	61.4	1259.4	176.5	540.3
2005	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
2010	918.2	711.5	21.5	714.7	21.5	138.9	1181.9	882.6	10.8	576.9	81.8	457.5

Table 6. Comparison of mean relative abundance values of stream macroinvertebrate functional groups between 2004-2005 (pre-harvesting) and 2010 (post-harvesting). BF = Booker Fork, SFN = Shelly Fork North, FC = filtering-collectors, GC = gathering-collectors, SCR = scrapers, SHR = shredders. nd = no data.

	BF - Intermittent					BF - Perennial				
Year	FC	GC	SCR	SHR	PR	FC	GC	SCR	SHR	PR
2004	2.1	51.3	7.9	28.6	10.1	4.4	41.1	13.3	25.5	15.6
2005	1.5	65.3	2.5	20.1	10.7	6.1	53.9	4.3	16.0	19.7
2010	0.9	33.3	2.9	57.9	5.0	0.1	50.0	3.7	42.7	3.5
	SFN- Intermittent					SFN - Perennial				
Year	FC	GC	SCR	SHR	PR	FC	GC	SCR	SHR	PR
2004	3.4	60.5	6.6	18.8	10.7	2.7	55.0	7.7	23.6	11.0
2005	nd	nd	nd	nd		nd	nd	nd	nd	
2010	2.3	77.8	2.3	15.1	2.3	0.9	48.8	6.9	38.7	4.6

Table 7. Comparison of functional feeding group (FFG) ratios between 2004-2005 (pre-harvesting) and 2010 (post-harvesting). BF = Booker Fork, SFN = Shelly Fork North, FC = filtering-collectors, GC = gathering-collectors, SCR = scrapers, SHR = shredders, TC = total collectors, nd = no data.

	BF - Intermittent			BF - Perennial		
Year	SCR / SHR + TC	SHR / TC	FC / GC	SCR / SHR + TC	SHR / TC	FC / GC
2004	0.10	0.53	0.04	0.19	0.56	0.11
2005	0.03	0.30	0.02	0.06	0.27	0.11
2010	0.03	1.69	0.03	0.04	0.85	< 0.01

	SFN - Intermittent			BF - Perennial		
Year	SCR / SHR + TC	SHR / TC	FC / GC	SCR / SHR + TC	SHR / TC	FC / GC
2004	0.08	0.29	0.06	0.09	0.41	0.05
2005	nd	nd	nd	nd	nd	nd
2010	0.02	0.19	0.03	0.08	0.78	0.02

Table 8. Comparison of mean density for all unique macroinvertebrate taxa obtained between 2004 (first column) and 2010 (second column). Unidentifiable taxa below the level of family were excluded provided that some material could be taken positively to the genus level. The 2005 data for Booker Fork (BF) was not included. SFN = Shelley Fork North. Taxa names in **bold type** indicate those exhibiting a consistent pattern increased density within a subwatershed between 2004 and 2010. Taxa listed in order of greatest- to least-overall abundance.

		SFN				BF			
Taxa		Intermittent		Perennial		Intermittent		Perennial	
Chironomidae		524.4	164.4	558.9	204.4	442.2	237.8	268.9	513.6
Leuctra	**sp.**	305.6	106.7	423.3	416.7	**416.7**	**910.0**	**353.3**	**1123.5**
Baetis	sp.	334.4	353.3	234.4	246.7	234.4	43.3	182.2	414.8
Paraleptophlebia	sp.	350.0	114.4	255.6	71.1	246.7	151.1	111.1	187.7
Ephemerella	sp.	102.2	2.2	103.3	35.6	40.0	3.3	55.6	190.1
Amphinemura	sp.	84.4	34.4	53.3	50.0	103.3	48.9	28.9	101.2
Cryptolabis	sp.	6.7	0.0	5.6	0.0	20.0	0.0	76.7	0.0
Sweltsa	sp.	58.9	0.0	93.3	1.1	104.4	4.4	44.4	9.9
Ameletus	sp.	73.3	18.9	40.0	2.2	93.3	76.7	33.3	32.1
Lepidostoma	sp.	61.1	1.1	56.7	1.1	62.2	1.1	35.6	0.0
Agapetus	sp.	46.7	0.0	58.9	0.0	25.6	0.0	74.4	0.0
Diplectrona	*modesta*	41.1	17.8	36.7	6.7	21.1	7.8	34.4	1.2
Epeorus	**sp.**	4.4	0.0	8.9	1.1	**27.8**	**34.4**	**25.6**	**76.5**
Stenelmis	sp.	11.1	2.2	51.1	26.7	27.8	3.3	26.7	7.4
Hexatoma	sp.	25.6	2.2	31.1	14.4	12.2	14.4	22.2	19.8
Wormaldia	*moestus*	6.7	0.0	8.9	0.0	2.2	6.7	2.2	2.5
Peltoperla	*arcuata*	14.4	0.0	14.4	3.3	52.2	4.4	24.4	0.0
Optioservus	sp.	6.7	4.4	20.0	38.9	20.0	1.1	13.3	12.3
Rhyacophila	*carolina*	8.9	0.0	17.8	0.0	25.6	14.4	15.6	25.9
Yugus	*kirchneri*	6.7	1.1	10.0	3.3	47.8	4.4	13.3	13.6
Polycentropus	sp.	28.9	2.2	12.2	0.0	11.1	5.6	16.7	4.9
Ectopria	*nervosa*	31.1	0.0	4.4	1.1	12.2	0.0	35.6	2.5
Simulium	sp.	20.0	1.1	20.0	1.1	13.3	0.0	40.0	0.0
Neophylax	sp.	23.3	0.0	3.3	1.1	38.9	3.3	12.2	2.5
Limnophila	sp.	43.3	0.0	17.8	0.0	20.0	0.0	4.4	0.0
Eurylophella	sp.	42.2	0.0	20.0	0.0	6.7	3.3	4.4	0.0
Maccaffertium	*meririvulanum*	20.0	1.1	14.4	0.0	14.4	2.2	11.1	2.5

Table 8. Continued.

Taxa		SFN Intermittent		SFN Perennial		BF Intermittent		BF Perennial	
Drunella	sp.	0.0	0.0	1.1	0.0	5.6	0.0	30.0	9.9
Remenus	*bilobatus*	5.6	3.3	8.9	13.3	4.4	1.1	8.9	6.2
Eccoptura	*xanthenes*	22.2	0.0	24.4	1.1	1.1	0.0	2.2	0.0
Isoperla	**sp.**	0.0	1.1	2.2	1.1	**0.0**	**13.3**	**2.2**	**8.6**
Oligochaeta		2.2	12.2	7.8	0.0	3.3	1.1	11.1	0.0
Bezzia	sp.	3.3	3.3	6.7	3.3	6.7	3.3	4.4	1.2
Ceratopogon	sp.	7.8	3.3	6.7	1.1	1.1	2.2	5.6	2.5
Dixa	sp.	2.2	5.6	4.4	0.0	2.2	0.0	0.0	7.4
Nigronia	*serricornis*	8.9	0.0	3.3	4.4	0.0	2.2	1.1	1.2
Rhyacophila	*nigrita*	2.2	0.0	5.6	0.0	4.4	2.2	4.4	0.0
Oulimnius	*latusculus*	2.2	0.0	3.3	0.0	2.2	0.0	10.0	1.2
Hemerodromia	**sp.**	**0.0**	**5.6**	**0.0**	**10.0**	0.0	0.0	3.3	0.0
Cinygmula	*subaequalis*	0.0	0.0	0.0	1.1	1.1	0.0	3.3	0.0
Sphaerium	sp.	13.3	0.0	1.1	0.0	1.1	0.0	0.0	0.0
Cordulegaster	sp.	5.6	0.0	2.2	0.0	1.1	0.0	6.7	0.0
Acroneuria	sp.	0.0	0.0	2.2	0.0	3.3	0.0	6.7	0.0
Tipula	*abdominalis*	3.3	0.0	5.6	0.0	0.0	0.0	6.7	0.0
Pycnopsyche	*gentilis*	4.4	0.0	2.2	0.0	3.3	1.1	1.1	1.2
Anchytarsus	*bicolor*	3.3	0.0	7.8	0.0	0.0	0.0	0.0	0.0
Ephemera	sp.	8.9	0.0	1.1	0.0	0.0	0.0	1.1	0.0
Stenacron	*interpunctatum*	0.0	2.2	0.0	1.1	0.0	0.0	4.4	1.2
Microcylloepus	*pusillus*	0.0	0.0	4.4	0.0	4.4	0.0	0.0	0.0
Pycnopsyche	sp. A	2.2	0.0	3.3	0.0	0.0	0.0	1.1	0.0
Molanna	sp.	3.3	0.0	2.2	0.0	0.0	0.0	1.1	0.0
Stenacron	sp.	2.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Stylogomphus	*albistylus*	4.4	0.0	1.1	0.0	0.0	0.0	0.0	0.0
Chelifera	sp.	0.0	0.0	0.0	0.0	1.1	0.0	1.1	0.0
Cambarus	sp.	0.0	0.0	1.1	0.0	1.1	0.0	2.2	0.0
Ostrocerca	sp.	1.1	0.0	0.0	0.0	0.0	3.3	0.0	0.0
Ceratopsyche	***ventura***	**0.0**	**1.1**	**0.0**	**3.3**	0.0	0.0	0.0	0.0
Tabanidae		3.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table 8. Continued.

Taxa		SFN Intermittent		SFN Perennial		BF Intermittent		BF Perennial	
Maccaffertium	*vicarium*	0.0	0.0	0.0	0.0	0.0	0.0	3.3	0.0
Stenacron	*carolina*	0.0	0.0	2.2	0.0	0.0	0.0	1.1	0.0
Siphlonurus	sp.	0.0	2.2	0.0	0.0	0.0	1.1	0.0	0.0
Diploperla	*robusta*	1.1	0.0	1.1	0.0	0.0	1.1	0.0	0.0
Dicranota	sp.	0.0	0.0	3.3	0.0	0.0	0.0	0.0	0.0
Tipulidae-3		0.0	0.0	2.2	0.0	0.0	0.0	0.0	0.0
Planariidae		0.0	0.0	0.0	0.0	1.1	0.0	1.1	0.0
Stenonema	*femoratum*	0.0	0.0	0.0	2.2	0.0	0.0	0.0	0.0
Prostoia	*similis*	0.0	0.0	0.0	0.0	2.2	0.0	0.0	0.0
Diplectrona	*metequi*	0.0	0.0	0.0	0.0	1.1	0.0	0.0	0.0
Molophilus	sp.	0.0	1.1	1.1	0.0	0.0	0.0	0.0	0.0
Nematoda		0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.2
Psychoda	sp.	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.2
Asellidae		0.0	1.1	0.0	0.0	0.0	0.0	0.0	0.0
Maccaffertium	sp.	0.0	0.0	0.0	1.1	0.0	0.0	0.0	0.0
Habrophlebia	*varians*	1.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Allocapnia	sp.	0.0	1.1	0.0	0.0	0.0	0.0	0.0	0.0
Cheumatopsyche	sp.	0.0	1.1	0.0	0.0	0.0	0.0	0.0	0.0
Lype	*diversa*	1.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Helichus	*basalis*	0.0	0.0	0.0	0.0	0.0	0.0	1.1	0.0
Psephenus	*herricki*	0.0	0.0	0.0	1.1	0.0	0.0	0.0	0.0
Ephydridae		0.0	0.0	1.1	0.0	0.0	0.0	0.0	0.0

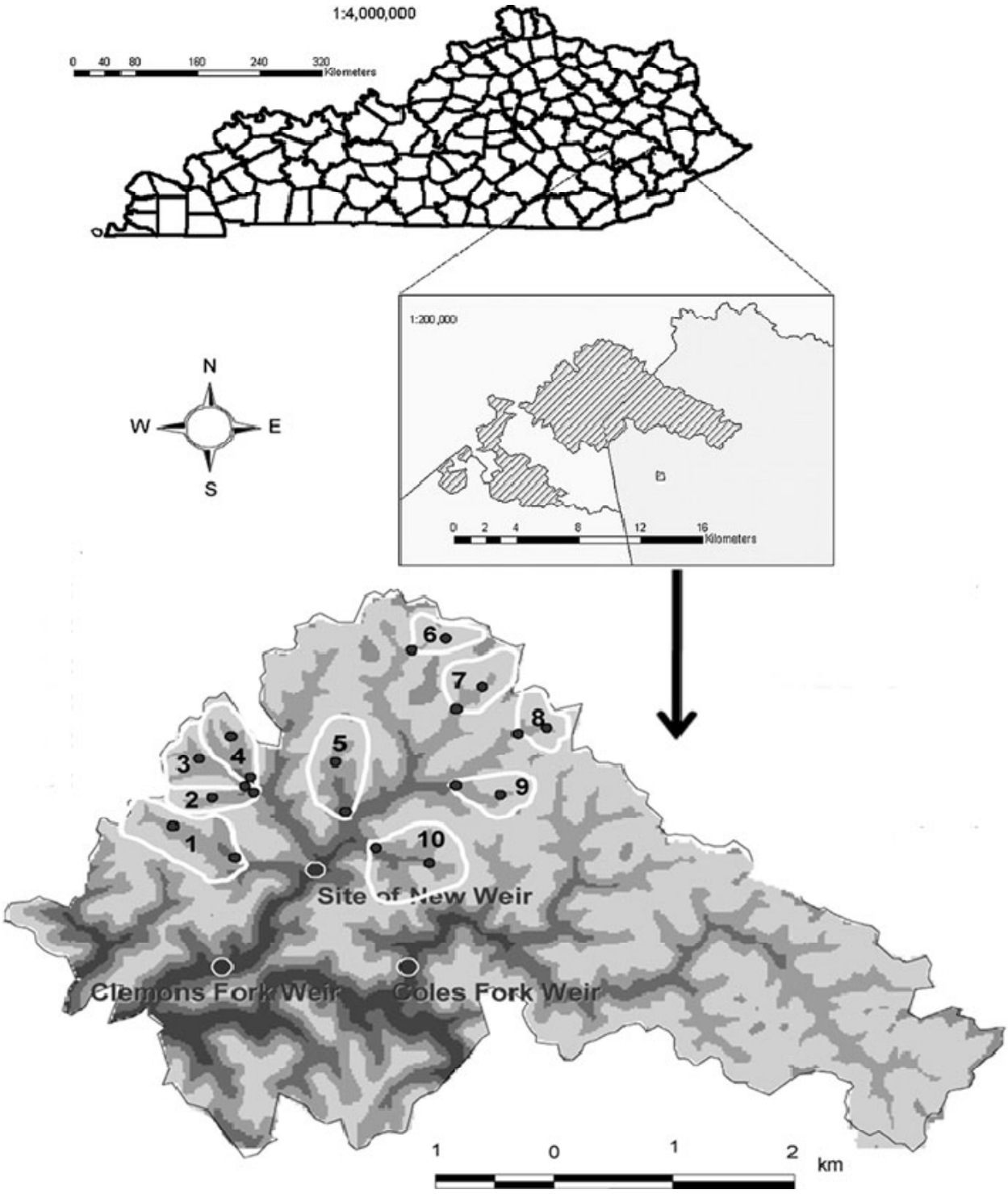

Fig. 1. Location of Robinson Forest in eastern Kentucky and the eight study streams. Map adapted from Cherry (2006). Streams draining watersheds 6 and 8 were not subject to forest harvesting treatments.

Bush honeysuckle induced aquatic hypoxia

Basic Information

Title:	Bush honeysuckle induced aquatic hypoxia
Project Number:	2010KY141B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Fourth
Research Category:	Biological Sciences
Focus Category:	Ecology, Acid Deposition, Water Quality
Descriptors:	exotic invasives, ephemeral systems, dissolved oxygen
Principal Investigators:	Richard Durtsche

Publications

1. Hayes, S.J., and R.D. Durtsche, 2010, Hypoxia in Aquatic Systems Resulting from Leaf Litter Leachate of the Invasive Amur honeysuckle (Lonicera maackii), in Proceedings ORBCRE Annual Meeting, Highland Heights, Kentucky.
2. Durtsche, R.D., 2010, Impact of the honeysuckle shrub (Lonicera maackii) on Anuran Larvae in Aquatic Ecosystems, in Proceedings ORBCRE Annual Meeting, Highland Heights, Kentucky.
3. Hayes, S.J., and R.D. Durtsche, 2010, Invasive Plant Induced Hypoxia in Aquatic Systems: Microbial Activity or Humic Content?, Kentucky Academy of Sciences Annual Meeting, Bowling Green, Kentucky.
4. Hayes, S.J., and R.D. Durtsche, 2011, Bush Honeysuckle Induced Aquatic Hypoxia, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 17-18.
5. Hayes, S.J., and R.D. Durtsche, 2011, Potential Agents of Induced Hypoxia in Aquatic Systems from the Invasive Amur Honeysuckle Shrub (Lonicera maackii), Kentucky Invasive Species Conference, Lexington, Kentucky.

Bush Honeysuckle Induced Aquatic Hypoxia

Project/Research Objectives: Our study addresses the impact of the exotic invasive Bush honeysuckle (*Lonicera maackii*) on inducing hypoxic conditions in ephemeral aquatic ecosystems which can have adverse effects on living organisms in these ephemeral systems. Our previous investigations using leaf teas from *L. maackii* and native riparian plants suggested that the leachates in these teas contained potential oxygen binding proteins. However, using several independent tests on these leaf tea leachates we did not find any evidence of these potential oxygen-binding proteins. Our focus in this study was to determine what components of the leaf leachates might bind oxygen. As metals form the basis of many oxygen-binding reactions, we evaluated both leaves and leaf leachates for their content of elemental metals. We also measured for the presence of organics or humics in the leachates as chelating ligands for the metal ions that could bind oxygen. In addition to these chemical analyses of the leaves and waters containing these leaves, we assessed the microbiological activity in leaf tea leachates. Any increase in microbial growth in aquatic systems has the potential to deplete dissolved oxygen levels in aquatic systems thereby resulting in hypoxic conditions.

Methodology: This research involves an undergraduate, Stephanie Hayes, who through directed studies was involved in all aspects of this project.

Leachates of *L. maackii* and native riparian tree leaves were prepared as teas from 0.75 g dry leaves/L of Reverse Osmosis (RO) water. Leaves dried to a constant mass at 60˚C in a drying oven were placed in empty tea bags, sealed with hot glue, and suspended in water for 48 hours prior to analyses. Dissolved oxygen readings in teas were determined with a galvanic dissolved oxygen probe connected to a multi-meter data-logger.

A full spectral analysis was performed on teas from both types of leaves, and from RO water with a Molecular Devices 190A 96 well plate reader. The spectral range from 190 – 500 nm was evaluated to look for any peaks that might indicate the presence of organic particles. A peak in the 240 - 260 nm wavelength range for example, would indicate the presence of an oxygen-binding protein. A BCA assay was also run. This analysis is only used to determine whether or not protein is present at all. If a protein is present, the solution turns purple, if not it remains clear. Finally, an SDS-PAGE with silver staining, 4% stacking gel and 10% resolving gel was used in order to separate any proteins based on size. Later, we became concerned about possible humics interfering with the analysis and decided to filter the teas using 3500 dalton dialysis tubing. Another SDS-PAGE was used, this time with a 4% stacking gel and 20% resolving gel in order to increase the range of proteins it could detect.

Chemical evaluation of potential oxygen-binding metals and other elements of leaf matter and teas for both *L. maackii* and native plants was determined using a ThermoFisher iCAP Inductively Coupled Plasma - Optical Emissions Spectrometer (ICP – OES) for mineral and element analyses. Prior to analysis of leaf materials with this

machine, leaves dried to a constant mass at 60˚C were acid digested under high pressure with a Milestone Ethos EZ microwave digester. A second set of analyses were made with teas of Amur honeysuckle or a mix of native riparian hardwood leaves.

To assess the impact of microbial activity on the reduction in dissolved oxygen in leaf leachates, a series of bacterial examinations were undertaken. Serial dilutions to 10^{-6} were made of teas using sterilized water, and TSA (tryptic soy agar) plates were streaked and incubated 24 – 48 hours to detect bacterial colonies. Microbial population estimates were made to determine following the standard plate count protocol from Brown, A.E. (2007). Samples taken from individual colonies were then evaluated to determine if they were gram positive or gram negative based on staining techniques, and visually inspected for morphotype and color under the microscope with oil-immersion at 1000x. Microbial strains were also assessed to be either aerobic, anaerobic, or both with thyoglycolate agar growth tubes. To eliminate bacteria from the leaves prior to creation of the leachates, dried leaves were exposed to UV radiation for 5 min. per side of each leaf. Teas were then derived from these irradiated leaves using sterile water, and TSA plates streaked to see if microbes were eliminated.

A second sterilization technique was used where tea of *L. maackii* was derived from heated but non-boiling water, and then sterilized by filtering through a sterile 0.2 nm filtration system. Dissolved oxygen readings were also taken of this sterilized tea after a 48 hour time period. Filtered tea from this sterilization technique was then used with technical agar to produce new media plates for growing bacteria specific for leaf degradation of *L. maackii*.

Microbial metabolic activity resulting from leaf decomposition in water was measured indirectly through the consumption of oxygen and production of carbon dioxide. Respirometry chambers (0.5 L) containing 0.4 L leaf teas (0.3 g leaf or standard tea conc.) of *L. maackii*, native riparian hardwoods, or control RO water (n = 5 chambers per condition) were sealed as static systems. Air samples (1ml) were withdrawn from above these teas and control water daily for 14 days, and injected into a Sable Systems gas analysis system with a CA-2A CO_2 Analyzer, and a FC-10A O_2 Analyzer. Calibrations, drift and barometric pressure (BP) adjustments were made with standardization gases: technical grade N_2 for 0 point, and CO_2 span gas for high point or natural air for O_2 content (20.95%). Measured CO_2 and O_2 levels were BP – drift adjusted prior to analyses.

Our most recent studies have determined the leaf composition ratio in native riparian leaf litter from areas with and without Amur honeysuckle. Leaf litter samples (15) were taken from St. Anne wetlands and separated by species. Leaf litter ratios were used to establish mesocosms of single and mixed leaf composition, with and without Bush honeysuckle. Roughly 14.33g of leaf material was added to each 18.9 L (5 gal.) bucket mesocosm to create concentrations (g dry leaf matter/L water) found in natural wetland ecosystems. These mesocosms allowed us to monitor dissolved oxygen levels in a "natural" setting in order to determine the degree and duration of hypoxia occurring naturally. Dissolved oxygen levels were monitored every day using a YSI dissolved oxygen probe.

Principle Findings and Significance: Spectral analyses across a range of spectra (170 – 500 nm) for both Amur honeysuckle and native riparian tree leaf teas and RO water show that Amur honeysuckle has peak absorbance more than 2 times that of the native tea at the low (UV) end of the spectrum (Fig. 1). These shoulders suggest that there is more particulate matter in the tea of *L. maackii* than that of the native tea or RO water. Such particulate matter

could be organic compounds like humics or an indication of microbial activity. Peaks within the 240 – 260 nm range suggesting oxygen binding proteins were not observed. Neither the BCA assay or SDS–PAGE analysis were positive for protein.

Evaluation of both leaves and teas for metal and elements from element analysis with the ICP-OES are shown in Table 1. Several metals including Calcium, Potassium, Magnesium, and Manganese show higher concentrations in the *L. maackii* tea than found in

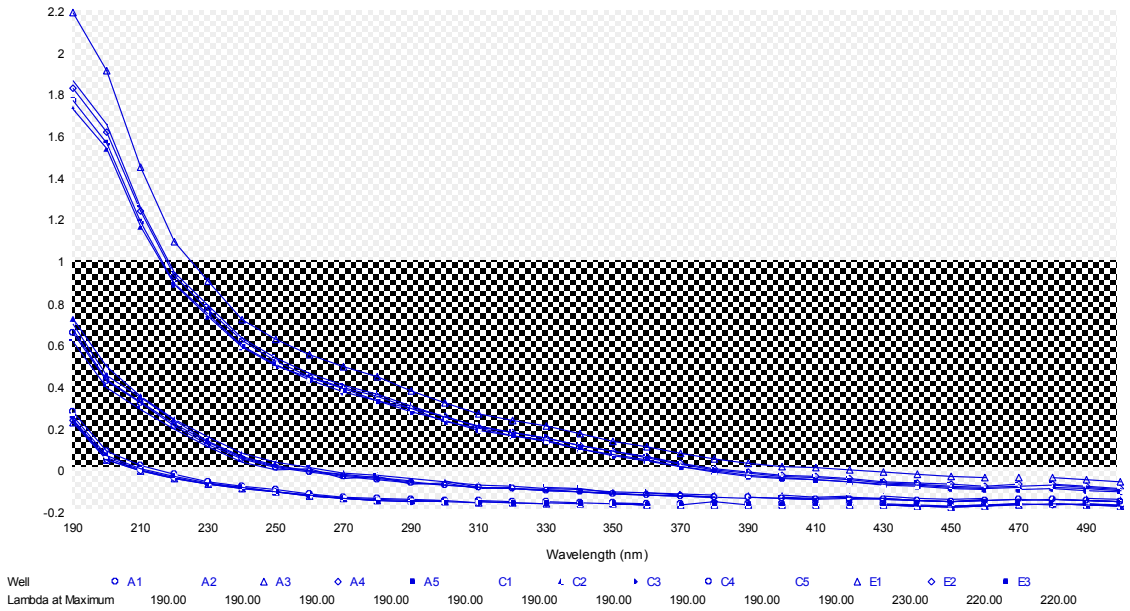

Fig. 1. This spectrogram shows the spectral absorbance from both teas and RO water. The largest peak is Amur Honeysuckle and the smallest is RO water with the native tea in the middle.

the leaves themselves suggesting that these elements are readily lost from the leaves relatively rapidly (48 hours) when placed in water. A similar pattern with native plant leaves was not observed. Table 2 shows a similar pattern in ephemeral pond water with and without *L. maackii* leaves. While potassium measurements were not registering during this analysis, the other metals did accumulate in the water containing the leaves.

Table 1. Chemical Analysis of Amur Honeysuckle and Native Tree teas using iCAP 6000 series spectrometer (values in ppm).

	Ca	Cr	Cu	Fe	K	Mg	Mn	Ni	Zn
AH Leaf	5.8633	0.0061	0.0091	0.1391	0.1377	0.1886	0.0109	Trace	0.0162
NT Leaf	14.6533	0.0019	0.0093	0.1037	2.334	0.8133	0.0312	Trace	0.0139
AH Tea*	19.4967	Trace	0.0061	Trace	5.8240	2.2137	0.05954	Trace	0.0198
NT Tea*	1.6977	Trace	0.0053	Trace	0.6659	0.2603	0.0098	Trace	0.0030

* Teas were made in RO water.

Table 2. Chemical Analysis of ephemeral pond water with and without Amur honeysuckle leaves using iCAP 6000 series spectrometer (values in ppm).

	Ca	Cr	Cu	Fe	K	Mg	Mn	Ni	Zn
FW Tea No AH	11.6067	0.0040	0.0404	3.0572	----	1.7473	0.0548	Trace	0.0267
FW Tea with AH	46.7733	0.0067	0.0062	3.2997	----	6.5223	0.3631	Trace	0.0375

Our investigations into any differences in microbial activity were quite striking. TCA plates streaked with teas from *L. maackii* contained 10^3 more bacteria than plates streaked from native plant teas (Fig. 2). Evaluation of the bacterial colonies found on the Amur honeysuckle plate suggest that most of the microbes were gram negative (Table 3). Colony B from the plate (one of the most abundant colonies) turned out to be a super society of three different bacterial types, 2 gram negative and one gram positive. When we streaked new plates with Amur honeysuckle tea prepared from leaves irradiated under UV lights, only gram positive microbes and a yeast were observed to grow. We did not get any bacterial growth in native plant teas where the leaves were irradiated.

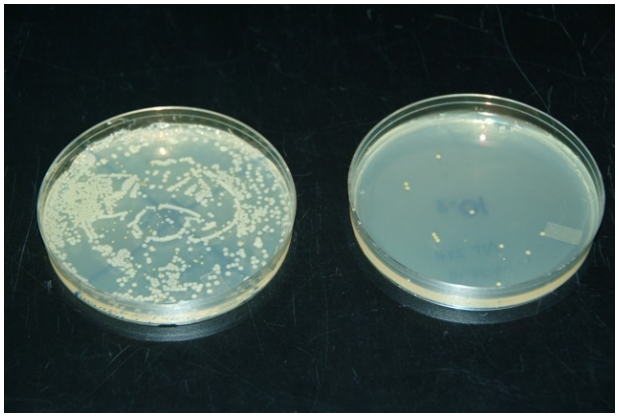

Fig. 2. Bacterial plates from both teas. Amur honeysuckle on the left, and native trees on the right.

The gram positive microbes of the irradiated Amur honeysuckle tea and the yeast all had some form of capsule around each organism, suggesting that these were the results of spores that survived the irradiation.

Table 3. Bacteria and yeast found within Amur Honeysuckle Teas.

	Gram + / -	Color	Shape	UV Y/N
Bacteria A	Negative	Yellow	Chains	N
Bacteria B*	2 neg./1 pos.	Tan	Mixed	N
Bacteria C	Positive	Peach/Tan	Rods	N
Bacteria D	Negative	Dark Yellow	Rods	N
Bacteria E	Positive	Gold	Rods	Y
Bacteria F	Positive	Yellow	Round	Y
Bacteria G	Negative	White	Mixed	N
Bacteria H	Negative	Yellow	Rods	N
Yeast A	n/a	White	n/a	Y

The thyoglycolate tubes were only used for the bacteria found in the initial plate count (Bacteria A-D). All four of these bacteria were found to be aerobic and depleted the allotted oxygen in 48 hours or less. Results of sterile filtration of teas prepared with hot water suggest no bacteria are growing after a period of 48 hours, and dissolved oxygen levels in these teas were not significantly different than that in RO water (Fig. 3). This suggests that bacteria may be responsible for hypoxic conditions in aquatic ecosystems receiving allochthonous leave litter from *L. maackii*.

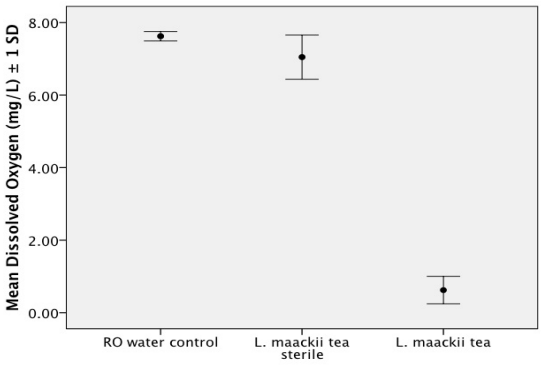

Fig. 3. Dissolved oxygen levels of reverse osmosis water, filter sterilized Amur honeysuckle tea, and unsterilized Amur honeysuckle tea.

After measuring the gas samples over a 12 day period in order to evaluate the potential metabolic activity of the bacteria, we ran a repeated

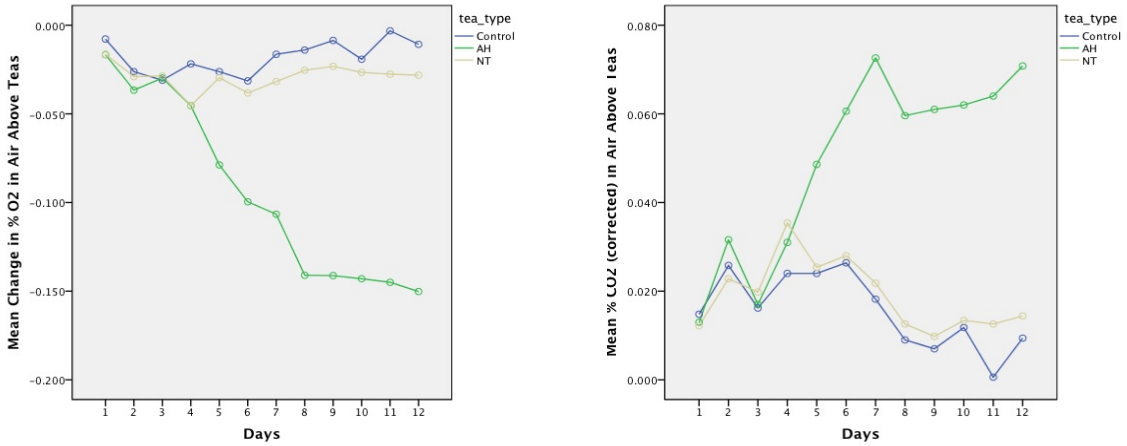

Fig. 4. Gas levels of the control (blue), native riparian teas (yellow) and Amur honeysuckle teas (green) for a) Oxygen $F_{2,12} = 614.07$ $P<.001$; and b) Carbon Dioxide $F_{2,12} = 178.45$ $P<.001$

measures ANOVA (Fig. 4a and 4b). The Amur honeysuckle tea obviously has a much larger amount of carbon dioxide produced and a much smaller amount of oxygen remaining than the native tea and RO water control.

The average proportions of species in the natural leaf litter of St. Anne wetlands can be found in Fig. 5. The monitoring of the mesocosms has just concluded and we will soon be compiling the data in order to run statistical analyses to determine if there is any significant difference in the dissolved oxygen levels of the 33 buckets.

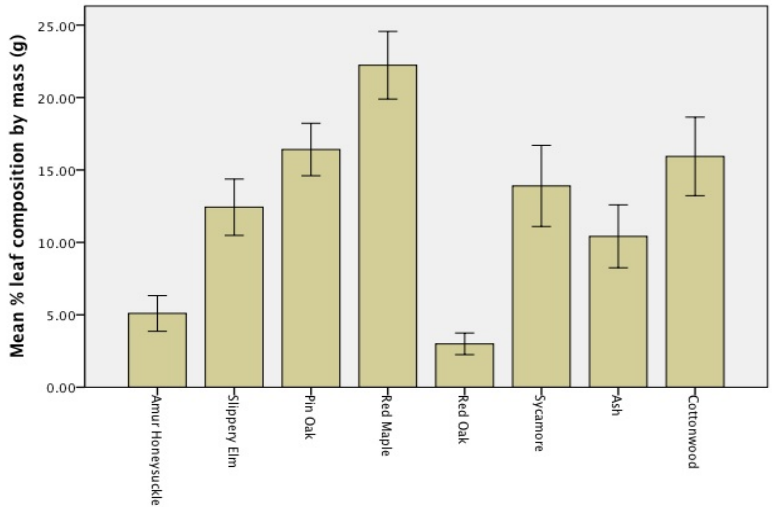

Fig. 5. Leaf composition of litter fall in a riparian wetland forest.

Biological process for manganese control in water supplies

Basic Information

Title:	Biological process for manganese control in water supplies
Project Number:	2010KY142B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Treatment, Water Quality, Methods
Descriptors:	drinking water, treatment
Principal Investigators:	Yi-Tin Wang

Publication

1. Snyder, Michael, and Y.T. Wang, 2011, A Laboratory Scale, Continuous Flow Bioreactor for the Removal of Manganese in Water Supplies, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 19-20.

Biological Process for Manganese Control in Water Supplies

Problem and Research Objectives

Manganese is a common contaminant in drinking water supplies in Kentucky, particularly in communities that rely on groundwater. Manganese imparts color, turbidity and taste and other undesirable water-fouling issues and has recently been implicated as a potential health concern by the USEPA[1]. Most drinking water treatment technologies for removal of soluble manganese rely on the addition of chemical oxidants to the raw water to form insoluble oxides followed by solid-liquid separation[2]. These technologies are vulnerable to failure in the presence of naturally occurring organic matter such as humic acids, which complex manganese and consume the chemical oxidants intended for the manganese. Additionally, cost of chemicals and formation of cancerous disinfection-by-products are of concern.

Our ongoing research interest is to investigate the efficacy of biological filters to remove/control manganese in the presence of humic substances. One limiting requirement for biological filters is sustainable amounts of biodegradable organic matter in the untreated water. The largest fractions of natural organic matter in freshwaters, macromolecular-sized humic acids, are generally recalcitrant to microbial biodegradation, and have limited the applicability of biological filters. It has been previously demonstrated that the metal catalyst Manganese Oxide (MnO_2) can degrade humic acids to low-molecular-weight products that may possibly serve as 'food' substrates for bacteria.[3] Our plan was to implement this concept in a continuous flow biological filter utilizing MnO_2-coated sand filter media that is supplied humic acids as the sole carbon source. We sought to evaluate the capability of a new manganese-oxidizing bacteria isolate in this filter to utilize the oxidation products of humic substances and control/remove manganese from the influent water stream. It was hypothesized that the Mn(II)-oxidizing bacteria will grow and attach to the sand filter media forming biofilms that remove both humic acids and manganese from the untreated water.[3] Preliminary batch studies utilizing our Mn(II)-oxidizing isolate have shown enhanced growth and Mn(II)-oxidation rates utilizing the low-molecular weight degradation products generated from the reaction of humic acids with MnO_2. This isolate appears to re-deposit the Mn-oxide on its outer cell surface.

One of the promising features of such a biological filter design is that the MnO_2-coated sand will be continuously regenerated with bacterially produced MnO_2, reducing or eliminating the need to replace the metal oxide coating on the granular media. The biological filter will be optimized for maximum removal of manganese in the presence of humic substances. The results from this study could potentially serve as the basis for an innovative drinking water treatment technique for the removal/control of both manganese and humic substances and also increase the use of biological filters.

The overall research objective of this ongoing research is to obtain a better understanding of suspended and fixed-film biological processes for manganese Mn(II) control/removal in water supplies provided humic acids as the sole carbon source. At present there is no reported information available on this subject.

Specific objectives of this research include:

I. Isolate a soil/freshwater Mn(II)-oxidizing bacteria and identify the species using 16s-rDNA gene sequencing. Determine optimal Mn(II)-oxidizing conditions (pH and temperature) and evaluate capacity of isolate to grow on selected substrates.

II. Test the hypothesis that Mn (III, IV) oxide can increase the biodegradability of humic acids.

III. Obtain the optimal ratio of humic acids /Mn (III, IV)-oxide concentrations and reaction (contact) time to maximize humic acids biodegradability.

IV. Investigate Mn(II)-oxidation rates using the isolate in batch and continuous flow biological reactors using Mn (III, IV) oxide treated humic acids as the sole carbon source. Evaluate Mn(II)-oxidation for different flow rates, reactor detention times and concentrations of Mn(II).

Methodology

Experimental evaluation was initiated by examining Mn(II)-oxidation rates in batch cultures of a Mn(II)-oxidizing bacteria we isolated (a *Pseudomonas Putid*[4] strain). This isolate, a Mn(II)-oxidizing, gram-negative, biofilm-forming, ubiquitous soil and freshwater bacterium, oxidizes Mn(II) to Mn(III,IV) oxides and accumulates the solids on its exterior surface. We have studied its ability to oxidize Mn(II) while utilizing glucose and then the oxidation products of humic acid and MnO_2 as the sole carbon source. The effect of pH and temperature on the relative rates of Mn (II) oxidation have been evaluated for both of these carbon substrates. These batch results will be used to evaluate the relationship between substrate utilization rate, rate of Mn (II) oxidation, and rate of bacterial growth. We are further evaluating the effects of MnO_2 on the oxidation of humic acids. We have hypothesized that both bacteria growth and Mn(II)-oxidation rates will be enhanced over controls when supplied humic acid-MnO_2 oxidation products.

Most recently we ran a continuous-flow, bench-scale bioreactor utilizing the *Pseudomonas Putida* isolate. The bioreactor, constructed from a 20.13 cm L x 2.28 cm ID plastic tube packed with 4 mm glass beads, was operated for 200+ days. A continuous recycle flow was incorporated in the bioreactor design and the bioreactor hydraulic retention time (HRT) was 48 hrs. (except for periods of increased influent flow rates for flushing undesired, releases of Mn from the biofilm into the surrounding liquid medium). The influent Mn(II) solution feed concentration was 0.500 ± 0.03 mg/L (500 ± 30 ppb) and the bioreactor solution pH and temperature were maintained at 6.50 ± 0.20 and 30.0° ± 1.0°C respectively for most of the operation period. The influent solution feed media consisted of α-glucose, nutrients (phosphate and nitrogen) and trace metals for most of the bioreactor operation period.

Principal Findings and Significance

A Mn(II)-oxidizing bacteria was isolated from a surface soil sample and identified as a member of the *Pseudomonas Putida* species based on its 16s-rDNA gene sequence. Subsequent batch experiments determined that this isolate oxidizes Mn(II) in the stationary growth phase, characteristic of other Mn(II)-oxidizing strains from this species. Screening tests found it capable of utilizing a variety of growth substrates as the sole carbon source. Among these substrates, isolate growth and Mn(II)-oxidation rates appear strongest for glucose and benzoate. Isolate growth and Mn(II)-oxidation rates utilizing the oxidation products of Aldrich Humic Acid (AHA) and Mn(III,IV) oxide as the sole carbon source were higher compared to controls (untreated AHA), validating the core thesis hypothesis.

Optimal Mn(II)-oxidation rates conditions for the isolate were determined to be pH 6.5 and 30°C utilizing glucose and oxidized humic acids as the sole carbon source.

Experiments to determine the optimal ratio of AHA/Mn(III,IV) oxide to maximize AHA biodegradability were completed utilizing two indicators of growth – viable cell count and biological oxygen demand (BOD). The results indicate that the biodegradability of AHA is dependent on the amount of Mn(III,IV) oxide and reaction time. Lower amounts of Mn(III,IV) oxide required longer reaction times and shorter reaction required higher amounts of Mn(III,IV) oxide to yield comparable results. The optimal contact time and Mn(III,IV) oxide (GreensandPlus) concentration was found to be 45 min. and 2 g for solution volumes of ca. 300 ml and 50 mg/L AHA. Isolate viable cell counts grown on these prepared solutions increased up to 10-fold compared to controls after 16 hours of growth. BOD levels in closed bottles following a 36-hr incubation on these solutions increased over 5-fold over the controls.

The pH of the reaction and concentration of calcium had a significant effect on the AHA-Mn(III,IV) oxide reaction. For reactions at pH 2.75, 7.0 and 9.25 with added Ca^{2+}, BOD increased ca. 3, 5, and 0.5-fold respectively compared to controls for the optimal AHA/Mn(III,IV) oxide ratios. For reactions at pH 7.0, Ca^{2+} had a significant effect on BOD for Mn(III,IV) oxide levels up to 1.5 g. At Mn(III,IV) oxide levels >1.5 g, Ca^{2+} had no observable effect on BOD.

Dissolved organic carbon (DOC (mg/L)) and uv-254 levels for 45-min. AHA-Mn(III,IV) reactions at pH 7.0 and 9.25 did not show significant changes. However, at pH 2.75, DOC decreased by ca. 4 mg/L.

Mn(II)-oxidation rates (Fig. 1) for the strain utilizing oxidized AHA were determined to be 0.14 mg/L/hr (from 5 hours to 24 hours after isolate inoculation) compared to 0.083 mg/l/hr for untreated AHA in separate 4-L batch reactors.

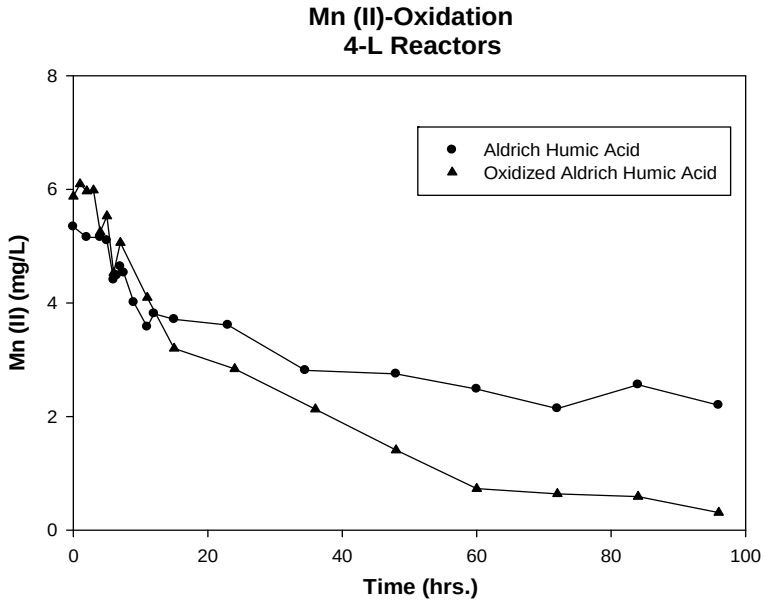

Figure 1. Strain Mn(II)-oxidation on AHA and oxidized AHA in separate 4-L batch reactors

Continuous-flow bioreactor: following bioreactor start-up, influent Mn(II) levels of 0.500 mg/L (500 ppb) were treated to effluent levels of approx. 0.01 mg/L (10 ppb). The bioreactor experienced several failures to remove Mn(II) below the Mn SMCL of 0.05 mg/L (50 ppb). One of the failures was characterized by a gradual increase in the effluent Mn(II) level over several days and is best explained as a lack of the proper nutrients in the α-glucose feed media to support *P. Putida's* ability to sustain Mn(II)-oxidation in the biofilms. Several sudden failures characterized by large releases of Mn from the biofilm are explained as being due to rapid drops in the solution pH in the bioreactor. Most of the releases of Mn were in the Mn(II) oxidation state, indicative of reduction processes occurring on the biofilm. In addition, it is noteworthy that separate, preliminary bioreactor studies of shorter operation time duration revealed that high influent feed solution loads of certain organic and inorganic chemicals induced sudden releases of Mn from the biofilms, loading conditions which were abated in the 200+ day bioreactor.

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Estimating impacts of land use and management on soil water and solute transport

Basic Information

Title:	Estimating impacts of land use and management on soil water and solute transport
Project Number:	2010KY145B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Ground-water Flow and Transport
Focus Category:	Agriculture, Groundwater, Solute Transport
Descriptors:	soil structure, soil properties, water flux
Principal Investigators:	Mark Steven Coyne

Publication

1. Kreba, S., O. Wendroth, and M. Coyne, 2011, Estimating Impacts of Land Use and Management on Soil Structure, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 109-110.

Estimating Impacts of Land Use and Management on Soil Water and Solute Transport

Problem and Research Objectives

Changes in land-use caused by demographic, economic, political, or cultural development have notable effects on soil quality. These changes can alter soil structure and have a negative impact on the environment. Soil use and management can affect physical and hydraulic soil properties that influence soil water and solute transport. Studying the effects of land use on soil properties affecting soil structure can help better determine the impacts of land use and management on soil and ground water systems and to evaluate ground water recharge and discharge. The original objectives of this project and its longer term goals were to estimate changes in soil water flux immediately after land use changed, and to evaluate the effects of changing soil use on ground water recharge and discharge. Also simulation of long-term impacts related to changing land use on soil water and solute transport. To meet these objectives, soil structure had to be characterized. Several measurements were performed and others are in progress to characterize and compare soil structure in two soil use systems, agriculture cropland and pasture.

Methodology

The research site is at Spindletop Research Farm, Lexington KY. The site has an area of 4125 m^2 with two established land use types, pasture and cropland (Fig. 1). A total of 60 sampling points are laid out in four transects. Forty-four of the 60 points are distributed at a regular interval of 5 m. In four nests located in the middle of each transect (Fig. 1), sampling locations are separated by 1m distance. Each nest consists of 6 points. The purpose of these nests with shorter sampling intervals was to quantify the variability structure and spatial association at short lag intervals. The cropland site was planted to corn in spring and harvested in fall 2009. Wheat was planted in fall 2009 and harvested in summer 2010. No-tillage cultivation was performed.

At each of the 60 locations, undisturbed soil cores were taken from 0.04-0.10 m depth for gas diffusivity and air-filled porosity measurements. Soil cores were 80 mm diameter and 60 mm height. Gas diffusivity was measured using a chamber method similar to that described by Rolston (1986). Diffusion chambers (1.88 L) were fabricated and a soil core was placed in the top of the chamber (Fig. 2). One end of the soil core was open to the diffusion chamber and the other to the lab atmosphere. Oxygen was chosen for estimating gas diffusivity and its concentration inside the chamber was reduced by rinsing the chamber with helium. The chamber had two valves (inlet and outlet) that were used to rinse the chamber and to keep the inside of the chamber at atmospheric pressure. A small fan inside the chamber provided mixing prior to sampling. A syringe was used to remove the gas sample from the diffusion chamber and to inject it into a gas chromatograph (GC) (GC-8A with TCD detector). The GC was used to measure the oxygen concentration and its readings were calibrated using the oxygen concentration in the atmosphere (210000 ppm) as a reference. The oxygen concentration inside the chamber is measured every 30 minutes for a 480 minute period. The oxygen diffusion coefficient was estimated at -333 cm soil matric potential. The matric potential was controlled using a pressure plate apparatus, and the air-filled porosity was calculated from the volumetric

water content and total porosity of the soil core. The gas diffusion coefficient was calculated based on the increase of oxygen concentration as a function of time and the difference of oxygen concentration between both open ends of the soil core as described by Rolston (1986).

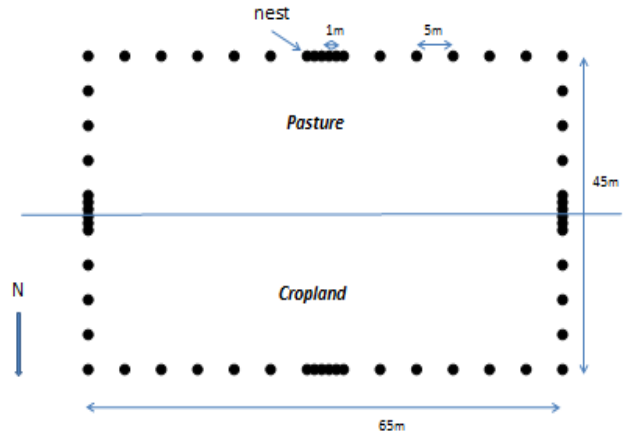

Fig 1: Site condition showing two land use systems (cropland and pasture) and 60 sampling points along four transects.

Fig. 2: Gas diffusion chambers.

Carbon dioxide and nitrous oxide fluxes were evaluated at each of the 60 locations along the four transects using a photo acoustic environmental gas monitor (INNOVA Model 1412) to measure CO_2 and N_2O concentrations. Collars (0.3 m diameter and 0.15 m height) were inserted 50 mm into the soil in each measurement location. A 0.3 m diameter chamber with air-tight fitting was placed in the top of the collar each time a measurement was taken. Gas concentration was sampled every minute for ten minutes from each location, and the gas flux was estimated by calculating the slope of cumulative gas concentration versus time curve. Soil moisture content was measured in 0.1-m depth increments down to 1-m depth each time the gas flux measurements were taken along the four transects using a capacitance probe. Access tubes were installed to 1-m depth in each measurement point and 1 m beside the gas flux collar. Soil temperatures were also monitored each time the gas flux was measured in each location using a thermometer inserted 50 mm into the soil surface beside the collar.

Principal Findings and Significance

The pasture has lower bulk density than the cropped area (Fig. 3). Average bulk density in the cropped site was 1.5 g/cm^3, and it was 1.4 g/cm^3 in the pasture site. Bulk density can be an indicator of soil structure, porosity, and soil water properties. Soils with higher bulk density tend to have lower porosity, and porosity has an important role in soil hydraulic properties. Porosity

controls water infiltration and water storage. Soils with larger pores tend to have higher infiltration rates.

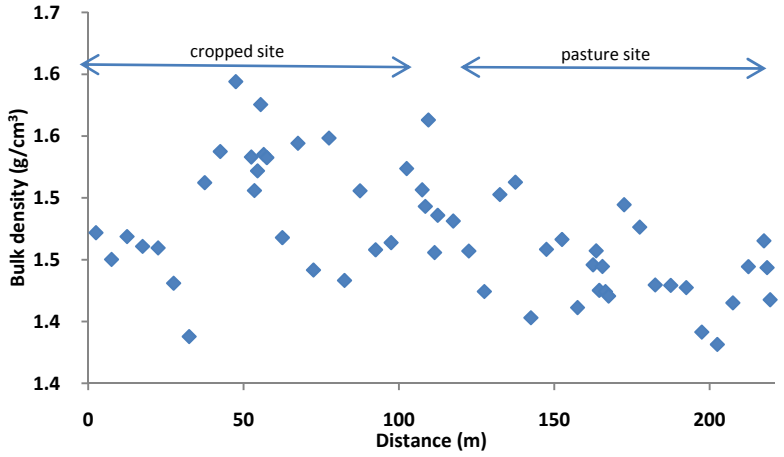

Fig. 3: Bulk density in cropland and pasture systems.

Soil water storage is an important component of soil water balance. Soil water content was measured on two days, September 16th and 17th, 2010. Soil moisture content for the top 0.1 m and soil water storage for the whole soil profile (1-m depth) showed that the cropped site had higher surface water content and water storage than the pasture site (Fig. 4). Average surface water content in the cropped site was 56% higher than that in the pasture site. Average water storage in the cropped site was 25% higher than that in the pasture site. The difference of surface water content and water storage between the two systems is due to evapotranspiration. The evapotranspiration in the pasture site was higher because there were no plants growing in the cropped site. Data shown in figure 4 indicates that there is a trend between surface water content and water storage in the cropland site (R^2=0.3) but this relation cannot be seen in the pasture site (R^2=0.02).

Gas diffusivity and air-filled porosity can be used to characterize soil structure in the two different soil management systems. Gas diffusivity is a reliable measurement for evaluating and assessing soil pores and pore continuity. Relative oxygen diffusion coefficient varied between 0.02 and 0.08 and air filled porosity varied between 0.06 and 0.16 cm^3/cm^3 in the pasture soil (Fig. 5) under 1/3 bar pressure (approximately field capacity). The result indicates that oxygen diffusivity is controlled by air-filled porosity. Average relative oxygen diffusion coefficient for ten soil cores taken from the cropped site was lower than that for the pasture soil (30 cores) by 22%. Measurements of oxygen diffusivity and air-filled porosity for the rest of the cropped site are in progress. Soils with higher gas diffusivity tend to be more structured and have more developed inter-aggregate pores. The results indicate that the pasture site has more structured soil than the cropped site. The result of gas diffusivity supports the result found with bulk density measurements. Pasture soil has higher porosity and higher oxygen diffusion coefficient. Moreover, based on the gas diffusivity result, it is expected that water transport in the pasture site is higher than that in the cropped site.

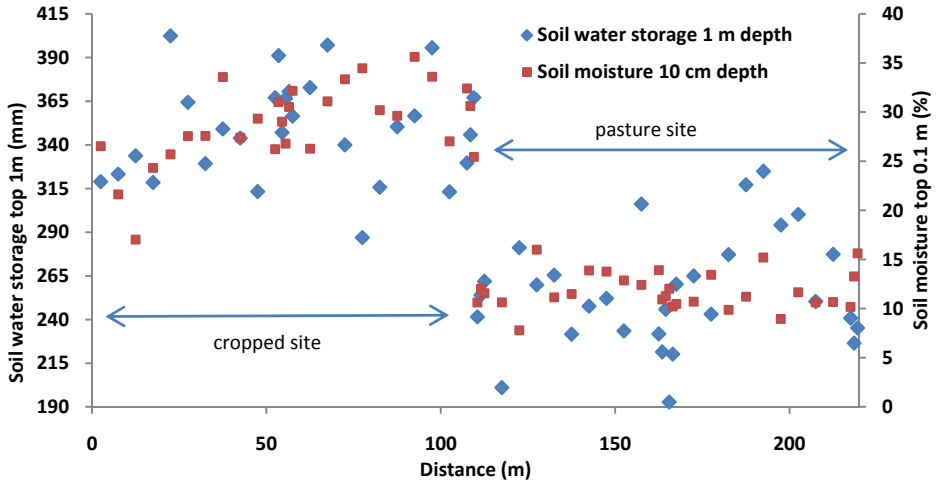

Fig. 4: Soil water storage, calculated by summation of ten depths (0-1 m depth), and soil water content for the top 0.1 m measured using the capacitance probe. Soil moisture content and storage measurements were not calibrated; therefore, the units are percentage. The measurements were taken on Sep. 16th and 17th, 2010.

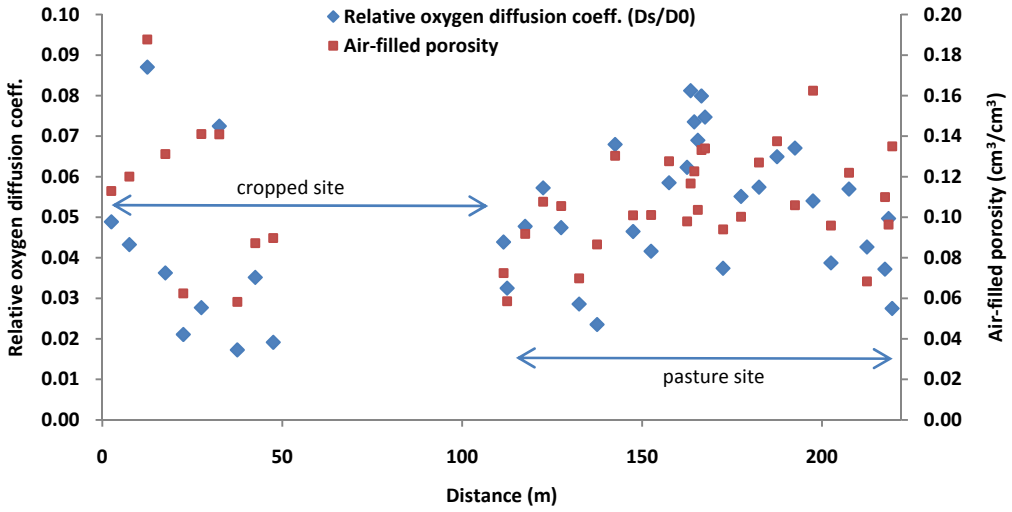

Fig. 5: Relative oxygen diffusion coefficient D_s/D_0 (relative to oxygen diffusion coefficient in the atmosphere, D_0=12.17 cm^2/min) and air-filled porosity under 3.33 m water pressure in both cropland and pasture systems.

As another method to assess potential impact of soil use and soil management on soil, CO_2 and N_2O fluxes from the soil surface were assessed. CO_2 and N_2O flux measurements were performed on two days (June 8th and 9th, 2010). Carbon dioxide flux was approximately 1000x the nitrous oxide flux (Figs. 6 and 7). Higher CO_2 flux occurred in the pasture site (average of 24 mg/m^2/min) than in the cropped site (average of 10 mg/m^2/min). There was no difference of average N_2O fluxes between the two soil use systems. There was diurnal variation of CO_2 and N_2O fluxes depending on soil temperature and water content in the cropped site, with greater flux as the soils became warmer and wetter over the sampling period. Carbon dioxide and nitrous oxide emissions reflect the microbial population and microbial activity in the soil. Microbial activity and carbon dioxide emission are affected by soil organic matter. It can be concluded that

structured soils have higher organic matter content and that results in higher carbon dioxide emission from the soil. Pasture soil has higher carbon dioxide emission because it has higher organic matter content and higher microbial activity and that indicates that it is a more structured soil.

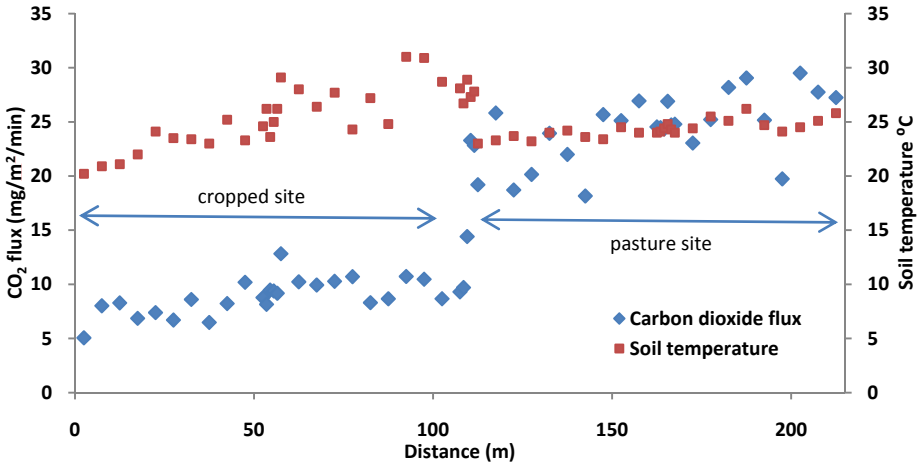

Fig. 6: Carbon dioxide emission and soil temperature (50 mm depth) in both land use systems measured on June 8th (cropped site) and June 9th (pasture site), 2010.

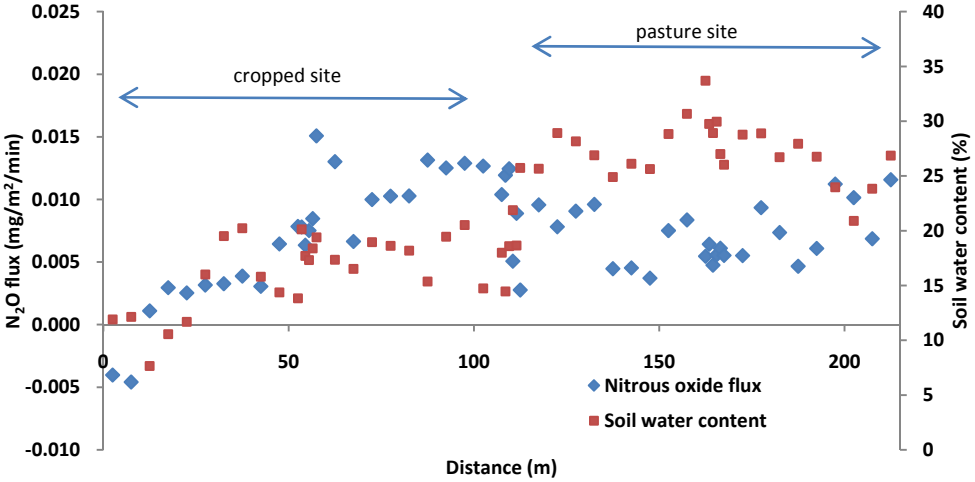

Fig. 7: Nitrous oxide emission and soil moisture (0.1 m depth) in both land use systems measured on June 8th (cropped site) and June 9th (pasture site), 2010.

Acknowledgements

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Caffeine as a marker for sewage contamination of Wilgreen Lake

Basic Information

Title:	Caffeine as a marker for sewage contamination of Wilgreen Lake
Project Number:	2010KY146B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Non Point Pollution, Water Quality, Methods
Descriptors:	limnology, anthropogenic markers
Principal Investigators:	Susan Godbey

Publication

1. Onjiko, Rosemary, and Susan Godbey, 2011, Determination of Caffeine as a Marker for Septic Tank Contamination of Wilgreen Lake, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 99.

Caffeine as a Marker for Sewage Contamination of Wilgreen Lake

Problem and Research Objectives

Wilgreen Lake in Richmond, Kentucky, is a freshwater reservoir that has been listed by both the Kentucky Division of Water, and the US EPA as nutrient-impaired. The lake drains a watershed with residential developments served by septic systems, cattle pasture, and some urban areas (sewered) in the City of Richmond. Uncertainty remains regarding the source of contamination. However, preliminary studies (Aquiar *et al*, 2008) suggest that a majority of the nutrients originate from human sources, most likely septic tank effluents from housing developments spanning the lakeshore. To investigate the contribution of human sources, caffeine was used as an anthropogenic marker to aid in determining the contribution of domestic activities to the impairment of the lake. Caffeine has been effectively used before to study the contribution of sewage treatment plants to contamination of surface waters (Buerge et al, 2003). Caffeine was chosen because of its specificity to human beings; it is the most consumed stimulant in beverages, soft drinks, pastries, and prescription drugs. Caffeine is also stable in surface waters. However, caffeine exists in the surface waters in very low concentrations, and therefore its quantitation calls for a very selective and sensitive method. Several methods have been used before to effectively quantitate caffeine (Verenitch et al, 2006). Liquid chromatography-tandem mass spectrometry (LC/MS/MS) has been used to determine the concentration of caffeine in surface waters (Cahil et al, 2004) and was the method chosen for the current study.

The purpose of this research was to establish whether domestic activities contribute to the contamination of Wilgreen Lake. To accomplish this, the levels of caffeine at key locations in the lake were determined to find if concentrations are higher near areas of suspected human contamination.

Methodology

Water samples were collected from different sections of the lake in February 2010 in 1-L pre-treated amber bottles, and refrigerated. The samples were then filtered in the lab, spiked with $^{13}C_3$-Caffeine internal standard, and pre-concentrated to 4 mL using Oasis HLB solid phase extraction cartridges within 48 hours of sampling. The concentrated samples were then spiked with $^{13}C_3$-Atrazine (instrument injection standard) and analyzed by injecting 20 µL into a Dionex HPLC. The separation was done using a multistep gradient of solvents (Table 1). The eluate was then passed through an Electrospray source, and detected using a Thermo LTQ XL mass spectrometer.

To determine the concentration of caffeine, an isotope dilution method was used. Five calibration standard solutions containing different concentrations of caffeine, 75ng/L 13C-Caffeine, and 50ng/L 13C-Atrazine were injected into the LC to make the calibration curve for caffeine (Figure 1). For MS/MS, two scanning segments were used to monitor the analytes' product ions (Table 2).

Table 1. Conditions for LC-MS/MS analysis

Instrument	Dionex HPLC, Thermo LTQ XL MS			
LC column	Waters Xtera C18, 10cm, 2.1 mm			
Ionization	Positive ESI			
Injection volume	20µL			
LC gradient program				
Time (min)	**Solvent flow mixture**	**LC flow rate**	**LC conditions**	
0	95% A : 5%B	0.15	Column	40°C
4	95% A : 5%B	0.25	Pressure	<345
22.5	12%A : 88%B	0.3	**MS conditions**	
23	100%B	0.3	Spray Voltage	5kV
26	100%B	0.3	Capillary temp.	240°C
26.5	95%A : 5%B	0.15	Capillary voltage	42V
33	95%A : 5%B	0.15	Tube lens	80V
A: 0.3% Formic acid and 0.1% Ammonium Formate				
B: 1:1 Methanol : Acetonitrile				

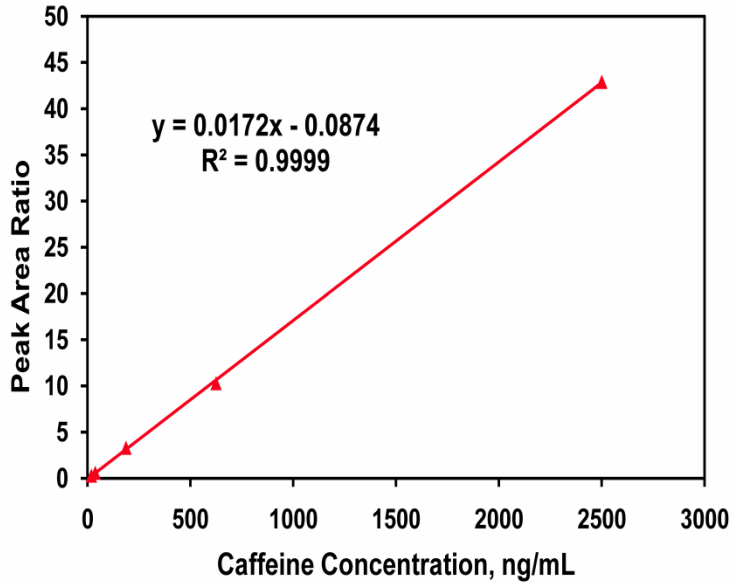

Figure 1. Calibration curve of caffeine generated using LC/MS/MS

Table 2. MS/ MS Scanning parameters

Analyte	Retention time	Parent (product) ion	Selected Ion (Isolation width)	Duration (min)	Scanned range	CID
Caffeine	9.5 mins	195 (138)	196.50 (4)	14.53	137.4 - 140.5	25
13C-Caffeine		198 (140)				
13C-Atrazine	16 mins	219.5 (177)	219.5 (1)	18.47	176.5 - 177.5	25

Principal Findings and Significance.

Detectable levels of caffeine were found at 6 of the 9 sites sampled. Figure 2 and Table 3 summarize the locations and concentrations of these samples.

The presence of caffeine in the upper Taylor Fork region suggests that the input could be from the adjacent neighborhood because caffeine was not found farther upstream at the inlet of the stream. On the other hand, the presence of caffeine at Old Town Branch inlet suggests that it could be from septic systems along the stream's course. The presence of caffeine at the dam (TF5d) could be due to direct human inputs (since this sample was collected from the dock) or from upstream. Representative samples need to be collected to establish if caffeine is indeed present throughout the entire lake. Additional work to gather more comprehensive data is underway.

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3. Cahill, D.J., Furlong, T.E., Burkhardt et al. Determination of pharmaceutical compounds in surface and ground water samples by solid-phase extraction and high performance liquid chromatography- electrospray ionization mass spectrometry. Journal of Chromatography. 2004, A 1041 pp 171-180.
4. Aguiar, T. A.; Borowski, W. S.; Layton, A. C.; and McKay, L. Using microbial distribution and abundance in a eutrophic lake as a tracer for nutrient inputs, Wilgreen Lake, Madison County, Kentucky, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, March 2008, p 51-52.

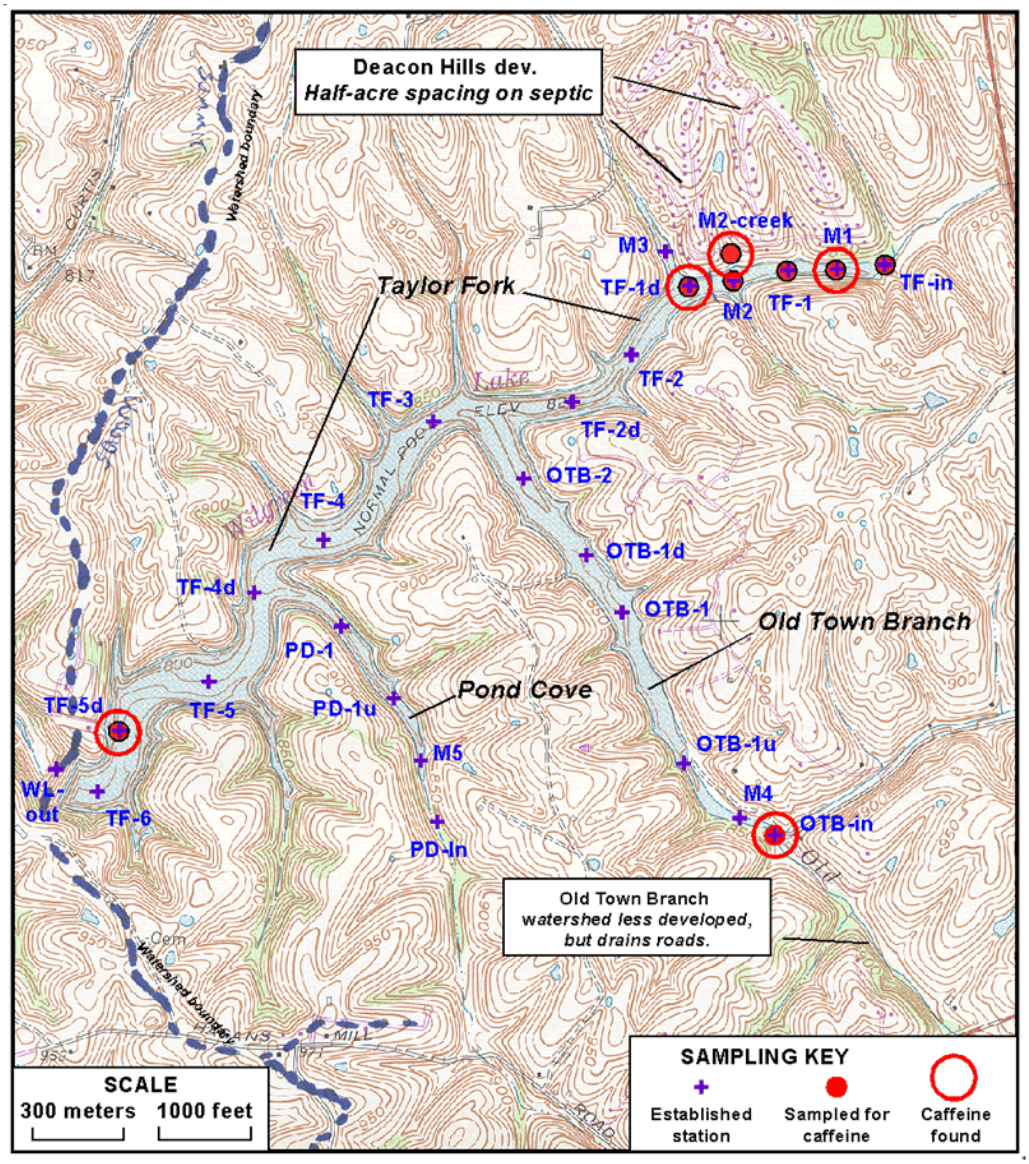

Figure 2. Sampling Locations

Table 3. Caffeine concentrations at sampled stations

Sample station	Caffeine concentration (ng/L)
OTB in	164
TF5d	329
M2	Not detected
M1	70
TF1d	89
TF1d-duplicate	133
TF1	Not detected
M2_Creek	346
TF_In	Not detected

Mobililty of Nitrogen-15 enriched E. coli within the Royal Spring basin, Kentucky

Basic Information

Title:	Mobililty of Nitrogen-15 enriched E. coli within the Royal Spring basin, Kentucky
Project Number:	2010KY147B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Groundwater, Water Quality, Methods
Descriptors:	pathogens, karst, groundwater trace
Principal Investigators:	Alan Fryar

Publication

1. Barton, A., and A. Fryar, 2011, Fate of Stable Isotope Label During Predation of N15-Tagged Wild-Type Escherichia coli by Protozoa, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 65-66.

Mobility of Nitorgen-15 Enriched *E. Coli* within the Royal Spring Basin, Kentucky

Problem and Research Objectives

Water can transmit pathogens such as O157:H7 *E. coli*, which is excreted through the feces of its main reservoir of ruminants. In 2002, Kentucky reported 2 deaths from O157:H7 that had been transmitted from untreated groundwater (Muniesa et al. 2006). Waterborne pathogens cause a variety of health problems, including diarrhea, dysentery, hemolytic-uremic syndrome, urinary tract infections, eye infections, and abscesses in the brain and lungs, just to name a few (Donnenberg 2002; Percival et al. 2004).

Much of Kentucky is underlain by limestone. These carbonate rocks have been partially dissolved by meteoric water and thus contain a variety of structures, including conduits, sinkholes, and springs. Inlets such as sinkholes and sinking streams allow water to travel directly to the aquifer without filtration. Karst aquifers are thus more prone to contamination than other sedimentary aquifers. In particular, karst aquifers are susceptible to pollution by particulate matter, including pathogenic microorganisms, which can be associated with sediment (Currens 1999).

Fecal indicator organisms are used to determine if potentially harmful bacteria are likely to be present. *Escherichia coli* (*E. coli)* is considered to be the best indicator for fecal contamination at this time. Ward (2008) found that ^{15}N-tagged *E. coli* behave differently than soluble fluorescent dyes or latex microspheres during a tracer test in a karst aquifer. It was also shown that in contrast to dye, both bacteria and microspheres can emerge during subsequent storm events after initial injection, but bacteria are flushed from the aquifer more quickly than microspheres. Bacteria may travel with suspended solids during periods of high velocity flow (such as storm flow), and settle during periods of slower velocities.

This research examined attenuation of the stable isotope signal within bacterial predators. Signal loss is anticipated during a future groundwater trace along Cane Run (approximate distance of 10 km from injection site to Royal Spring). One of the anticipated losses of the nitrogen signal during the trace is predation of the tagged bacteria. Information from this study will be used in a future tracer test conducted in the Cane Run watershed utilizing ^{15}N-tagged bacteria.

Objective 1: Selection of protozoa. Protists are an important component of microbial ecosystems in aquifers, acting as predators of bacteria and other microorganisms. Bacteria may be the sole nutrient source for protists in natural aquatic systems (Drake and Tsuchiya 1977). Protozoa have been selected based on the likelihood of being found in a karst system and choice of *E. coli* as a food source. *Tetrahymena* can be used as a representative protozoan predator, as it is considered typical for its phylum, with nutrition, morphology, and reproduction similar to other protozoa (Hill 1972). *Tetrahymena pyriformis* is a freshwater ciliate that is present where plant or bacterial decomposition occurs, such as water contaminated by fecal material. Although dimensions are commonly listed as 50 × 30 μm in the literature, sizes ranging from 25-90 μm are possible (Bick 1972).

Although flagellates are the most common protists in typical clastic aquifers, ranging from 10^2 per gram of dry aquifer material to several orders of magnitude higher in aquifers exposed to organic pollution, ciliates may play an important role, with *Colpoda steinii* being a major constituent (Novarino et al. 1997). *Colpoda steinii* is a ciliate found in all types of water with a preference for high bacteria counts. Although these organisms may struggle with

temperatures common in karst, they can tolerate conditions of low dissolved oxygen and high NH_4^+. *C. steinii* are typically 15-60 μm (Bick 1972).

Objective 2: Capture and separation of *E. coli* and protozoa for isotope analysis. *E. coli* are 2 × 0.5 μm, which is significantly smaller than *T. pyriformis* and *C. steinii*. For isotope analysis, protozoa were captured on glass fiber filters with nominal pore sizes of 2.7 μm, while most *E. coli* remained on filters of 0.7 μm.

Objective 3: Isotopic analysis. Protocol generally followed those of Ward (2008) and Warden (2010). Samples were filtered onto glass fiber filters, which were weighed, dried, and analyzed using an elemental analyzer, gas chromatograph, and isotope ratio mass spectrometer.

Methodology

Tetrahymena pyriformis were obtained from Carolina Biological Supply and maintained in tubes of proteose media until use. Tetrahymena medium was prepared by mixing 5.0 g proteose peptone, 5.0 g tryptone, and 0.2 g K_2HPO_4 in 1.0 L deionized (DI) water before autoclaving. *Colpoda steinii* Maupas (ATCC® 30920) were established from frozen culture and kept in T-25 tissue culture flasks with a media of Cerophyll and DI water. Aliquots of log phase *E. coli* were injected into the flasks as prey. Subcultures were maintained until use.

Wild-type *E. coli* used by Warden (2010) were enriched with ^{15}N using the methods described in Ward (2008). Isotopically enriched NH_4SO_4 (98 atomic % ^{15}N) was diluted with non-enriched NH_4SO_4 to generate a more manageable $\delta^{15}N$ value for this research. This was achieved by mixing 0.003 g isotopically enriched $^{15}NH_4SO_4$ with 2.997 g NH_4SO_4 while creating the M9 media.

Isotope abundances can be expressed in delta notation, which compares the ratio of the rare isotope to the common isotope in a sample to that of a standard. For nitrogen, the international standard is air, which has a $^{15}N/^{14}N$ value of 0.0036765 (Fry 2006). Delta notation is measured in units of per mil (‰) and is calculated using the following formula:

$$\delta^{15}N(‰) = \left[\frac{{}^{15}N_{sa} / {}^{14}N_{sa}}{{}^{15}N_{std.} / {}^{14}N_{std.}} - 1 \right] \times 1000$$

Filter-sterilized water was collected from Royal Spring and placed in Erlenmeyer flasks (500 mL each). Triplicates were made of each of the following conditions: sterile Royal Spring water only, ^{15}N-enriched *E. coli*, *T. pyriformis* + non-enriched *E. coli*, *C. steinii* + non-enriched *E. coli*, *T. pyriformis* + enriched *E. coli*, and *C. steinii* + enriched *E. coli*. Because control conditions of protozoa were provided with non-enriched *E. coli* to keep them alive, triplicates were also made of natural *E. coli* in 250-mL quantities. IDEXX was used to obtain a most probable number (MPN) for *E. coli* enumeration. A serial dilution was made and Colilert® snap packs were added to each dilution. Dilutions were poured into a Quanti-Tray/2000 and run through a Quanti-Tray sealer. Trays were incubated at 35°C for 24 hours. Yellow cells and cells that fluoresce under long-wave UV light were counted to obtain the MPN. Flasks were stored in the dark at approximately 14°C in a wine chiller.

Samples were taken at 0 and 7 days. Prior to filtration, filters were ashed in a furnace at 550°C to remove any residual nitrogen that may have been present from manufacturing or shipping. Protozoa and bacteria were separated through filtration of 100-mL aliquots for each sample. Protozoa were captured on Whatman Grade GF/D glass fiber filters, which have a pore size of 2.7 μm. Filtrate was passed through glass fiber filters (nominal pore size 0.7 μm) under vacuum and captured in test tubes in order to capture bacteria. Filters were folded in half, placed in foil packets, which were opened so that the filters were exposed, and dried in an oven at 40°C for 12 to 24 hours. Packets were then closed, weighed, and packaged into 9×10 mm tin capsules. These were combusted at 980°C in an elemental analyzer (EA), passed through a gas chromatograph, and analyzed on an isotope ratio mass spectrometer (IRMS). Values were compared to a standard of acetanilide (ACE) and an in-house reference of fish protein (DORM 3).

Details of the first trial are omitted here, as initial results showed cross-contamination of ^{15}N. Steps were taken to assess the origin of contamination. The following possibilities were analyzed: blank filters, volatilization within the oven, the filtration apparatus, gloves, swabs taken within the autoclave, incomplete combustion within the reaction tube of the EA, and low voltage in the mass spectrometer. Results indicated that one autoclave had become contaminated with ^{15}N, which was passed on to the filter sterilized water. A second trial was conducted, which yielded the results described below.

Principal Findings and Significance

Trial 1 showed cross-contamination of ^{15}N throughout the samples. This can be seen in the highly elevated $\delta^{15}N$ values prior to any addition of enriched bacteria (Table 1, Fig. 1). This can also be seen in data obtained for Royal Spring water in December's calibration run as compared to values obtained in Trial 1 (Fig. 2). Natural samples should be close to 0 ‰ but can reach as high as 20 ‰ if there is organic/fecal contamination.

Trial 2 showed evidence that the stable isotope signal is passed on to predators. *T. pyriformis*, which is considered a representative organism of the protozoa phylum, showed a statistically significant increase in ^{15}N over the 7 day period (Fig. 3). *C. steinii* did not show this drastic change, which may be explained by environmental stress and the encysted form that was seen during enumeration. Elevated $\delta^{15}N$ values for time zero for protozoa mixed with enriched bacteria may be a result of not rinsing all of the enriched bacteria from the filter, as samples taken at t=0 were filtered prior to the ingestion/clearing rate times listed in the literature in order to provide delta values representative of organisms prior to assimilation.

E. coli populations decreased in the presence of a predator over the course of the trial, while protozoa populations increased (Table 3), supporting the hypothesis that these predators were feeding on bacteria and assimilating the isotope signal into their biomass. Control groups of *E. coli* stayed fairly constant, indicating that population decrease was a result of predation.

Although the ^{15}N signal from enriched *E. coli* was assimilated into protozoan biomass, this should not significantly influence the ability to detect the ^{15}N signal within a trace. This should, however be considered when trying to calculate the number of bacteria present and making interpretations about their movement within a conduit.

The ease of ^{15}N cross-contamination needs to be in the forethoughts of any investigator using this application in future studies. Enumeration of bacteria and protozoa needs to be performed in conjunction with isotopic data obtained.

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Figures and Tables

Table 1. Elevated $\delta^{15}N$ (‰) values obtained in Trial 1. Masses on filters and amplitude have been included.

Sample	Mass (mg)	Ampl 28 (mV)	$\delta^{15}N$ (‰ vs. air)
ACE	0.982	4807	-0.504
ACE	1.105	5457	-0.616
ACE	1.097	5285	-0.516
Royal Spring Water	5.367	253	1728.376
E. coli	0.149	317	1233.998
T. pyriformis	0.02	782	483.536
C. steinii	0.034	600	614.158
T. pyriformis fed enriched *E. coli*	41.115	592	1096.981
Enriched *E. coli* fed to *T. pyriformis*	0.36	258	1330.038

Table 2. $\delta^{15}N$ (‰) obtained for samples in Trial 2.

Sample	Avg. $\delta^{15}N$ (‰ vs. air)	
	t=0 days	t=7 days
Royal Spring Water		4.443
Non-enriched *E. coli*	16.080	5.730
T. pyriformis	22.882	23.376
C. steinii	17.460	25.475
E. coli fed to *T. pyriformis*	47.501	56.949
E. coli fed to *C. steinii*	58.108	54.583
T. pyriformis fed enriched *E. coli*	93.296	181.415
C. steinii fed enriched *E. coli*	114.764	120.179
Enriched *E. coli* fed to *T. pyriformis*	632.090	
Enriched *E. coli* fed to *C. steinii*	649.622	
Enriched *E. coli*	732.483	704.281

Table 3. Population data for protozoa and *E. coli* from Trial 2.

Control Conditions

Organism	Population at t=0 (cells/mL)	Population at t=7 days (cells/mL)
E. coli (enriched)	5.67×10^8	1.18×10^8
T. pyriformis	1.57×10^4	5.42×10^4
E. coli (non-enriched)	2.14×10^9	4.10×10^4
C. steinii	2.87×10^3	1.44×10^4
E. coli (non-enriched)	2.14×10^9	3.93×10^5

Experimental Conditions

Organism	Population at t=0 (cells/mL)	Population at t=7 days (cells/mL)
T. pyriformis	1.57×10^4	4.58×10^4
E. coli (enriched)	5.67×10^8	9.70×10^4
C. steinii	2.87×10^3	2.30×10^4
E. coli (enriched)	5.67×10^8	2.06×10^6

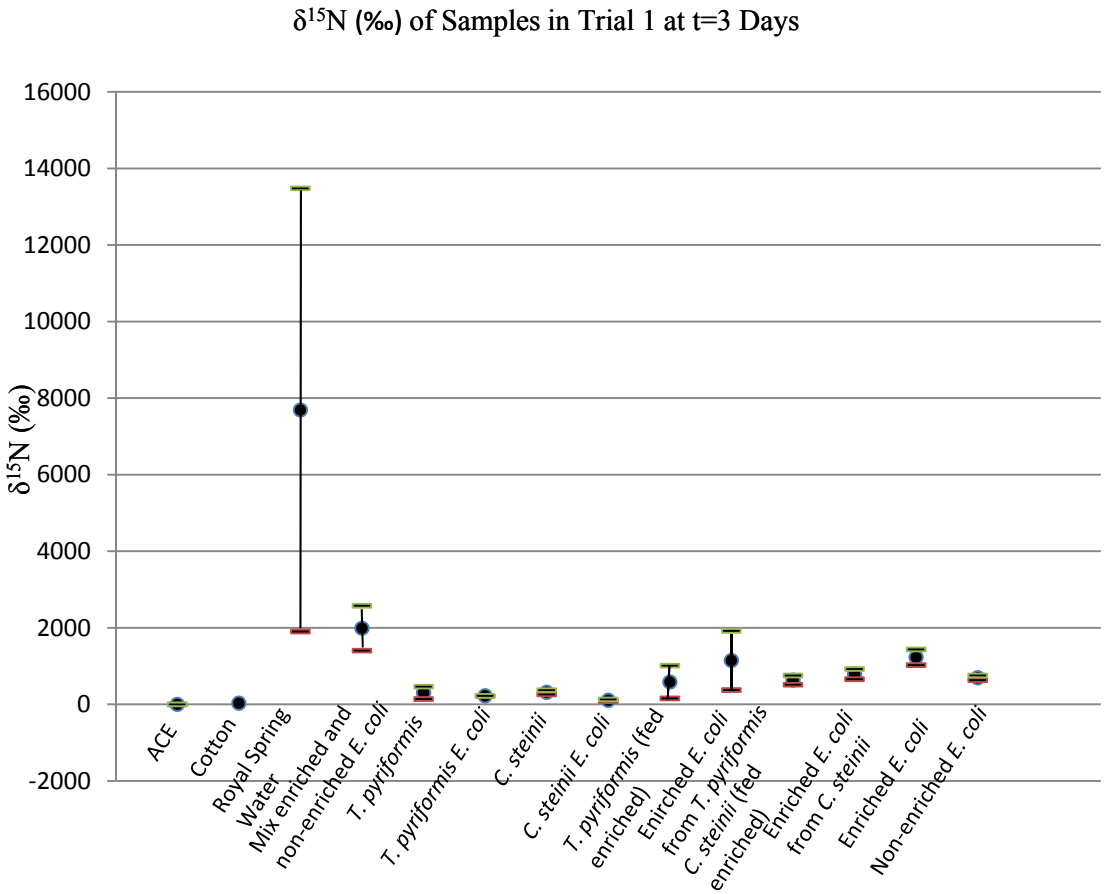


Figure 1. Elevated $\delta^{15}N$ (‰) values obtained in Trial 1.

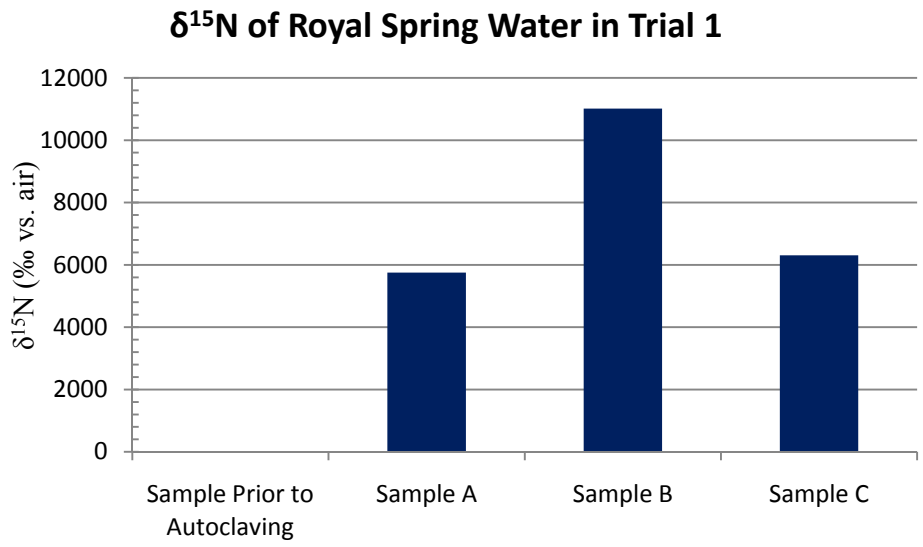


Figure 2. Comparison of $\delta^{15}N$ (‰) of sterilized Royal Spring water in December, prior to autoclaving, and after autoclaving in February.

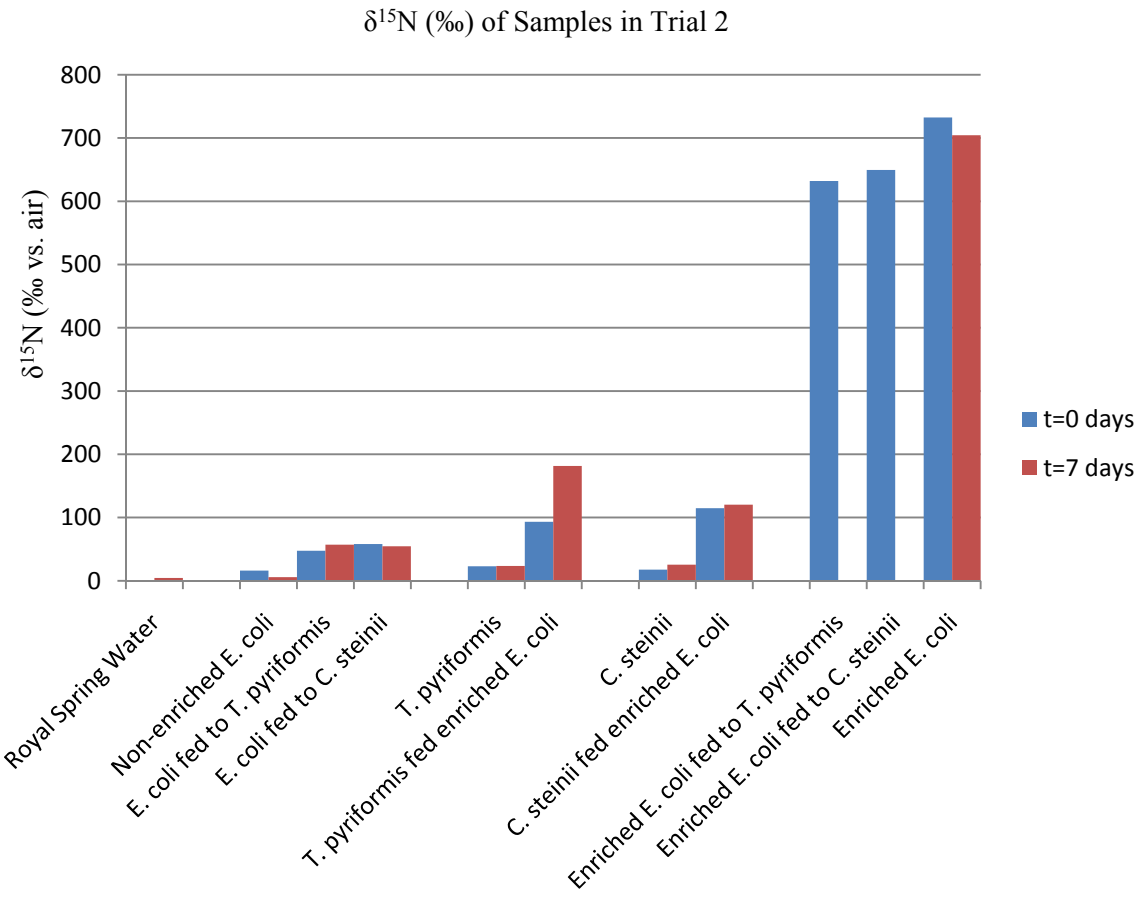


Figure 3. Chart comparing $\delta^{15}N$ (‰) obtained at time zero and seven days for Trial 2.

Filtration column for the covalent removal of arsenic from water

Basic Information

Title:	Filtration column for the covalent removal of arsenic from water
Project Number:	2010KY148B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Water Quality
Focus Category:	Methods, Toxic Substances, Water Quality
Descriptors:	treatment, reagent, drinking water
Principal Investigators:	David A. Atwood

Publications

1. Jana, Partha, and David Atwood, 2011, Aqueous Arsenic Removal by Thiol-Containing Filtration Columns, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 27.
2. Atwood, David, and Partha Jana, 2010, A Permanent Solution to Environmental Merury Contamination, in Proceedings 31st Annual Meeting of the Society for Environmental Toxicologists and Chemistry (SETAC), #474.

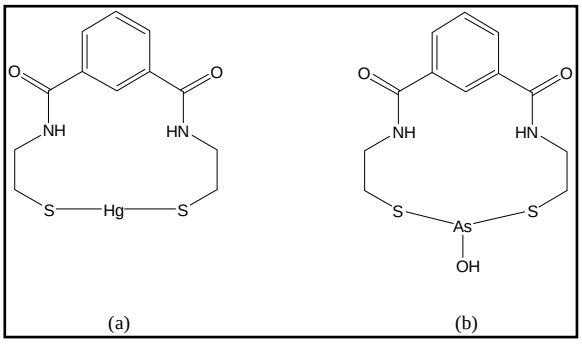
Filtration Column for the Covalent Removal of Arsenic from Water

Problem and Research Objectives

The contamination of surface and ground water with metals and metalloids is a major environmental concern. Arsenic is a widespread surface, ground, and drinking water contaminant that adversely affects human health due to its acute and long term toxicity. It is poisoning the drinking water in more than 70 countries with major concerns in India, Bangladesh, the United States, China, Vietnam (and other Asian countries) and Argentina. Arsenic ingestion causes arsenicosis, cancer, and a recent study in the U.S. linked arsenic consumption at part-per-billion levels (ppb) to cardiovascular disease and diabetes. This study was conducted in a metropolitan area in the U.S. and the arsenic source was drinking water.

The Environmental Protection Agency, the National Research Council, and the World Health Organization have set the maximum contaminant level (MCL) for arsenic at 10 ppb. Drinking water in many countries is commonly drawn from wells, which is good in that such water is generally free of microbial contamination that may exist in surface water. However, in these countries, groundwater is often contaminated with naturally-occurring arsenic. This research project address the critical need of providing a filtration material that can remove arsenic from anoxic drinking water.

N,N'-bis(2 mercaptoethyl)isophthalamide) has emerged as the preeminent reagent for the complete precipitation of mercury from water (< 5 ppb) through the formation of a neutral, water-insoluble compound having strong Hg-S covalent bonds (~ 200 kJ/mol) (see figure (a) below).[1-5] Given the stronger covalent bonding found for arsenic and sulfur (~ 300 kJ/mol) as compared to mercury, it was anticipated that the compound would be equally effective in removing As(III) from water. This was found to be the case, and provides the impetus for the current research program. The compound is abbreviated, $BDTH_2$, in the literature but is known commonly as "B9."

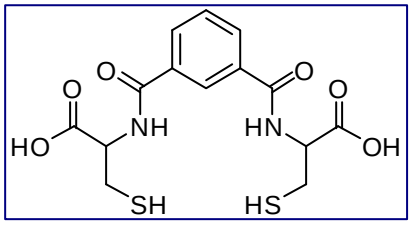


B9 removes As(III) from water, through covalent bond formation (see figure (b) to right) and subsequent precipitation with no subsequent leaching at pH values from 4 – 8. B9 binds only

As(III), the primary form of arsenic in anoxic water. This was demonstrated in studies with 50:50 mixtures of As(III)(150 ppb) and As(V) (150 ppb). The bound arsenic can be removed to regenerate the ligand by treatment with acid at a pH below 4. Moreover, the efficiency of B9 in removing As(III) from groundwater was demonstrated in field studies conducted in 2005 and again in February 2008 in West Bengal, India, for 48 separate samples in four locations. Well-water containing arsenite (As(III)) at levels of 200 – 40 ppb was passed through hand-held filtration columns containing B9 mixed with sand. The resulting arsenic levels in the water were below the limit of detection, < 5ppb, of the graphite furnace atomic absorption analyzer.

This project focused on creating a solid-supported reagent having the arsenic capture ability of $BDTH_2$ to be used as filtration bed or in a filtration column to remove arsenic (III) from groundwater and thereby provide clean drinking water. The objective was to covalently attach a mono-carboxylic acid derivative of B9 referred to commonly as "AB9" (for "acid B9"), to silica beads in order to prepare a column packing material with sufficient flow rates for point-of-use drinking water treatment as a proof-of-concept. The next stage of the program (beyond the scope of the current project) will be to covalently attach AB9 to the highly porous, hydrophilic polymer hydroxyethyl methacrylate (HEMA).[6, 7]

The research was conducted in two phases, both of which were successful. The Phase I study demonstrated the synthesis of the mono-carboxylic acid derivative of B9, AB9 (the structure of AB9 is shown in the adjacent figure. Preliminary work has demonstrated the synthesis of this compound but the reaction needed to be optimized to obtain larger amounts of the compound. Once this was accomplished (in the first month) the compound was attached to silica beads through known techniques.[8-10]



Methodology

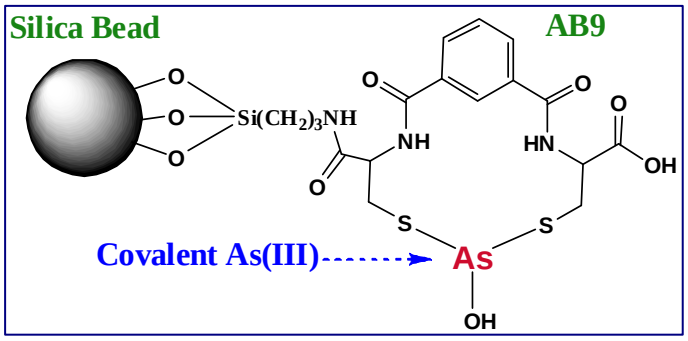
SiAB9 was prepared in two steps. The first step was the modification of silica-60 surface hydroxide groups with γ-aminopropyltriethoxysilane to form Si-O-Si bonds with pendant propylamine groups. The material was then treated with chlorotrimethylsilane to protect the potentially reactive silanol groups by converting them to trimethylsiloxy groups. With the

application of AB9 in heated ethanol, the AB9 carboxyl groups condensed with the free propylamines, forming amide (-CONH-) linkages.

The IR spectra of SiAB9 showed the characteristic absorption for SH at 2545 cm^{-1}. A broad peak was observed at 3399 which could be attributed to either the remaining COOH group on AB9 or to the amide NH. Silica-60 (Si-60) has a sharp absorbance at 3447 cm^{-1} for the O-H bond and weak intensity absorption for the Si-O at 963 cm^{-1}. Both of these absorbances were absent in SiAB9, confirming that the SiOH group had been displaced by amine and treatment with Me_3SiCl (to create Si-O-$SiMe_3$ linkages). The AB9 loading on SiAB9, as determined by ICP-OES analysis for sulfur, was found to be 0.712- 0.773 mmol sulfur per gram SiAB9 beads or an AB9 loading of 0.356 - 0.386 mmol per gram of beads for the 20 g scale preparations.

The Phase II portion of the study demonstrated that SiAB9 was capable of removing As(III) from water. This was conducted by performing a series of "batch" tests where a measured amount SiAB9 was mixed with a solution containing As(III) at a concentration of ~ 200 ppb. As demonstrated in Tables 1-3, 100 % arsenic removal was achieved by adding the SiAB9 in about a 400-fold excess compared to the arsenic for 12 h. When used in a filtration column SiAB9 would, in effect, be in a near infinite excess for each molecule of arsenic(III) passing through the column, until a significant portion of the arsenic binding sites in the column were occupied. These results also indicate that there is an optimum capture at pH 7 when less of an excess of SiAB9 is used.

Principle Findings and Significance



AB9 immobilized on silica may be used as a filtration column packing material. This project provided the foundation needed to pursue this goal by demonstrating the synthesis of SiAB9 and the utility of the material to remove 100 % of the arsenic(III) from an aqueous solution. Within a filtration column one gram of SiAB9 would treat approximately 60 L of water contaminated by arsenic(III). With additional funding, the program plans to create a demonstration filtration column packed with SiAB9 and test it against arsenic(III) through a

range of flow rates. If successful, the arsenic filtration column could then be used to remove As(III) from water drawn from contaminated aquifers.

Table 1. Arsenic(III) Removal by SiAB9 at pH 5

Sample ID	Conc. (µg/L)	Stdev.	% Capture
As stock	208.45	± 10.86	N/A
0.2 g Si AB9	115.40	± 7.27	44.6%
0.6 g Si AB9	< 5.0	N/A	100%

Table 2. Arsenic(III) Removal by SiAB9 at pH 7

Sample ID	Conc. (µg/L)	Stdev.	% Capture
As stock	192.80	± 0.79	N/A
0.2 g Si AB9	51.50	± 0.14	73%
0.6 g Si AB9	< 5.0	N/A	100%

Table 3. Arsenic(III) Removal by SiAB9 at pH 9

Sample ID	Conc. (µg/L)	Stdev.	% Capture
As stock	218.20	± 5.02	N/A
0.2 g SiAB9	156.80	± 10.98	28.1%
0.6 g Si AB9	< 5.0	N/A	100.0%

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Flood impact due to dam failure: an assessment of current studies and the appropriateness of the methodologies applied to the Dix Dam

Basic Information

Title:	Flood impact due to dam failure: an assessment of current studies and the appropriateness of the methodologies applied to the Dix Dam
Project Number:	2010KY149B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY Sixth
Research Category:	Climate and Hydrologic Processes
Focus Category:	Floods, Models, Water Supply
Descriptors:	HEC-RAS, BreZo4.0, simulations
Principal Investigators:	Scott Yost

Publication

1. Cantrell, Allen, and Scott Yost, 2011, Flood Impact Due to Dam Failure: An Assessment of Current Studies and the Appropriateness of the Methodologies Applied to the Dix Dam, in Proceedings Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, p 21-22.

Flood Impact Due to Dam Failure: As Assessment of Current Studies and the Appropriateness of the Methodologies Applied to the Dix Dam

Problem and Research Objectives

The aging condition of the Dix Dam has come under more scrutiny over the past few years. There is concern as to the impact (economic, society, recreational, safety, etc) if the dam were to fail. This study was to look at the downstream impact to the communities and people living along the Kentucky River if the dam were to fail. Previous studies were conducted [Dix Dam Breach Study, Kentucky Utilities] to help understand the extent of the impact and the time available to give the communities warning. The past studies relied on widely accepted one-dimensional (1D) models (HEC2/HEC-RAS) (U.S. Army Corps of Engineers) to simulate the flood wave resulting from a dam break/breach. While the models are widely accepted, they are based on dated techniques and decades old understanding. The purpose of the research was to determine the adequacy of a 1D water flow analysis model compared to a two-dimensional (2D) model. Previous models have been completed using 1D modeling, but have been questioned based on the limited flow modeling capabilities. This study focused on the potential downstream flooding impact along the Kentucky River if the dam were to fail. The following were the procedures for the research plan.

1. Review the past studies to categorize the methodologies, input data completeness, and assumptions.
2. Relate the information categorized in (1) to the outcomes/results from the various studies to ensure appropriateness and completeness.
3. Compare the advantages and disadvantages of 1D modeling to current 2D dam break/breach models to see if the 1D limitations warrant more advanced techniques.
4. Depending on the results of (3), establish the framework for performing the same analysis, but with current 2D models.

The investigation was oriented towards determining whether or not a 1D simulation provides an adequate amount of accuracy or if a current state-of-the-art 2D model will be warranted. The need for this information comes from the direct impact a dam breach will have on human lives, property, and the environment downstream. It is anticipated that a 2D simulation will be necessary to formally quantify the differences, similarities, and applicability of both 1D and 2D modeling results. The investigation determines model applicability with the goal of producing the most accurate/reliable results for policy makers. The water surface elevations and floodwave propagation calculated in the simulations will have a direct impact downstream of the dam breach, which may require changes in floodplain mapping and warning time for affected communities.

This study compares the accuracy and efficiency of the widely used 1D model developed by the US Army Corps of Engineers® Hydrologic Engineering Center, HEC-RAS (U.S. Army Corps of Engineers), and a cutting edge 2D model, BreZo4.0 (Sanders). Both models are free to download which makes them easily obtainable by the engineering community. The study

compares the modeling of linear one-dimensional rectangular ideal channels, two-dimensional ideal rectangular channels with a confluence, as well as a linear channel with transitions between rectangular and trapezoidal shaped cross sections. It is necessary to test simplified models before progressing to natural topography so that differences in the results developed by the computation methods can be noted. Since the Dix and Kentucky Rivers are continuous channels with constantly changing properties, it is important to isolate the differences in the computation methods from the differences created by the channel properties. Using identical ideal rectangular channels, differences resulting from the 1D and 2D calculations are easier to recognize.

When conducting hydraulic modeling of natural topography, a finite area of interest is chosen. Since the modeling is essentially cutting out a piece of the topography, boundary conditions need to be specified at each of the cut lines. These boundary conditions will identify which areas allow for water to pass through and which ones act as barriers so that the real world situation will be represented as accurately as possible. Also, for numerical modeling stability issues associated with any hydraulic modeling, initial conditions will need to be specified as well. A well-known problem with many hydraulic modeling systems is the behavior of water on dry cells in the finite area. The initial condition settings reduce this problem by decreasing the effects of a shock in the system. BreZo4.0 incorporates a revolutionary method (Begnudelli & Sanders, 2006) for dealing with this problem in relation to the unstructured grid.

Methodology

Identical hydrographs, initial conditions, boundary conditions and channel properties were used for each simulation completed in each model during the investigation. The first simulation compared was a single straight rectangular channel configuration with a constant slope. This was followed by rectangular channels with lateral inflow configuration as well as a straight channel with transitions between rectangular and trapezoidal shaped channels. Additional simulations modeling inline structures such as locks, dams and bridge foundation piers as well as a natural topography will provide more information on flow patterns as well as the wetting and drying of computation cells.

Since all input parameters were the same, the dissimilarities in the simulations will come from the differences in the computation domain/grid and the governing equations used in the 1-dimensional and 2-dimensional modeling. HEC-RAS solves the 1D energy equation between cross sections perpendicular to the main direction of flow, whereas BreZo4.0 solves the 2D shallow-water (momentum) equations across an unstructured triangular grid or an optional structured quadrilateral grid.

Principal Findings and Significance

The single straight rectangular channel simulation showed identical results in both models. This was expected since this was essentially a 1D hydraulic simulation. Figure 1.a. shows the floodwave propagation downstream at a time of 30 minutes into the simulation, just before the floodwave exits the channel. Figure 1.b. shows the same floodwave propagation versus time. The results depicted are expected since this 1D situation is consistent with HEC-

RAS capabilities. Since the 2D calculations are a superset of the 1D situation, it is expected the BreZo4.0 would have no problems with the same analysis. The results are basically identical.

The rectangular channel with lateral inflow simulation started to show differences in the results downstream of the confluence. Upstream of the junction, the flow is basically identical to the first simulation, but once the junction is reached, the floodwave propagation differs in the models. This configuration slightly modifies the 1D configuration by adding a confluence not parallel with the main channel flow. This starts to show the differences between 2D and 1D computations since the flow upstream of the junction is essentially in one direction, but once it reaches the confluence, the flow has a choice of which direction it will travel therefore creating a 2D flow field. These results were expected since BreZo4.0 can more accurately represent the momentum carried in the floodwave through the junction. This results in the floodwave in BreZo4.0 being further downstream of the junction than in HEC-RAS at a given time as well as the floodwave propagating upstream of the junction that is not as far upstream of the main channel at the same time. The results from this simulation in the channel downstream of the junction can be seen in Figure 2.a. This figure shows that near the junction, a higher water surface elevation is computed by BreZo4.0 as well as a flood wave that propagates faster downstream compared to that of the HEC-RAS results. Figure 2.b. shows the results from this simulation in the upstream portion of the channel after the junction. This figure shows the floodwave propagating upstream more slowly in BreZo4.0 than that of HEC-RAS. The floodwave propagation for the downstream continuation in the main channel is visible in Figure 2.c. Overall the floodwave is identical for each model until the junction is reached. BreZo4.0 produces a floodwave that moves faster downstream than HEC-RAS after the junction is reached. The junction, being 2D in nature, is handled more naturally by a 2D model than a 1D model with internal boundary conditions.

The straight channel with transitions between rectangular and trapezoidal shaped channels also showed differences between the models (Figure 3). BreZo4.0 shows a deeper and sharper hydraulic bore (backwater affect) upstream of the transition from rectangular to trapezoidal, slightly slower flow in the trapezoidal channel and a more distinct hydraulic jump downstream of the transition back to the rectangular channel. These results were also expected since HEC-RAS has a tendency to smooth out shocks in the system due to numerical stability issues. Because of this, the hydraulic bore and hydraulic jump in HEC-RAS are not as pronounced as they appear in BreZo4.0. Figure 3.a. shows the water surface elevations after the floodwave has already passed through the entire channel, but at the point where the hydraulic bore and hydraulic jump are most distinct. Figure 3.b. shows the floodwave propagation versus time for the simulation. The zones where the BreZo4.0 results are greater than the HEC-RAS results indicate that the flow is moving quicker downstream and vice versa. It is important to note that for a given flow, as the velocity decreases, the water elevation increases. Natural topography situations include irregular channels that get wider (expansion) and smaller (contraction) as the channel progresses. These transitions will act as energy dissipaters through

interaction with the channel bed and walls which slow down the flow resulting in increased water surface elevations. The differences between the two models in these figures are from a combination of 2D flow around the transitions as well as HEC-RAS smoothing out the shock of the hydraulic bore and hydraulic jump.

Additionally, with BreZo4.0, the entire area of interest can be represented and modeled, but HEC-RAS will only be able to represent slices of the channel every so often. This immediately poses a problem in the natural topography applications for HEC-RAS. But this also brings up another issue of computation time related to stability and accuracy. In order for all numerical models to remain stable and not develop incorrect values, the computation time step needs to correspond to an appropriate length interval. Normally, one would suspect that a more refined grid would always increase the accuracy, but in fact if the time step is not adjusted at the same time, then there is no added benefit to results because refined (smaller) grid nodes will be smeared over if the time step is too large. The Courant Number is one such indicator of numerical stability that can be calculated based on the flow velocity, time step and the length interval. BreZo4.0 uses the Courant Number for each calculation to ensure stability and if this number becomes unstable at any point, the simulation will cease and the time step or length interval will have to be adjusted to accommodate numerical stability. HEC-RAS has similar indicators of numerical instability, but the smallest time step possible in HEC-RAS is 1 second, which limits how close the cross sections can be. Since BreZo4.0 largest possible time step is 1 second, this allows for a more compact grid to be used for computation. However, most current digital elevation models that are readily available are 30 meter data, meaning that an increase in computational grid resolution smaller than this cannot add significant information.

Seeing the results from these simplified configurations, it is expected that larger, more compounded differences will be noticeable in the natural topography application to the Dix River and Kentucky River. This could result in the floodwave propagating downstream faster than modeled before in HEC-RAS indicating a shorter warning time for downstream communities as well as deeper flooding than has been modeled before. This research will continue into the summer of 2011 where the Dix River and Kentucky River application will be modeled as part of a thesis research.

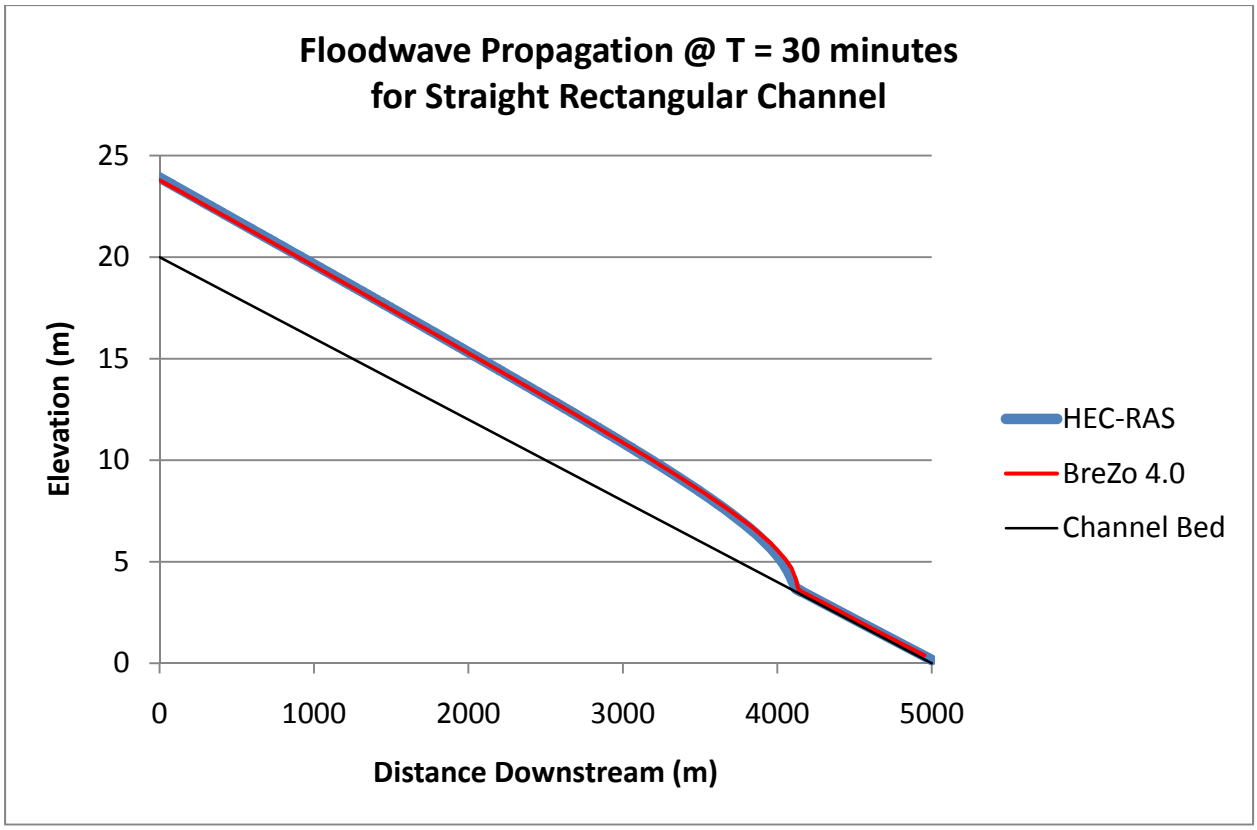

Figure 1.a. - Straight rectangular channel simulation

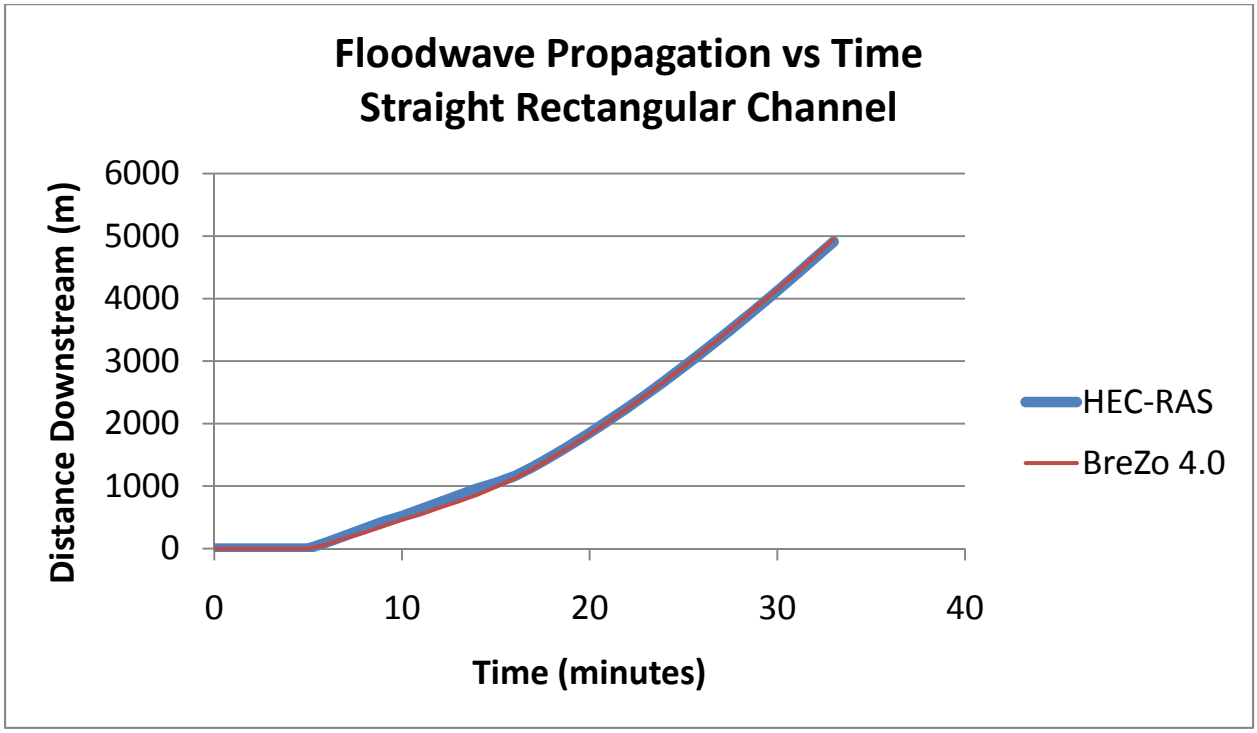

Figure 1.b. – Straight rectangular channel simulation

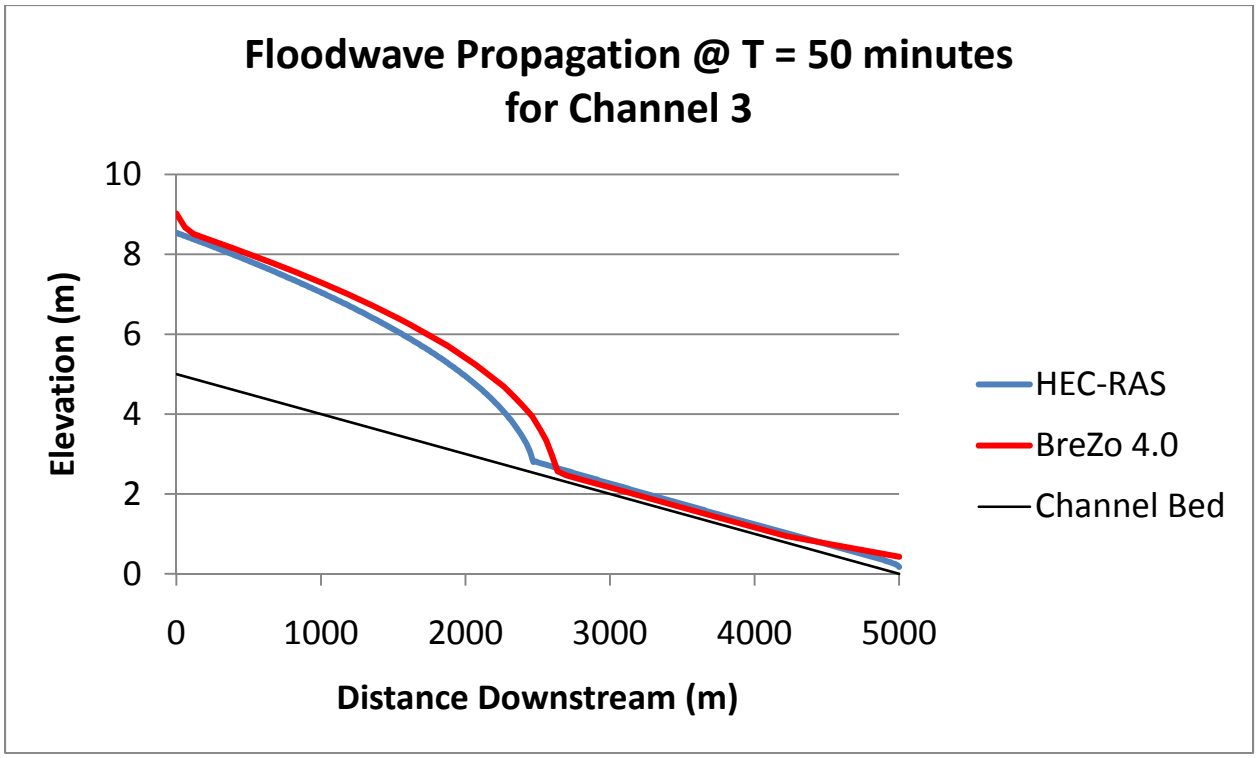

Figure 2.a. – Downstream of confluence

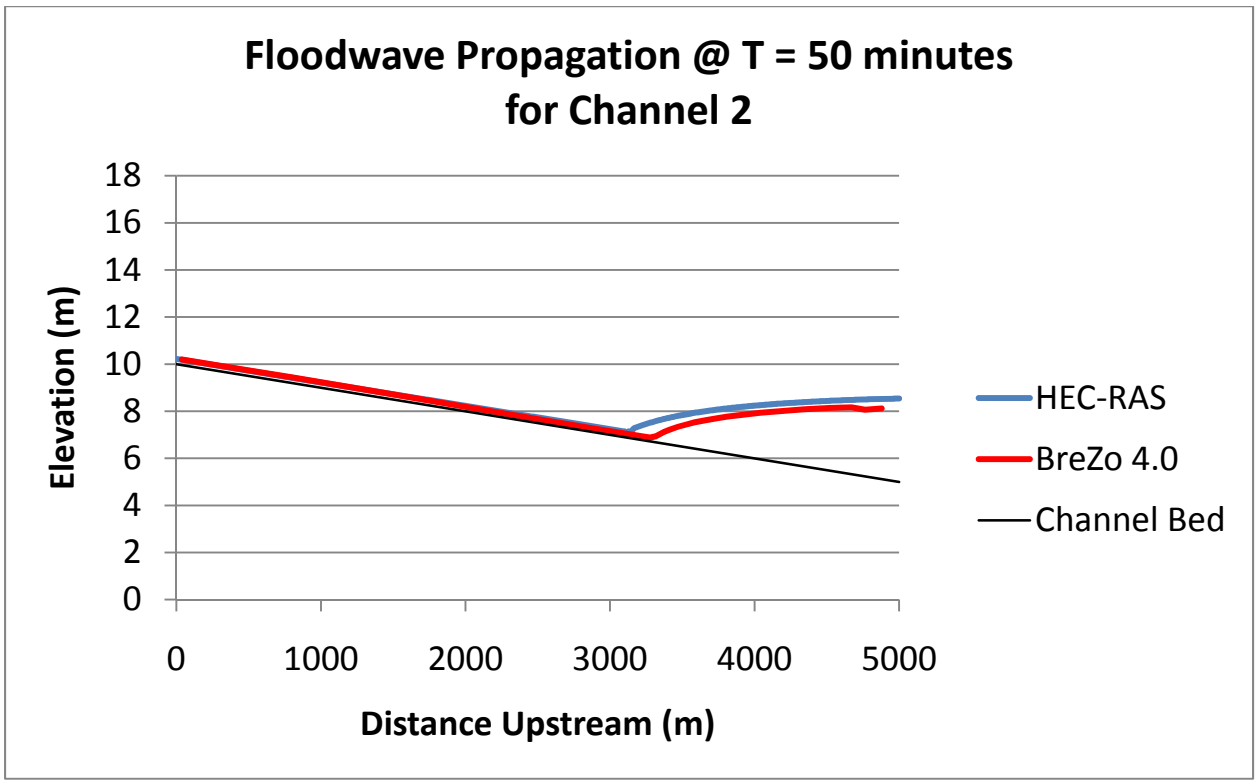

Figure 2.b. – Upstream of confluence

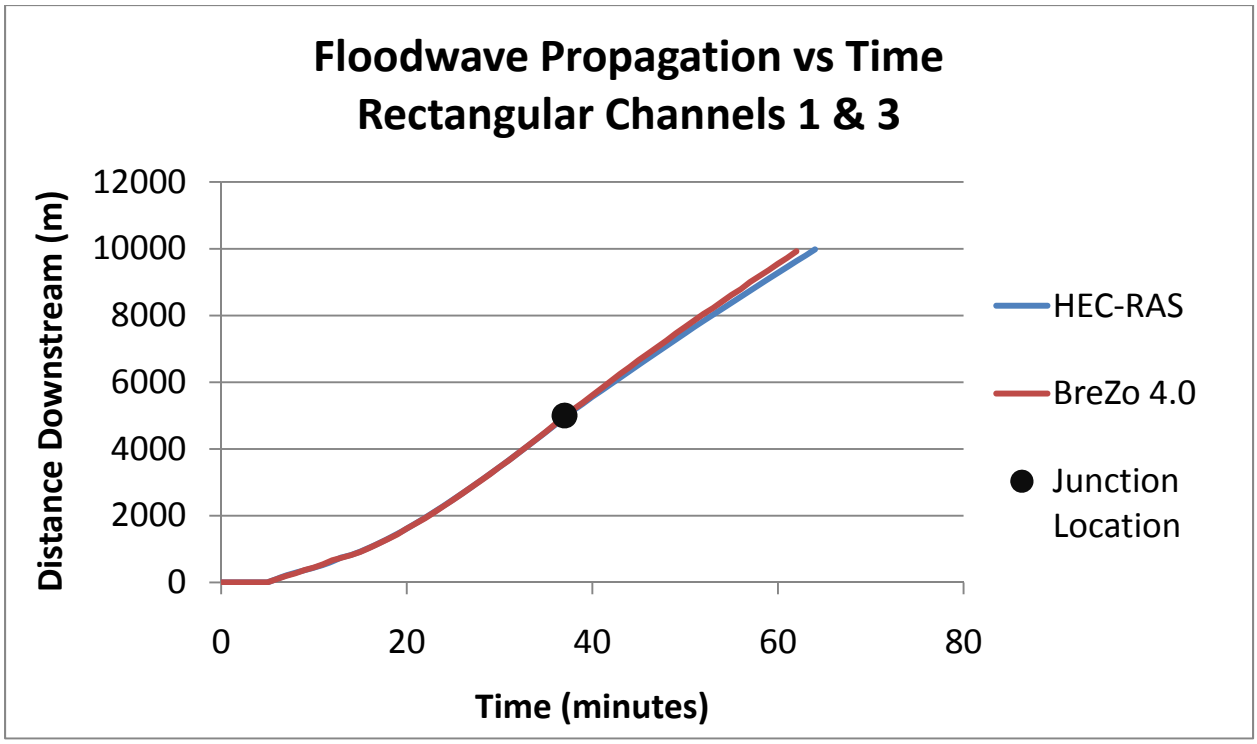

Figure 2.c. – Lateral inflow channel and main channel downstream of confluence.

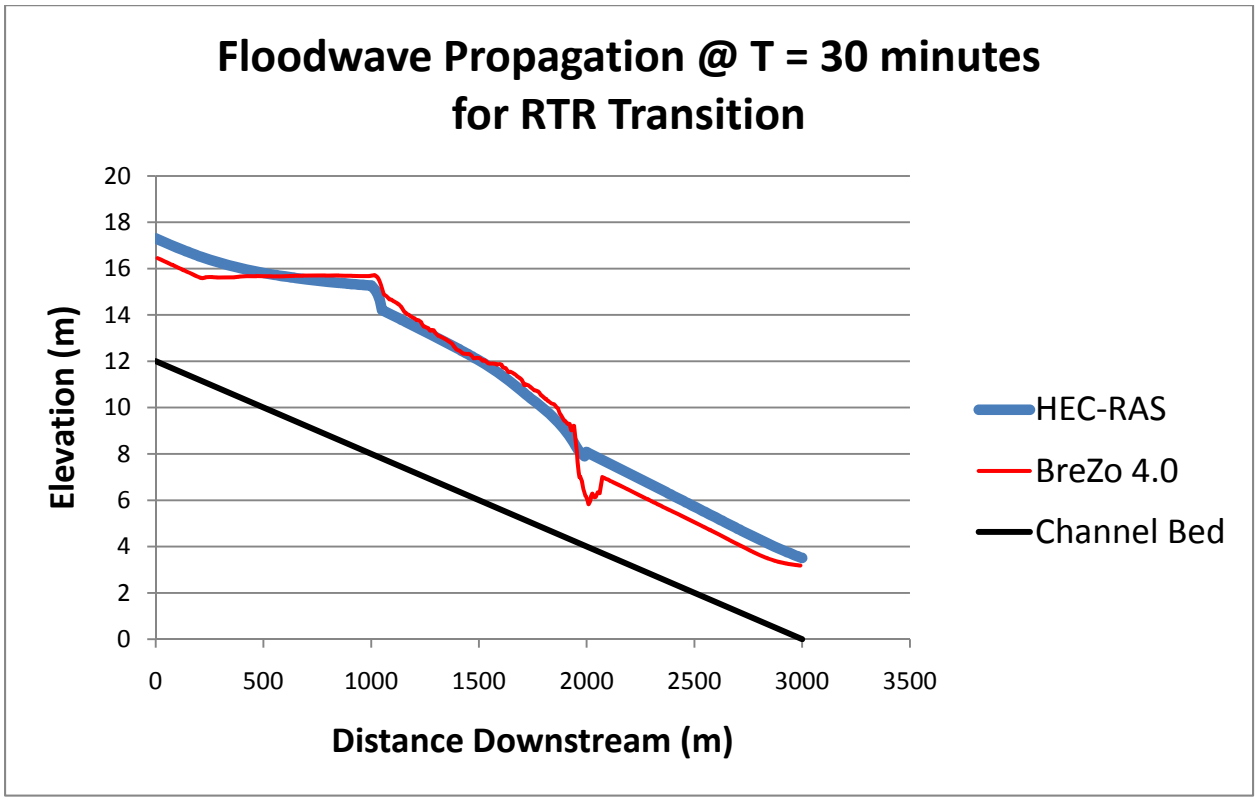

Figure 3.a. – Transition between rectangular and trapezoidal shaped channels simulation.

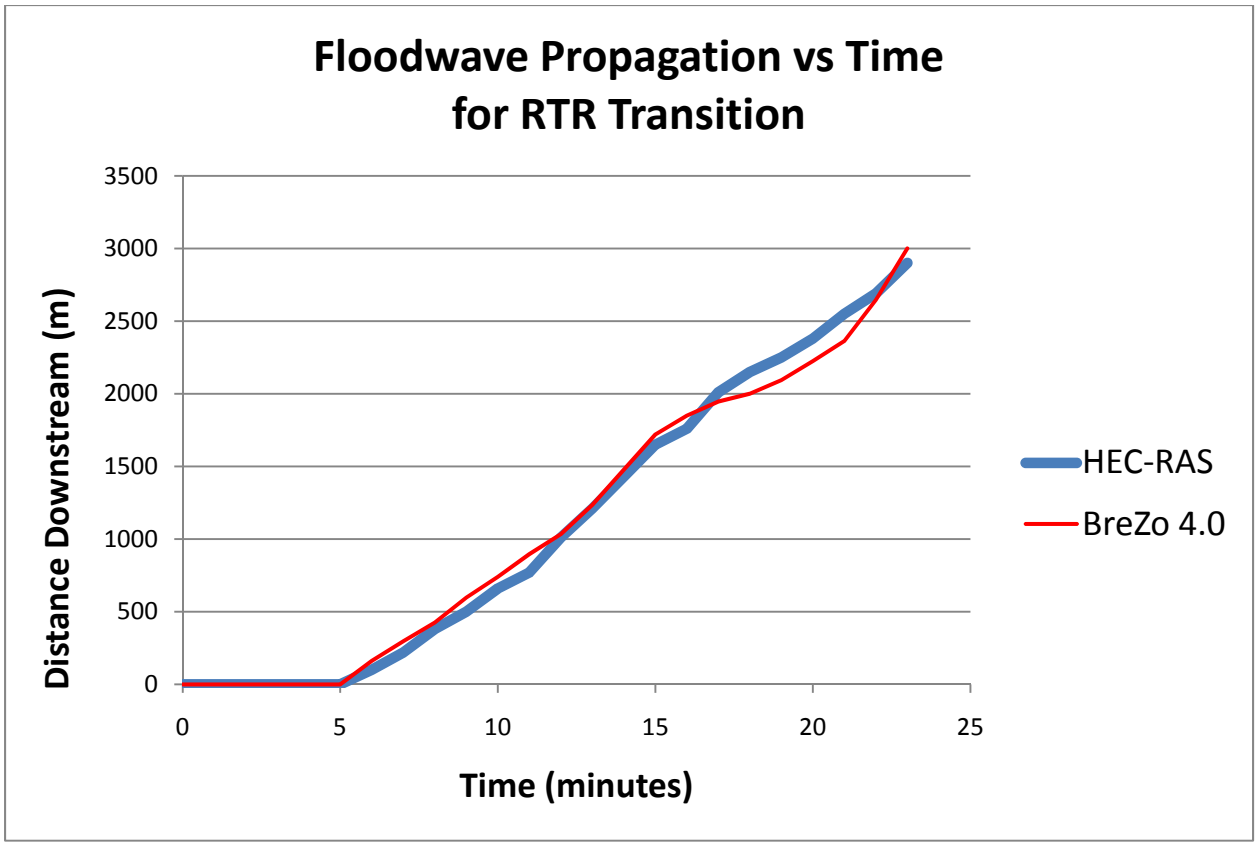

Figure 3.b. - Transition between rectangular and trapezoidal shaped channels simulation.

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U.S. Army Corps of Engineers. (n.d.). *HEC-RAS*. Retrieved 2011, from US Army Corps of Engineers Hydrologic Engineering Center Web Site: http://www.hec.usace.army.mil/software/hec-ras/

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 site. The institute also participates in and supports numerous other minor technology and information transfer activities throughout the year.

Kentucky Information Transfer Activities

Basic Information

Title:	Kentucky Information Transfer Activities
Project Number:	2010KY154B
Start Date:	3/1/2010
End Date:	2/28/2011
Funding Source:	104B
Congressional District:	KY 6th
Research Category:	Not Applicable
Focus Category:	None, None, None
Descriptors:	symposium, web site
Principal Investigators:	Lindell Ormsbee, Anna Goodman Hoover, Jim Kipp

Publication

1. Proceedings 2011 Kentucky Water Resources Annual Symposium, Kentucky Water Resources Research Institute, Lexington, Kentucky, 112 p.

Kentucky Information Transfer Activities (2010KY154B)

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

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 KWRRI 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 KWRRI 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.

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 "Beyond the Black Box: Integrating Advanced Characterization of Microbial Processes with Subsurface Reactive Transport Models" by Dr. Tim Schiebe on August 31, 2010.

An open house was held on Wednesday evening 10/13/2010 during 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 continues participation in the Bluegrass Partnership for a Green Community cooperative effort between the University of Kentucky, the Fayette County public school system, and the Lexington-Fayette Urban County Government, and numerous other partners including a number of citizen watershed groups in the community. Staff members are active with the water/stormwater team.

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 Institute assisted the University of Kentucky Superfund Research Program in planning and presenting "Nutrition and Superfund Chemical Toxicity: Implications in Risk Assessment" on December 2, 2010. The University of Kentucky Superfund Research Program Research Translation Core (directed by KWRRI) also provided seminars including "Emerging Issues with Vapor Intrusion" presented by Kelly Pennell of Brown University on March 25, 2010 and "Engaging Communities to Advance the Science of Environmental Justice" presented by Nigel Fields on April 20, 2010.

The Kentucky Water Resources Annual Symposium was held on March 21, 2011. Although the date of the symposium fell outside of FY2010, most of the planning and preparation occurred during the fiscal year. Two concurrent sessions provided time slots for 32 oral platform presentations. Twenty-four posters were also presented during a separate poster session. The 12 student research enhancement projects funded during FY2010 presented their results. 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, 2011, Kentucky Water Resources Research Institute, Lexington, Kentucky, 112 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. The Institute's newsletter WATERWORKS is also available in electronic format through a link on the web page.

USGS Summer Intern Program

None.

Student Support					
Category	**Section 104 Base Grant**	**Section 104 NCGP Award**	**NIWR-USGS Internship**	**Supplemental Awards**	**Total**
Undergraduate	4	0	0	0	4
Masters	5	0	0	0	5
Ph.D.	4	0	0	0	4
Post-Doc.	0	0	0	0	0
Total	13	0	0	0	13

Notable Awards and Achievements

The Kentucky Water Resources Research Institute at the University of Kentucky was designated as a Center of Excellence for Watershed Management by Region 4 of the US Environmental Protection Agency. With this designation, KWRRI becomes the first such Center of Excellence in Kentucky intended to help communities identify watershed-based problems and assist with the development and implementation of locally sustainable solutions. In becoming recognized, KWRRI demonstrated: 1) technical expertise in identifying and addressing watershed needs, 2) involvement of students, staff, and faculty in watershed research, 3) the capability to involve the full suite of disciplines needed for all aspects of watershed management, and 4) a willingness to partner with other institutions. Started in 2007, the EPA Region 4 Centers of Excellence for Watershed Management Program works with colleges and universities from across the Southeast to provide hands-on, practical products and services for communities to identify watershed problems and solve them.

2010KY138B - The experience and skills gained through this KWRRI-sponsored research made the student (Ester Renee Kirtman) competitive for a national scholarship awarded by the Garden Club of America for additional research to be conducted during summer 2011. The KWRRI supported research experience and the Garden Club of America award will make Kirtman a competitive candidate when applying for graduate school.

2010KY141B - The student (Stephanie Hayes) received the following awards as a result of her involvement in the research funded by this project: 1) 2010 First Place Undergraduate Research Competition, Ecology and Environmental Science Division, Kentucky Academy of Sciences, 2) 2010 First Place Undergraduate Research Competition, Ohio River Consortium for Research and Education, 3) 2011 Second Place Undergraduate Research Competition, Frank G. Brooks Award, Tri-Beta Regional Meetings, Huntsville, Alabama, 4) 2011 John Thieret Undergraduate Research Award, Department of Biological Sciences, Northern Kentucky University, and 5) 2011 Northern Kentucky University Faculty Senate Award presented to a graduating senior exhibiting exceptional skill and productivity in research and scholarly work.

2010KY148B - United States Patent Application: "Thiol-Containing Compounds for the Removal of Elements from Contaminated Milieu and Methods of Use." Inventors: Boyd E. Haley and David A. Atwood, U.S. Patent Appl. Serial No. 12/892,464, September 28, 2010. International Patent Application (same title and inventors), PCT/US 10/50512, September 28, 2010. This invention relates to the synthesis and use of new compounds to covalently capture contaminant elements in water (such as aresenic and mercury). This includes placing the contaminant capture agent on a solid support. The 2010 104B project demonstrated that a silica-supported reagent could remove arsenic (III) from water.