Maryland Water Resources Research Center
Annual Technical Report
FY 2014

Introduction

During Funding Year 2014, the Maryland Water Resources Research Center (MWRRC) supported a variety of research and outreach activities that address the diversity of water issues in the State and the Chesapeake Bay Region. The Center's outreach/information transfer event focused on the Anacostia River and Watershed, which has been transformed by cultivation and urbanization, with severe degradation of water quality, but which is undergoing widespread restoration efforts. Two research projects were continued from the previous funding year (under new project numbers, as an administrative arrangement to deal with the Federal Sequestration-related budget reduction in 2013); these projects deal with two emerging problems related to human effects on natural waters: novel methods to diagnose intersex conditions in freshwater fish, and tracing the pathways of road salt in surface and groundwater. A new research project focuses on the ecology of wetlands, with implications for restoration of natural wetlands throughout the region. Three graduate students received summer fellowships: investigating urban streams as sources of greenhouse gases, analyzing the water quality impacts of the use of steel slag in highway structural fills, and investigating the chemical kinetics of toxic iodine-containing organic compounds formed during drinking water treatment.

Research Program Introduction

With 104B funding, the Maryland Water Resources Research Center supported three research projects and three graduate student summer fellowships in Funding Year 2014. Two of the research projects, although given new project numbers, were essentially continuations of projects funded in FY2013. Originally selected for two years of funding in 2013, each of these projects had to be cut to one year when the 104B funding was reduced to 60%. Because the PIs were making appropriate progress, each was awarded a new 104B project to cover the second year of their research.

- Tracing the rates of road salt runoff movement from impervious surface to stream and the effects on soil and aquifer geochemistry (2014MD313B, *continuation of 2013MD306B*); PIs: Joel Moore and Steven Lev (Physics, Astronomy & Geosciences, Towson University).

- Wetland restoration: Experimental effects of soil carbon:nitrogen ratio on growth of invasive and native Phragmites australis (common reed) (2014MD314B); PI: Andrew Baldwin (Environmental Science & Technology, University of Maryland College Park).

- Validation of non-lethal laparoscopic technique for detection of intersex in regional black bass populations (2014MD315B, *continuation of 2013MD305B*); PI: Lance Yonkos (Environmental Science & Technology, University of Maryland College Park).

- Steel Slag-Induced Surface Water and Groundwater Contamination (Graduate Fellowship) (2014MD316B); Asli Yalcin Dayioglu (Ph.D. student, Civil & Environmental Engineering, University of Maryland, College Park; Advisor: A. Aydilek)

- Impacts of Stormwater Management on Greenhouse Gas Fluxes from Urban Streams (2014MD317B); Rose Smith (Ph.D. student, Geology, University of Maryland, College Park; Advisor: S. Kaushal)

- Kinetics of the Iodination of Natural Organic Matter and Iodinated Disinfection Byproduct Formation in Drinking Water Treatment (Graduate Fellowship) (2014MD318B); Michael Rose (Ph.D. student, Geography and Environmental Engineering, Johns Hopkins University; Advisor: A.L. Roberts)

A University of Maryland project selected for support under the IWR/NIWR program in 2012 was completed during this performance period:

- The Effectiveness of a Computer-Assisted Decision Support System Using Realistic Interactive Visualization as a Learning Tool in Flood Risk Management (2012MD299S), Bahram Momen (Environmental Science & Technology, University of Maryland, College Park).

A University of Maryland project selected for support under the 104(G) program in 2014 was active during this performance period:

- Environmental Concentrations and Exposure Effects of Environmental Gestagens on a Sentinel Teleost Fish (2014MD321G), Edward Orlando (Animal & Avian Sciences, University of Maryland, College Park).

The Effectiveness of a Computer-Assisted Decision Support System Using Realistic Interactive Visualization as a Learning Tool in Flood Risk Management

Basic Information

Title:	The Effectiveness of a Computer-Assisted Decision Support System Using Realistic Interactive Visualization as a Learning Tool in Flood Risk Management
Project Number:	2012MD299S
USGS Grant Number:	G12AP20058
Sponsoring Agency:	USACE IWR
Start Date:	3/1/2013
End Date:	11/30/2014
Funding Source:	Supplemental
Congressional District:	MD 5
Research Category:	Social Sciences
Focus Category:	Management and Planning, Models, Floods
Descriptors:	Decision Support System
Principal Investigators:	Bahram Momen, Kaye Lorraine Brubaker

Publications

1. Olsen, V. Beth Kuser, Cynthia McCoy, and Bahram Momen. “HAZUS Flood Risk Analysis: Regional vs Local Data," Poster Presentation, University of Maryland Bioscience Day, College Park: PS 35 Environmental Science: November 27, 2012.
2. Olsen, V. Beth Kuser, Cynthia McCoy, and Bahram Momen. “HAZUS Flood Risk Analysis: Regional vs Local Data,” Poster Presentation, University of Maryland Marine, Estuarine, and Environmental Science Graduate Program 2012 Colloquium, Institute of Marine and Environmental Technology: PS 26: October 19, 2012.
3. Olsen, V. Beth Kuser, 2014, "Proposing A Realistic Interactive Visualization Model And Testing Its Effectiveness In Communicating Flood Risk," Ph.D. Dissertation, Marine-Estuarine-Environmental Sciences (MEES), University of Maryland, College Park, MD, 308 pp.
4. Abdissa, Aman, Geoffrey Chan, Yevheniya Kupchyk, Derek Lam, and Marianne Varkiani. “Comparison of HAZUS vs. Google EarthTM in Flood Damage Analysis," Poster Presentation, University of Maryland "Maryland Day," College Park, Maryland: Apr. 27, 2013.
5. Olsen, V. Beth Kuser, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link, and Bahram Momen. “Sustainable pathways: Using innovative modeling techniques to communicate flood information to communities at risk: Are we reaching all demographics?” Poster Presentation (PS 82-117), Ecological Society of America Annual Conference, Minneapolis, Minnesota, August 9, 2013.
6. Olsen, V. Beth Kuser, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link, and Bahram Momen. “Sustainable pathways: Using innovative modeling techniques to communicate flood

information to communities at risk: Are we reaching all demographics?" Poster Presentation (PS 19), University of Maryland Marine, Estuarine, and Environmental Science Graduate Program 2013 Colloquium, Horn Point Environmental Science Center, September 27, 2013.

7. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "Using innovative modeling to prepare for flooding due to climate change in Maryland/DC Important Bird Areas," Poster Presentation, Maryland Ornithological Society (MOS) Annual Conference, Solomons, Maryland, May 17, 2014.
8. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "A user-friendly, inexpensive model interface communicates flood risk-reduction options effectively using realistic interactive visualization," Poster Presentation (PS 70-128), Ecological Society of America (ESA), August 14, 2014.

Completion Report

U.S. Army Corps Engineers Institute for Water Resources (IWR) and the National Institute for Water Resources (NIWR) Grant

U.S. Geological Survey Award No. G12AP20058

"The Effectiveness of a Computer-Assisted Decision Support System using Realistic Interactive Visualization as a Learning Tool in Flood Risk Management"

March 1, 2012 – November 30, 2014

Prinicpal Investigator: Bahram Momen, Ph.D.
Co-Principal Investigators: Kaye L. Brubaker, Ph.D.
Stacy M. Langsdale, P.E., Ph.D.
Report prepared by: V. Beth Kuser Olsen, Ph.D.

I. Summary

This project advances the field of flood risk management by using technology to bridge the gap between science and local community decision-making. Currently, flood risk management meetings use a computer-assisted decision support system (DSS) to illustrate various flood scenarios and facilitate collaborative discussions among participants. The DSS is a set of sophisticated models structured by geographic information systems (GIS) technicians.

This study proposed a "stakeholder-built" DSS. Stakeholders are defined here as those directly at risk of flooding. This method utilized improved user interface capabilities while retaining the technical rigor and robustness of a nationally-recognized GIS software package. There are times when a simple model may serve as an introduction to GIS technology. There are also situations where the cost of the sophisticated models may place them out of reach. The stakeholder-built DSS was proposed as a compliment to the sophisticated models by providing greater access to a DSS for end-users.

The stakeholder-built DSS, in which stakeholders construct their own models, uses realistic interactive visualization as a learning tool. Realistic visualization represents information using virtual reality. The intent is to trigger awareness of risk through emotional response to images. Stakeholders use interactive visualization when constructing the model. Awareness of the flood scenario is enhanced by the constant attention required of the model-builder as they make connections between hand-eye coordinated motions and the cognitive information they are modeling. Knowledge accumulates as multiple steps are completed.

The effectiveness of the stakeholder-built DSS was tested during community flood risk management meetings in Federal Emergency Management Agency Region III, the mid-Atlantic area. A DSS based on a nationally-recognized GIS software package was also tested to serve as a comparison. Data were collected in pre- and post-surveys and follow-up interviews.

The stakeholder-built and national GIS software DSS both performed equally well in communicating knowledge of flood risk and risk-reduction options, resulting in significant learning outcomes. To maximize the intent by stakeholders to take actions to reduce risk,

meetings using the stakeholder-built DSS in high-quality meeting facilities performed best. In addition, the stakeholder-built model was less expensive and found to be more user-friendly for stakeholders.

Research Objectives, Methods, Findings and Significance

These are reported in detail in V. Beth Kuser Olsen's University of Maryland dissertation: "Proposing a realistic interactive visualization model and testing its effectiveness in communicating flood risk." The anticipated publication date is August 22, 2015. A pre-publication copy is attached.

II. Presentations

A. Oral presentations:

1. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "A User-friendly, Inexpensive Model Interface Communicates Flood Risk Information Effectively and Motivates Action to Reduce Risk Using Realistic Interactive Visualization" XXth International Conference of the Society for Human Ecology: Oral Session: October 22-26, 2014.

2. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "Can a model make a difference in how well we communicate flood risk to the vulnerable? Chesapeake Modeling Symposium, Annapolis, Maryland: Oral Session: Making the Connections: Modeling Living Resources, Economic, and Social aspects of Estuary Protection and Restoration: May 29, 2014.

3. Olsen, V. Beth Kuser. "Proposing a realistic interactive visualization model and testing its effectiveness in communicating flood risk" Marine, Estuarine, and Environmental Science Graduate Program Dissertation Defense, University of Maryland, College Park: April 18, 2014.

4. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "Are we reaching the vulnerable when we communicate flood risk?" ESA Mid-Atlantic Chapter Conference, University of Maryland, College Park: Oral Session I:5: March 29, 2014.

5. Abdissa, Aman, Geoffrey Chan, Yevheniya Kupchyk, Derek Lam, and Marianne Varkiani (2013) "Flood Risk Management at University of Maryland: A Comparison of HAZUS vs. Google™ in Flood Damage Analysis." University of Maryland, College Park, Maryland.

6. McCoy, Cynthia, Beth Olsen, Bob Pierson, and Farhad Tavassoli (2012). "Damage Estimate Analysis for Alexandria, Virginia." Federal Emergency Management Agency Emergency Management Institute, Emmitsburg, Maryland.

B. Scientific poster presentations:

1. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "A user-friendly, inexpensive model interface communicates flood risk-reduction options effectively using realistic interactive visualization" Ecological Society of America (ESA): PS 70-128: August 14, 2014.

2. Olsen, V. Beth Kuser, Bahram Momen, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link. "Using innovative modeling to prepare for flooding due to climate change in Maryland/DC Important Bird Areas" Maryland Ornithological Society (MOS) Annual Conference, Solomons, Maryland: PS: May 17, 2014.

3. Olsen, V. Beth Kuser, Kaye L. Brubaker, Gerald E. Galloway Jr., Lewis (Ed) Link, and Bahram Momen. "Sustainable pathways: Using innovative modeling techniques to communicate flood information to communities at risk: Are we reaching all demographics?" Ecological Society of America Annual Conference, Minneapolis, Minnesota: PS 82-117: August 9, 2013 and University of Maryland Marine, Estuarine, and Environmental Science Graduate Program 2013 Colloquium, Horn Point Environmental Science Center: PS 19: September 27, 2013.

4. Abdissa, Aman, Geoffrey Chan, Yevheniya Kupchyk, Derek Lam, and Marianne Varkiani. "Comparison of HAZUS vs. Google Earth™ in Flood Damage Analysis: University of Maryland" Maryland Day, College Park, Maryland: April 27, 2013.

5. Olsen, V. Beth Kuser, Cynthia McCoy, and Bahram Momen. "HAZUS Flood Risk Analysis: Regional vs Local Data" University of Maryland Marine, Estuarine, and Environmental Science Graduate Program 2012 Colloquium, Institute of Marine and Environmental Technology: PS 26: October 19, 2012 and University of Maryland Bioscience Day, College Park: PS 35 Environmental Science: November 27, 2012.

III. **Students supported**

Total number of students supported:

Undergraduates: 7
Masters: 2
Ph.D.: 2

This grant provided a fellowship to Ph.D. candidate, V. Beth Kuser Olsen to assist with costs of research toward completion of her Ph.D. dissertation. She successfully defended her dissertation, "Proposing a realistic interactive visualization model and testing its effectiveness in communicating flood risk" in April 2014. It was accepted for publication in August 2014. Under this grant, her accomplishments include:

1. Completion of the Collaborative Institutional Training Initiative (CITI Program) Human Subjects Research 11-module course: Social & Behavioral Research - Basic Curriculum
2. Completion of approved University of Maryland, College Park, Human Subjects Internal Review Board application and renewals

3. Development of a research website: https://sites.google.com/site/floodmodeling1/
4. GIS training in "HAZUS for Floods" at the Federal Emergency Management Agency Emergency (FEMA) Emergency Management Institute
5. Service as the search committee for hiring six research assistants
6. Training of six research assistants and four undergraduate students as GIS Technicians in Environmental Science Research Institute (ESRI) ArcGIS 10™, FEMA HAZUS-MH for Floods, and the Stakeholder-built model used in this study that links to Google Earth™ maps and the FEMA National Flood Hazard Layer Google Earth™ application and uses Google Earth™ drawing tools to build the model.
7. Training of research assistants as Meeting Facilitators and Meeting Assistants in collaborative learning techniques and participant-centered learning.
8. In coordination with FEMA Region III Community Coordination and Outreach (CCO) meetings, introduction of the project to city and county municipal planners in twelve FEMA CCO and Open House meetings
9. Introduction of the project in 18 local community outreach meetings
10. Completion of ten data-collection meetings with a total of 98 participants, completed exploratory interviews with key leaders in the communities prior to each meeting, and completed follow-up interviews with selected participants
11. Presented four scientific posters, five oral presentations, and completed one publication
12. Reviewed a journal article for the *Journal of the American Water Resources Association*
13. Joined www.linkedin.com Group, Collaborative Modeling for Decision Support as an invited member
14. Joined ResearchGate Workgroup at http://www.researchgate.net/group/Sigma_Xi_The_Scientific_Research_Society/ as an invited member

This grant provided salaries for four research assistants during GIS technician and meeting facilitator training:

Undergraduate: Alison Karp

Masters:
1. Michael Riedman
2. Xiaoyu Bi Murtha

Ph.D.: Quentin A. Stubbs

Each completed Environmental Science Research Institute (ESRI)™ certifications for successful completion of three ESRI ArcGIS 10™ online courses and eleven ESRI™ Federal Emergency Management Agency Multi-hazard Loss Estimation Methodology (HAZUS) online courses, followed by training in the use of real data. The ESRI courses completed are listed in the addendum of V. Beth Kuser Olsen's dissertation, attached.

This grant provided salaries for six research assistants during Meeting Assistant training:

Undergraduates:

1. Alison Karp
2. Maureen Kelly
3. Aman Abdissa

Masters:

1. Michael Riedman
2. Xiaoyu Bi Murtha

Ph.D.: Quentin A. Stubbs

This grant covered travel, lodging and meals to present project introductions to potential data-collection communities for:

Undergraduate: Alison Karp

Masters: Michael Riedman

Ph.D.: V. Beth Kuser Olsen

This grant covered travel, lodging and meals for pre-data-collection preparation and flood risk management meetings for ten communities for:

Undergraduates:

1. Alison Karp
2. Maureen Kelly
3. Aman Abdissa

Masters:

1. Michael Riedman
2. Xiaoyu Bi Murtha

Ph.D.:

1. V. Beth Kuser Olsen
2. Quentin A. Stubbs

This grant covered GIS training for:

Undergraduates:

1. Aman Abdissa
2. Geoffrey Chan
3. Derek Lam
4. Marianne Varkiani
5. Yevheniya Kupchyk
6. Maureen Kelly

III. Publication

Olsen, V. Beth Kuser. In-print: 2015. “Proposing a realistic interactive visualization model and testing its effectiveness in communicating flood risk” Electronic dissertation: ProQuest. University of Maryland, College Park. Anticipated publication date: August 22, 2015.

Validation of non-lethal laparoscopic technique for detection of intersex in regional black bass populations

Basic Information

Title:	Validation of non-lethal laparoscopic technique for detection of intersex in regional black bass populations
Project Number:	2013MD305B
Start Date:	7/1/2013
End Date:	6/30/2015
Funding Source:	104B
Congressional District:	MD-005
Research Category:	Water Quality
Focus Category:	Surface Water, Toxic Substances, Methods
Descriptors:	None
Principal Investigators:	Lance Yonkos, Lance Yonkos

Publications

There are no publications.

Administrative Completion Report: 2013MD305B
PI: Dr. Lance Yonkos

In FY 2013, the Federal portion of the MWRRC budget was reduced to 60% several months after research projects and summer graduate fellowship recipients were selected. To meet the imposed budget constraints, 2013MD305B, originally selected as a 2-year project, was funded for only one year. In FY 2014, the PI was awarded another 1-year project, 2014MD315B. Although it has a different 104B number, this project is essentially a continuation of 2013MD305B.

Administratively, 2013MD305B is completed as of this Annual Report, although the research is continuing under 2014MD315B. For the technical progress report, please refer to 2014MD315B in this Annual Report.

Tracing the rates of road salt runoff movement from impervious surface to stream and the effects on soil and aquifer geochemistry

Basic Information

Title:	Tracing the rates of road salt runoff movement from impervious surface to stream and the effects on soil and aquifer geochemistry
Project Number:	2013MD306B
Start Date:	6/1/2013
End Date:	2/29/2016
Funding Source:	104B
Congressional District:	MD 3
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Solute Transport, Water Quality
Descriptors:	None
Principal Investigators:	Joel Moore, Steven Lev

Publication

1. Moore J., S. M. Lev, R. E. Casey, 2013, Modeling the effects of road salt on soil, aquifer, and stream chemistry in 2013 MODFLOW and More Conference Proceedings, Integrated GroundWater Modeling Center, Golden, CO. 5 pp.

Administrative Completion Report: 2013MD306B
PI: Dr. Joel Moore

In FY 2013, the Federal portion of the MWRRC budget was reduced to 60% several months after research projects and summer graduate fellowship recipients were selected. To meet the imposed budget constraints, 2013MD306B, originally selected as a 2-year project, was funded for only one year. In FY 2014, the PI was awarded another 1-year project, 2014MD313B. Although it has a different 104B number, this project is essentially a continuation of 2013MD306B.

Administratively, 2013MD306B is completed as of this Annual Report, although the research is continuing under 2014MD313B. For the technical progress report, please refer to 2014MD313B in this Annual Report.

Tracing the rates of road salt runoff movement from impervious surface to stream and the effects on soil and aquifer geochemistry

Basic Information

Title:	Tracing the rates of road salt runoff movement from impervious surface to stream and the effects on soil and aquifer geochemistry
Project Number:	2014MD313B
Start Date:	6/1/2014
End Date:	2/29/2016
Funding Source:	104B
Congressional District:	2
Research Category:	Water Quality
Focus Category:	Hydrogeochemistry, Geochemical Processes, Water Quality
Descriptors:	None
Principal Investigators:	Joel Moore, Joel Moore

Publications

There are no publications.

Progress Report

Tracing the rates of road salt runoff movement from impervious surfaces to stream and the effects on soil and aquifer geochemistry

2013-MD-306B & 2014-MD-313B

March 1, 2013 – February 28, 2015

1) Narrative summary – Progress Report

Problem and Research Objectives

Runoff from impervious surface that contains dissolved road salt (NaCl) clearly affects the chemistry of soils, groundwater, and streams. However, effects on soils, groundwater, and streams typically have been studied in isolation from each other. Also, the effects of stormwater management basins on road salt movement from impervious surfaces to streams have not been investigated. The research funded by this grant has the goal of addressing the following questions about road salt movement from impervious surfaces to streams in a system with stormwater management basins. The study site is located in Owings Mills, MD located in a suburban part of Baltimore County, MD.

a) What are the rates of Na and Cl movement from impervious surface to stream?
b) Where and how along the flow paths from road to stream are Na and Cl retained?
c) What is the effect of Na retention on soil and shallow aquifer chemistry, particularly on pH and the concentrations of important plant macronutrients like calcium (Ca), magnesium (Mg), and potassium (K) in the cation exchange complex?

Methods

The methods employed include collection of

- Physical data such as stream discharge and water table levels
- Collection of water and soil/aquifer matrix samples at the field site
- Chemical analysis of those samples in the Urban Environmental Biogeochemistry Laboratory at Towson University including
  - Concentration of cations and anions in water via ion chromatography
  - Composition of the cation exchange complex via $BaCl_2$ extraction and then ion chromatography
  - Soil pH via 0.1M $CaCl_2$ suspension and pH probe
- Deployment of pressure and conductivity sensors to continuously monitor water level and the total dissolved content

Principal Findings

Stream discharge and water table levels increased from fall into winter as trees shed their leaves and evapotranspiration rates decreased. Na^+ and Cl^- were loaded into groundwater via stormwater management basins (SMBs) and then moved via surface and groundwater flow into first- and second-order streams. In groundwater below the SMBs, Cl^- concentrations increased rapidly at the beginning of the 2013–14 winter and approached seawater concentrations (Fig. 1). Through early spring, Cl^- and Na^+ concentrations decreased slowly. A similar, though smaller amplitude, Cl^- and Na^+ concentration increased from fall into winter and then decline into early spring was seen in groundwater downgradient of the SMBs.

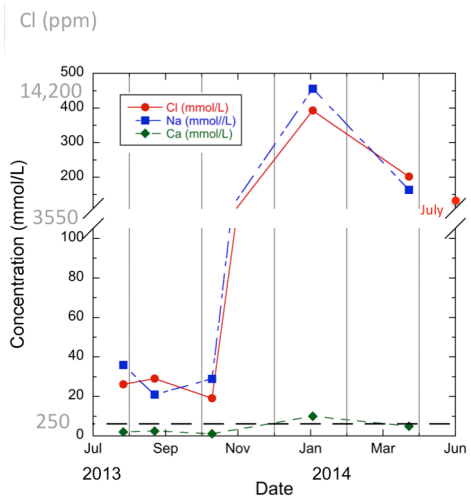


Fig. 1: Calcium, chloride, and sodium concentrations in groundwater from July 2013 to June 2014. The groundwater was collected from under a stormwater management basin that collects runoff from a ~200 unit townhome community along with secondary and tertiary roads. Sodium and chloride concentrations were ~75% the concentration of sea water and were ~2100 times higher than concentrations measurd in groundwater collected from a forested watershed ~15 km to the northeast.

The movement of high Na^+ water through groundwater has increased the pH and Na^+ concentrations in soils and aquifer matrix downgradient of the SMBs (Fig. 2). Compared to unaffected soils, soil pH increased from a value of ~4.5 to pH 6–7 and the Na^+ fraction of the cation exchange complex increased from 0 to 20–40%. Measured Na^+ fractions of the cation exchange complex in road salt–affected soils matched well with predictions from the PHREEQC geochemical model. Na^+ replaced H^+, Mg^{2+}, and, to a lesser extent, Ca^{2+} in the cation exchange complex.

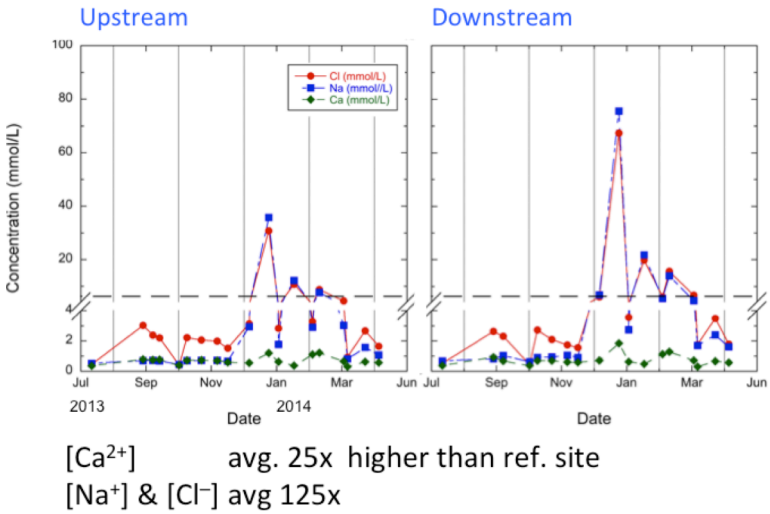


Fig. 2: Calcium, chloride, and sodium concentrations from July 2013 to June 2014 from locations upstream and downstream where water from two stormwater management basins enters the stream via surface and groundwater. Concentrations are compared to a reference watershed ~15 km to the northeast with similar bedrock and a similar size.

Significance

Preliminary results of this study increase understanding of how road salt affects the chemistry of soils/aquifer matrices, groundwater, and streams. Understanding groundwater and stream chemistry across seasons will provide information that will be useful in predictive models of road salt impacts in the future. Understanding of the effects of high Na^+ water on soil and aquifer matrix chemistry provides information that is important for assessment of the longer term impact of road salt.

2) Publication citations

Conference proceedings

Moore J., S. M. Lev, R. E. Casey, 2013, Modeling the effects of road salt on soil, aquifer, and stream chemistry in *2013 MODFLOW and More Conference Proceedings*, Integrated GroundWater Modeling Center, Golden, CO. 5 pp.

Abstracts

Moore J. (2014) Stormwater management basins and road salt loading to groundwater in Baltimore County. *Twenty-third Annual Maryland Ground Water Symposium*, Baltimore, MD.

Moore J. (2014) Stormwater management basins and road salt loading to groundwater and streams. *Twentieth Annual Conference of the Maryland Water Monitoring Council*, North Linthicum, MD.

Moore J., Sandosky B.**, McGuire M., Casey R. E., Snodgrass J. W., Lev S. M. (2014) Alteration of Critical Zone chemistry due to addition of rapidly dissolving rock salt. *Goldschmidt Conference*, Sacramento, CA.

Sandosky B.**, Lev S. M., Casey R. E., Snodgrass J. W., McGuire M., **Moore J.** (2014) Increased Chemical Fluxes to the Critical Zone due to road salt (halite) addition. *Geological Society of America, Northeastern Section Meeting*, Lancaster, PA.

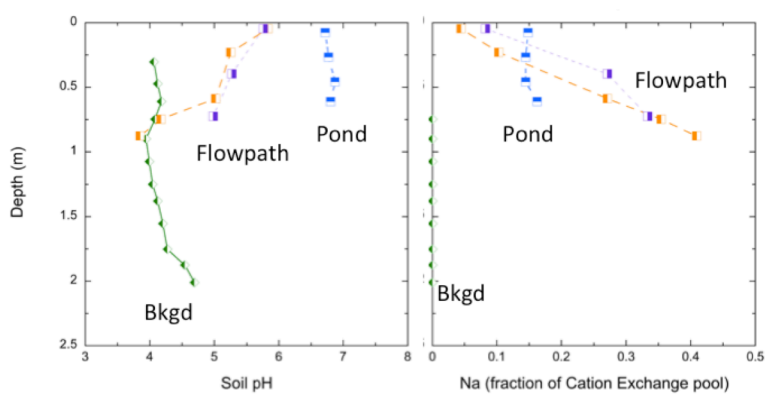


Fig. 3: Soil pH and Na as a fraction of total cation exchange pool versus depth in soils and aquifer materials from outside the road salt–affected area (background), within the flowpath from stormwater management basins (SMBs) to stream, and under the SMBs (pond). Soils and aquifer materials in road salt–affected areas exhibit a pH increase of 1–2.5 units and a Na increase from 0 to up to 0.4 of total cation exchange.

Moore J., Sandosky B.**, McGuire M., Casey R. E., Snodgrass J. W., Lev S. M. (2013) Salt overload: How quickly does road salt move from road to groundwater to stream in Baltimore County, MD and what are the effects on soil, groundwater, and stream chemistry? *Fall American Geophysical Union Meeting*, San Francisco, CA.

Moore J., Lev S. M., Casey R. E. (2013) Modeling the effects of road salt on soil, aquifer, and stream chemistry. *MODFLOW and More 2013: Translating Science into Practice*, Integrated GroundWater Modeling Center, Colorado School of Mines, Golden, CO.

3) Student Support

MWRRC and matching funds have supported stipends for 1 undergraduate student and 2 masters students. Research associated with this grant, including analytical costs, have supported 2 additional undergraduate students.

4) Notable achievements and awards

The sensor deployment originally proposed in this grant is being built upon through funds applied for, and received from, the School of Emerging Technology at Towson University. The $31,725 in funds was used to purchase additional conductivity and pressure sensors as well as Isco instruments for measuring pipe flow in culverts. Joel Moore is collaborating with Michael McGuire, a computer scientist specializing in spatio-temporal data mining algorithms, on this project. Additionally, a computer science graduate student is being supported by the Towson University funds in the summer of 2015 to apply spatio-temporal data mining techniques and develop ways for visualizing the sensor datasets collected as part of the work supported by this grant.

Joel Moore applied for, and was awarded, a three-year endowed chair position awarded to pre-tenure faculty (Jess and Mildred Fisher Endowed Chair of Geological Sciences) in part to expand on the work originally proposed in the MWRRC grant.

The work supported by this grant has been well received. Dr. Moore gave the following talks related to this research:

Research seminars

Sept. 2014: A high salt diet: How stormwater management ponds affect road salt impacts on soils, groundwater, and streams. *Department of Geology Seminar, Kent State University*.

Dec. 2014: Groundwater that looks like seawater: stormwater management basins & road salt loading into suburban watersheds. *Maryland-DC-Delaware Water Science Center, US Geological Survey*.

Mar. 2015: Groundwater that looks like seawater: stormwater management basins & road salt loading into suburban watersheds. *Department of Geography & Environmental Planning, Towson University*.

Talks to local and regional governmental groups

Mar. 2015: Groundwater that looks like seawater: stormwater management basins & road salt loading into suburban watersheds. Department of Environmental Protection & Sustainability, Baltimore County, MD.

Apr. 2015: Effects of road salt on dissolved sodium and chloride in suburban Baltimore groundwater and streams. Reservoir Technical Group, Baltimore Metropolitan Council.

Additionally, in part related to the work supported by this proposal, Dr. Moore was nominated and elected to serve a three-year term on the Maryland Water Monitoring Council (starting January 2015).

Wetland restoration: Experimental effects of soil carbon:nitrogen ratio on growth of invasive and native Phragmites australis (common reed)

Basic Information

Title:	Wetland restoration: Experimental effects of soil carbon:nitrogen ratio on growth of invasive and native Phragmites australis (common reed)
Project Number:	2014MD314B
Start Date:	3/1/2014
End Date:	2/29/2016
Funding Source:	104B
Congressional District:	5th and 8th Congressional District of Maryland
Research Category:	Biological Sciences
Focus Category:	Wetlands, Nutrients, Invasive Species
Descriptors:	None
Principal Investigators:	Andrew Baldwin, Stephanie Ann Yarwood

Publication

1. Baldwin, Andrew H., Amr E. Keshta, Martina Gonzalez Mateu, Diane E. Leason, and Stephanie A. Yarwood, 2015. Carbon cycling, invasives, and restoration: an overview of current research on the tidal Patuxent. Patuxent River Conference, Jefferson-Patterson Park and Museum, St. Leonard, Maryland (Poster presentation).

Progress Report

Wetland restoration: Experimental effects of soil carbon:nitrogen ratio on growth of invasive and native *Phragmites australis* (common reed)

Prepared for: Maryland Water Resources Research Center

Project Number: 2014MD314B

Project duration: March 1, 2014 - February 28, 2016

Principal Investigator:
Andrew H. Baldwin
Department of Environmental Science and Technology
University of Maryland, College Park, MD 20742
baldwin@umd.edu 301-405-7855

Co-PI: Stephanie A. Yarwood

Prepared by: A.H. Baldwin, S.A. Yarwood, and M. Gonzalez Mateu (graduate student)

Submitted: June 16, 2015

Project Goals, Methods, and Research Accomplishments

Objectives

Objective 1: Assess aboveground and belowground growth, ecophysiology, and competitive performance of native and invasive *Phragmites* in mixtures and monocultures, planted in soil from either restored or natural wetland and subjected to increased, decreased, or unaltered soil C:N ratio (manipulated by addition of wood shavings or urea nitrogen).

Objective 2. In the monoculture plantings used in Objective 1, quantify AM fungi colonization of roots of native and invasive *Phragmites* and relate AM fungi abundance to plant responses to C:N ratio treatments in soils from restored and natural wetlands.

Objective 3: In a separate experiment, determine N and C mineralization rates in soils from restored and natural wetlands subjected to increased, decreased, or unaltered soil C:N ratio.

Methods

We conducted Objective 3 first because it was a lab study that only required collecting soil from the field and could be accomplished in a short period of time. Soils were sampled in October of 2014 from two freshwater tidal wetlands. Soils were amended with urea, glucose, or sawdust, corresponding to a low C:N (urea), a high C:N with a labile carbon source (glucose) and a high C:N with a recalcitrant C source (sawdust). To determine Potential Nitrogen Mineralization (PMN), the amended soils were incubated under anaerobic conditions in glass serum bottles at 40C for 10 days, and soil extracts analyzed for NH4-N. To determine emissions of carbon dioxide and methane, amended soils were incubated at 35C for 25 days and gas samples analyzed on a gas chromatograph.

Objectives 1 and 2 were originally planned as greenhouse experiments, but we decided to conduct the study as a field experiment instead because: 1. We had unforeseen micronutrient limitation in the greenhouse that needed to be solved before greenhouse experiments could be conducted; 2. We were encouraged by results from Objective 3 that field treatments would be effective, and 3. Field studies, while harder to maintain environmental control, are more realistic than greenhouse studies. (The micronutrient problem has since been addressed, and found to be reduced iron limitation, in a separate experiment conducted by a group of undergraduate Capstone students in Environmental Science and Technology, ENST). Early in 2015 we collected additional rhizomes, propagated them in the greenhouse (Figure 1), and planted them in the field at Jug Bay Wetlands Sanctuary, in a tidal freshwater marsh where invasive *Phragmites* had been sprayed with herbicide during the fall of 2015. A boardwalk was constructed to reduce disturbance to the marsh substrate (Figures 2 and 3). Plants of native

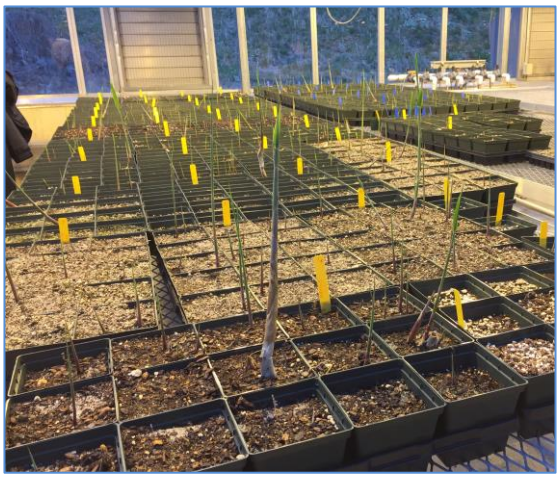


Figure 1. Native (yellow tags) and invasive (blue tags) Phragmites shoots emerging from rhizomes collected on the Patuxent river. Plants were grown to abou 1 m height and planted in the field C:N ratio experiment.

and invasive *Phragmites* were planted in monoculture and mixture, and treatments to alter soil C:N ratio have been added (Figure 4). The treatments are: urea (22g), wood shavings (1.25kg), both, or no treatment to 1-m^2 plots.

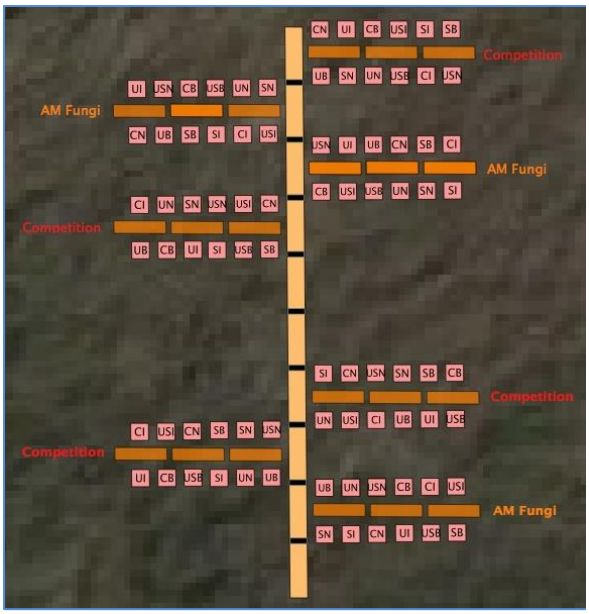


Figure 3. Jug Bay site showing the RCBD experimental design. We will facilitate the access to each block by building boardwalks, which will also help reduce the disturbances during sampling. Each treatment is assigned in each block and can be identified by the following letter: U=urea, S=sawdust, C=control, N=Native, I=Invasive, B=Both native and invasive.

Figure 2. Experimental site at Jug Bay Wetland Sanctuary (also pictured on the cover page).

Figure 4. Application of carbon treatment to experimental plot (pine wood shavings, 1.5 kg/m^2).

Research accomplishments

In the laboratory incubation study (Objective 3), Potential Nitrogen Mineralization was affected by the increased carbon amendments only in the natural wetland soil. Both the glucose and sawdust treatments effectively decreased nitrogen mineralization, compared to the control soil. Since there were no statistical differences between these two amendments, either one could be used for restoration purposes but sawdust represents a cheaper alternative. In the case of the urea treatment, its application significantly increased mineralization for both natural and restored sites compared to the control and the other amendments as expected (Figure 5).

The carbon addition treatments significantly increased carbon dioxide production in both types of wetland soils, while the urea treatment had no significant effect on CO2 production compared to controls (Figure 6). However, the dynamics of the gas production was different between glucose and sawdust (results not shown). Glucose addition caused a more rapid but short-lived response, where a lot of CO2 was produced after only 4 days of adding the treatment. This is the result of an increase in microbial respiration as the microbial community increases its biomass by incorporating the labile carbon source. After a few days, carbon mineralization slows down probably due to nitrogen limitations and as a result there is less CO2 production. A similar but more delayed and gradual response was observed for the sawdust treatments. These produced less amount of CO2 (Figure 6) but a longer lasting, because sawdust is a more recalcitrant carbon source and can't be incorporated to the microbial biomass as easy as glucose.

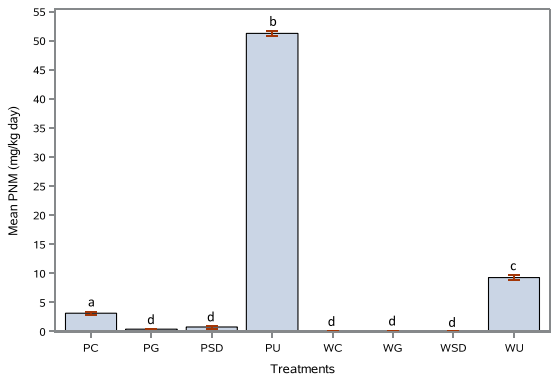


Figure 5.Potential nitrogen mineralization after 10 days of anaerobic incubation. Means and standard error bars are shown, and different letters indicate statistical significance between treatment means (P<0.001). P=Patuxent Wetland Park, W=Wooton's Landing,

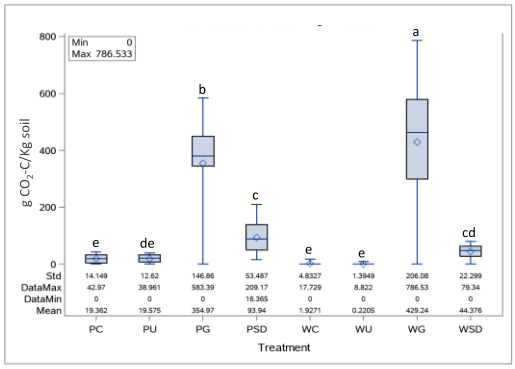


Figure 6. Boxplot showing total carbon dioxide production (gCO2-C/Kg of soil per day) after 25 days of anaerobic incubation. Different letters indicate statistical significance (P<0.001). P=Patuxent Wetland Park, W=Wooton's Landing, C=Control, G=Glucose, SD=Sawdust, U=Urea.

For the field C:N ratio competition experiment (Objective 1), the site was selected, boardwalks constructed, plots cleared of dead and standing vegetation, and experimental plots were planted. Pre-treatment plant measurements were taken (height, basal stem diameter), and treatments of urea and wood shavings applied. A monitoring well was installed at the site for continuous logging of water level, electrical conductivity, and temperature. Treatments and monitoring will continue throughout the growing season. We will take different measurements of the plants during two growing, which will be the duration of the experiment seasons (the experiment will continue beyond the end of the funded project period). Every two weeks we will measure stem height, light availability (PAR) and we will count the number of stems per plot. We will also analyze different soil characteristics such as pH, conductivity, texture, and C:N ratio. When the experiment concludes, we will harvest and estimate the above and belowground biomass, as well as leaf area and specific leaf area. Then we will use that information to explore the net balance of native and invasive *Phragmites* interaction, using an index of relative interaction intensity proposed.

To study AM fungi, we will collect rhizome samples from some plots in the middle of the growing season, and separate the below and aboveground biomass. We will wash and separate the roots from the soil, weigh them, and store them in 50% ethanol. For the quantification we will use microscopic and molecular biology techniques. The microscope observations will be done on roots dyed with Trypan blue and using the grid line intersect procedure. This technique will be complemented with terminal

restriction fragment length polymorphism analysis (TRFLP) to obtain further information about the mycorrhizae community.

As mentioned previously, we encountered micronutrient limitation of native *Phragmites* plants grown in the greenhouse. The primary symptom was leaf chlorosis (yellowing) and ultimately poor growth. To identify the limiting nutrients, the PIs worked with a group of students in the ENST undergraduate Capstone course to develop and carry our a two-semester greenhouse experiment. Based on corresepondece with colleagues, we identified two possible limiting minerals: magnesium (Mg2+) and reduced iron (Fe2+). These were applied to native *Phragmites* in a factorial combination of Mg and Fe (all of which received Osmocote slow-release fertilizer), plus an additional treatment that received no minerals or fertilizer. A number of measuresments were made, but the results are evident in the photograph in Figure 7: reduced iron was the primary cause of chlorosis (in fertilized plants) and strongly limiting growth.

Figure 7. Native *Phragmites* grown under different supplements of reduced iron, magnesium, and fertilizer. The experiment was carried out by undergraduate ENST Capstone students.

Publications and Presentations

Baldwin, Andrew H., Amr E. Keshta, Martina Gonzalez Mateu, Diane E. Leason, and Stephanie A. Yarwood, 2015. Carbon cycling, invasives, and restoration: an overview of current research on the tidal Patuxent. Poster presentation. Patuxent River Conference, Jefferson-Patterson Park and Museum, St. Leonard, Maryland.

Students supported

Undergraduate

The project partially supported a group of 7 undergraduate students in the two-semester ENST Capstone course series, who developed and carried the micronutrient limitation experiment on native *Phragmites*, under the mentorship of the PIs.

Additionally, about 5 undergraduate students working in the PIs' labs have participated in the project and discussed the project during presentations given by the graduate student during lab group meetings.

M.S.

The project provides the primary research funding for Martina Gonzalez Mateu, an international M.S. student in the MEES program co-advised by the PIs. At least 2 other M.S. students from the PIs' labs have also participated in the research or heard presentations on the project.

Ph.D.

No Ph.D. students were directly supported by the project, although at least 2 Ph.D students in the PIs' labs have participated in the research or contributed to the project during presentations at lab group meetings.

Validation of non-lethal laparoscopic technique for detection of intersex in regional black bass populations

Basic Information

Title:	Validation of non-lethal laparoscopic technique for detection of intersex in regional black bass populations
Project Number:	2014MD315B
Start Date:	7/1/2014
End Date:	6/30/2015
Funding Source:	104B
Congressional District:	MD-005
Research Category:	Biological Sciences
Focus Category:	Surface Water, Toxic Substances, Methods
Descriptors:	None
Principal Investigators:	Lance Yonkos

Publications

1. Yonkos, Lance; Christine Kim; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu, 2014. "Poultry Litter-Induced Intersex in Regional Largemouth Bass Populations," Chesapeake and Potomac Regional Chapter of the Society of Environmental Toxicology and Chemistry Annual Meeting. Grasonville, MD. April 28, 2014 (poster presentation).
2. MacLeod, Alexander; Lance Yonkos, 2014, "Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass (Micropterus salmoides) for Non-Lethal Detection of Intersex (Testicular Oocytes)," Chesapeake and Potomac Regional Chapter of the Society of Environmental Toxicology and Chemistry Annual Meeting. Grasonville, MD. April 28, 2014 (poster Presentation).
3. Yonkos, Lance; Christine Kim; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu, 2014, "Testicular oocytes and vitellogenin induction in largemouth bass (Micropterus salmoides) exposed to 17 -estradiol and poultry litter at different ages," Society of Environmental Toxicology and Chemistry 35th Annual Meeting. Vancouver, BC. November, 2014 (poster presentation).
4. MacLeod, Alexander; Lance Yonkos, 2014, "Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass (Micropterus salmoides) for Non-Lethal Detection of Intersex (Testicular Oocytes)," Society of Environmental Toxicology and Chemistry 35th Annual Meeting. Vancouver, BC. November, 2014 (poster presentation).
5. MacLeod, Alexander; Lance Yonkos, 2015. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass (Micropterus salmoides) for Non-Lethal Detection of Intersex (Testicular Oocytes), Association of Mid-Atlantic Aquatic Biologists, Cacapon, WV, March 25, 2015 (poster presentation).
6. Kim, Christine; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu; Lance Yonkos, 2014, "Poultry Litter-Induced Intersex in Regional Largemouth Bass Populations," University of Maryland College Park, 2014 Maryland Day, College Park, MD, April 26, 2014 (poster presentation).
7. Kim, Christine; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu; Lance Yonkos, 2014, "Poultry Litter-Induced Intersex in Regional

Largemouth Bass Populations," University of Maryland College Park – Undergraduate Research Day, College Park, MD, April 30, 2014 (poster presentation).

NIWR PROGRESS REPORT

PROJECT ID:
MWRRC PROJECT # 2013-MD-305B
MWRRC PROJECT # 2014-MD-315B

TITLE: VALIDATION OF NON-LETHAL LAPAROSCOPIC TECHNIQUE FOR DETECTION OF INTERSEX IN REGIONAL BLACK BASS POPULATIONS

DATE: June 15, 2015

PRINCIPAL INVESTIGATOR
Lance Yonkos, Assistant Professor
University of Maryland College Park
Environmental Science and Technology
1451 Animal Science Bldg.
College Park, MD 20742
TEL: 301-405-7871
Email: lyonkos@umd.edu

1. PROGRESS REPORT: SUMMARY

Regional Water Problem

Intersex in the form of testicular oocytes (TO) has been reported with high prevalence in regional smallmouth bass *(Micropterus dolomieu)* and largemouth bass *(Micropterus salmoides)* populations [Blazer et al 2007; 2011; Iwanowicz et al 2009; Yonkos et al 2014]. Prevalence in males from Potomac, Shenandoah, Susquehanna, and several Eastern Shore Watersheds often exceeds 80%. The presence of TO is generally accepted as evidence of exposure to endocrine disrupting compounds (EDCs) and suggestive of reproductive compromise and population level effects [Hinck et al 2009]. This raises concerns for the aquatic food web and for the quality of regional drinking water resources. Potential sources of estrogenic contaminants are numerous (e.g., WWTP effluent, agricultural and urban runoff, etc), but, as yet, no causal link to any particular source or contaminant has been identified that adequately explains the spatial heterogeneity of TO occurrence and severity [Blazer et al 2012]. Continued surveillance of resident fish populations is necessary to determine areas of particular concern and to employ a weight-of-evidence framework for identifying culprit contaminants/activities.

Black bass (largemouth and smallmouth) are climax predators within regional freshwater aquatic food webs. Because they appear uniquely sensitive to EDCs, they are the logical choice as sentinel species for monitoring contaminant effects. However, current methods for identification of TO in require field collection and sacrifice of fish for histological identification of oocytes within testis of male specimens. Generally a minimum of ten male fish is considered necessary to accurately assess TO prevalence and severity at a particular site. Because black bass are only minimally sexually dimorphic (and only during the spawning season), sampling efforts also involve collection and sacrifice of numerous females before achieving the requisite number of male fish. Where bass populations are already imperiled through over-exploitation, habitat modification, contaminant-impacts, and/or competition from invasive species, this level of experimental collection necessarily places an additional impact on population number. Thus, populations in regions where analysis of fish is most warranted are among those least able to accommodate this additional pressure. Employing a non-lethal means of determining TO prevalence will serve to alleviate this pressure while still allowing field assessment of endocrine disruption within impacted populations. Further, targeted regions can be re-sampled over multiple seasons or years to observe temporal shifts including recovery or further decline without undue influence to fish populations.

Project Objectives

Fish populations in many Chesapeake tributaries are sufficiently threatened that any additional burden of research-related collection is difficult to justify. This makes development of an effective non-lethal monitoring technique paramount. To that end, we examined the efficacy of endoscopy via the genital pore (laparoscopy) as a non-lethal, minimally-invasive sampling technique for collection of largemouth bass *(Micropterus salmoides)* gonadal tissue for the detection of testicular oocytes (TO). Laparoscopic insertion into the body cavity allows testis

tissue collection via biopsy without penetration through the body wall. The method has been demonstrated previously to be effective for non-lethal gender identification in largemouth bass by allowing internal visualization and biopsy of gonadal tissue with high survival, rapid healing, and minimal infection [Matsche 2013]. Our research expands on previous efforts by collecting multiple testis biopsies (n=5) equidistant along a single testis lobe with the purpose of quantifying TO prevalence and severity. This was addressed in four non-sequential phases: *(1) Model Development:* determination of testis biopsy number and size necessary to quantitatively establish TO prevalence and severity; *(2) Recovery and Healing:* determination of post-operative survival and healing following multiple-biopsy tissue collection; *(3) Spawning Capability:* determination of post-operative spawning capability; and *(4) Field Validation:* comparison of non-lethal laparoscopic tissue collection and conventional tissue dissection for efficacy of detecting TO in native fish collected from regional waters with a history of high intersex prevalence.

Hypotheses

H_1: Laparoscopic testicular tissue collection from mature male largemouth bass will heal rapidly and result in minimal mortality

H_2: Laparoscopic testicular tissue collection will not interfere with natural spawning of mature male largemouth bass

H_{3a}: Laparoscopic testicular tissue collection from mature male largemouth bass will allow non-lethal detection of testicular oocyte <u>prevalence</u> comparable to conventional methods that require sacrifice and dissection of specimens

H_{3b}: Laparoscopic testicular tissue collection from mature male largemouth bass will allow non-lethal quantification of testicular oocyte <u>severity</u> comparable to conventional methods that require sacrifice and dissection of specimens

Methods

The research project uses a non-lethal laparoscopic technique (pioneered by fish pathologist Mark Matsche, MD DNR, Cooperative Oxford Laboratory, Oxford, MD) to collect testis biopsy samples prior to routine collection of tissue via dissection. Comparison of efficacy of TO detection by laparoscopic tissue collection versus routine sacrifice and dissection was undertaken to determine the appropriateness of the method for field assessment of TO prevalence and severity. Post-laparoscopic survival, healing and spawning ability were also investigated to estimate the actual level of impact the procedure would have if employed on native bass populations. Efforts toward satisfaction of the several research objectives occurred concurrently with fish from various sources satisfying multiple purposes. For example, hatchery-reared and field-collected largemouth bass were used variously to ensure sufficient numbers of specimens for method comparison, model development, and/or post-laparoscopic survival, healing, and spawning assessment. Field sampling was done in coordination with on-going MD DNR, USGS, and US FWS population assessment efforts allowing use of already targeted-collections to

minimize additional sampling impacts. Processing of biopsied and dissected tissues was by routine histological processing and necessarily required sacrifice of research fish. Briefly, tissue samples were dehydrated in alcohol, embedded in paraffin, sectioned at 6 µm, and stained with hematoxylin and eosin [Luna, 1992].

Laparoscopy – Refinement of non-lethal testis tissue collection

Methods for testis tissue collection have been adapted from Matsche [2013] to allow collection of multiple testis biopsies. Briefly, individual largemouth bass were anesthetized with tricaine methanesulfonate (Finquel®, Argent Laboratories, Redmond, WA) and placed dorsal side down between angled plastic positioning blocks with procedures initiated immediately after movement ceases. The urogenital opening was gently dilated using a blunt obturator with surgical lubricant (Surgilube®) followed by insertion of the examination sheath through the urogenital opening to the urinary bladder [Matsche 2013]. The exposed end of the examination sheath was then angled 30° towards the posterior of the fish (to direct the tip toward the head) and 30° toward the left body wall. The obturator was then removed and replaced with the laparoscope and biopsy forceps which were used to perforate the urinary bladder and access the body cavity. To assist with laparoscopic viewing the coelom was inflated using a low-pressure air supply connected to a stopcock on the examination sheath. Guided by the laparoscope, the examination sheath was advanced into the coelom caudally along the left dorsal region of the body wall until the gonad was encountered and gender determined. If determined to be female, procedures were discontinued and the fish was revived (or sacrificed) as necessary. If determined to be male, the five biopsies were collected along an approximately equidistant transect from the left gonad and placed in 10% neutral buffered formalin. The examination sheath remained in body cavity with only the flexible forceps removed during all five biopsies. If survival was to be assessed, a passive integrated transponder (PIT) was inserted into the body cavity prior to removal of the examination sheath. If comparison with testis cross-sections was required, fish were sacrificed (via decapitation) and testis removed via routine dissection.

Phase I – Biopsy Validation

The primary goal of this project was to determine whether biopsies could detect TO with the same precision as existing methods using cross-sections from sacrificed fish and whether a severity index could be constructed to facilitate future research. The first step involved determining the instrument size and number of biopsies necessary to provide sufficient tissue for quantitative analysis of TO prevalence and severity. This was necessary to ascertain the utility and limitations of the laparoscopic technique as a non-lethal means of quantifying TO. The approach required taking multiple biopsies at discrete intervals along the testis using instruments of several sizes with cross-sections taken subsequently from regions between biopsies. Briefly, five biopsies were taken along the left testis lobe using forceps meant for small fish (1.7 mm diameter) and five biopsies were taken along the right testis lobe using forceps meant for larger fish (3.3 mm diameter). Routine histological processing of all biopsies and cross-sections allowed comparison of the methods for detection and enumeration of TO. For each specimen, at least one histological section from each of the five tissue segments and ten biopsies was examined via light microscopy for TO presence and other pathology. Establishment of TO prevalence involved observation of one or more discernible oocytes within preserved testis cross-

sections and/or biopsies from an individual specimens. All tissue samples of adequate quality were examined for the presence of oocytes under low and moderate magnification (4x and 10x objectives, respectively) with confirmation of potential oocytes determined under high magnification (40x objective). Determination of TO severity conformed as closely as possible to the ranking system described by Blazer et al [2007] for smallmouth bass (*Micropterus dolomieu*). Briefly, five cross-sections taken equidistant along the testis lobe were examined by light microscopy using a 10x objective (approx. 4 mm^2 of tissue) and scored for severity as follows: *Focal Distribution* (Score 1): single oocyte within a microscopic field; *Diffuse Distribution* (Score 2): more than one oocyte in a field of view without physical association with neighboring oocytes; *Cluster Distribution* (Score 3): more than one but less than five closely associated oocytes; *Zonal Distribution* (Score 4): more than five closely associated oocytes or numerous clusters in a field of view. Ranks for each section were averaged to determine a severity rank for the individual specimen. This system is currently being modified in an attempt to provide equivalent results for biopsy collected tissues. Issues that remain to be resolve include: (1) whether biopsies can consistently collect tissues from appropriate regions of the testis and (2) how many biopsy sections are necessary to provide quantitatively similar results to conventional methods.

Phase II – Survival Study

In order be deemed effective post-laparoscopy survival must be high with minimal long-term compromises to fish health. Maintaining batches of fish after the laparoscopic procedure allowed for monitoring of infection and assessing post-operative healing and survival. Fish were anesthetized and, as described above, approximately five biopsies were taken equidistant along the length of the left testis. Post-operative survival was investigated initially as part of a dedicated "survival" study (March 2014) using hatchery-reared fish (SmartFish Farms, Auburn, KY). Additional survival data was accrued on fish used in satisfaction of other project objectives. In the dedicated survival study a subset of fish was sacrificed immediately after the procedure (e.g., controls), another cohort was sacrificed at one week to investigate incidence of infection or other pathology, and the remainder were sacrificed at one month to investigate healing and establish long term survival. On sacrifice testis tissue was also collected for comparison of biopsy vs routine dissection. This served to make more robust the sample size for method validation and model development.

Phase III – Reproduction Study

The third phase of the project was intended to determine if biopsy collection of the testis adversely affects reproductive capability of male fish. It is important to determine whether fish remain competent to spawn following laparoscopic biopsy of testis. Releasing a wild-caught mature male largemouth bass rendered sterile by the laparoscopic procedure could potentially be more damaging to a population than actually sacrificing the fish on collection. He will continue to occupy a territory, consume forage species and attract females to spawn but will not fertilize eggs so will contributing no progeny.

We attempted to complete the reproduction study by field collecting largemouth bass and using laparoscopy to definitively establish gender. Females received no additional treatment while

males had the standard five biopsies collected from the left testis. Fish were then segregated by gender and held for a prescribed recovery interval (approximately 14 days) before being paired in spawning raceways at the Manning Hatchery (Cedarville, MD) and monitored for spawning activity. Unfortunately, highly variable weather conditions during the spring of 2014 made the largemouth bass spawning season unpredictable. Paired bass at the Manning Hatchery (both those treated via laparoscopy and control fish) failed to spawn. Because bass spawn only once per year we were unable to pursue this project phase any further. We were able to use the male fish from this portion of the project to further investigate survival and healing and to make more robust the sample size of fish used in comparing biopsy examination to conventional dissection.

Phase IV – Field Validation Study

The final phase of the study involved field validation of laparoscopic testis sampling as an effective tool for identifying TO prevalence and severity. To achieve this endpoint largemouth bass were collected regionally via boat electrofishing from the Potomac and Anacostia Rivers during the spring and summer 2014; largemouth and smallmouth bass was collected in conjunction with USGS efforts in Vermont during summer 2014; and largemouth bass collections were performed during spring 2015 in collaboration with University of Georgia researchers in a lake in Georgia with a history of high TO prevalence. In each system females were released once identified to minimize the number of fish removed from the population. An effort was made to collect testis via laparoscopic biopsy from a minimum of 10 mature male fish for assessment of TO prevalence and severity. An electronarcosis system was used rather than MS222 to avoid the need for quarantine. This allowed immediate release of females. Males were treated as described previously (i.e., after collection of biopsies fish were sacrificed, testis was removed and cross-sectioned, and all tissues were processed for histological examination). In all, approximately 100 field collected male bass were examined for this study. To increase the sample size (i.e., number of specimens available for comparison of TO detection via biopsy and cross-section), preserved testis tissue was sampled via biopsy from approximately 100 male largemouth and/or smallmouth bass previously collected and examined by USGS personnel. Tissues were collected "blind" from fish populations known to have high TO prevalence.

Application to Other Species

At the request of MD DNR, US FWS, and USGS fisheries biologist, the laparoscopic method has been adapted for use with several other fish species of interest:

- First is the smallmouth bass which has been found in the Potomac and Shenandoah Rivers to have a high incidence of TO. This species has also suffered population declines in these regions as a result of fish kills over the past decade. While no causal link has been established, the possibility exists that TO induction and premature mortality are related to a common contaminant exposure occurring at some earlier life stage. Laparoscopic tissue collection will allow non-lethal investigation of TO prevalence in those regions where smallmouth bass populations are at particular risk. As this species is very similar to the largemouth bass, only minimal adaptation of the method is required. Also, since smallmouth bass generally occupy a distinct range from that of largemouth bass, this expands the inland surface waters systems that can be investigated for TO using this method.

- The northern snakehead *(Channa argus)* is a large fast-growing predatory fish species native to China but now invasive and established in many Chesapeake Bay tributary rivers. The species competes effectively with largemouth and smallmouth bass but grows faster so adds an additional stressor to systems where it is present. Currently very little is known about the reproductive biology of the snakehead including spawning frequency and other fecundity criteria. Even knowledge of whether females are single or multiple spawners is lacking. For this reason the laparoscopic method is being adapted to non-lethally investigate gonad development in female northern snakehead. The intent is to track gonad development in individual female fish over protracted periods to determine seasons of maturation and establish whether fish are single or multiple spawners. This information is considered paramount when devising management strategies. As the northern snakehead differs morphologically in significant ways from the largemouth bass, modification of the laparoscopic method will be more substantial and may require different instruments.

Preliminary Findings

Laparoscopic tissue collection shows promise for detecting testicular oocytes. However, results described below are preliminary as tissue examination and analysis is on-going. Therefore, endorsement of the method is not, as yet, justified. Preliminary results do indicate that laparoscopic imaging of the body cavity provides rapid, non-lethal gender identification. This allows immediate release of females so minimizes population impacts of scientific sampling. Preliminary findings for individual objectives are as follows:

- *Model Development* — Determination of the ability of testis biopsies to detect TO requires a multitude of fish with the intersex condition, preferably demonstrating a range of severities. During initial collection efforts we encountered few fish with TO. This limited the ability to make a meaningful comparison between the two methods. Subsequent live (Vermont, June 2014; Georgia, Mar 2015) and post-mortem (USGS, Apr 2015) tissue collections have dramatically increased the number of specimens with TO. While tissue collection and processing efforts are complete, analysis of histological slides is ongoing and won't be complete until the end of June. Therefore a detailed comparison of TO detection via biopsy collection vs dissection is not yet be completed. Preliminary results suggest that overall, biopsy collection via laparoscopy was able to detect TO prevalence with approximately 80% of the efficacy of conventional cross-sections from sacrificed fish. There was, however, significant variability between sampled populations with biopsy collection actually exceeding cross-section sampling in at least one instance. Due to the relatively small amount of area available in biopsies for observation (1.7 mm diameter) compared to conventional cross-sections (≥10 mm diameter), assessment of TO severity was somewhat limited. Preparation of additional sections may address this concern but will necessitate modification of the severity index to account for the increase in observed sections.

- *Survival* — Results from the dedicated "survival" study indicate post-surgery survival to 28 days is high (≥ 80%) as is healing based on integrity of the urinary bladder (≥ 90%). Survival of laparoscoped fish employed in satisfaction of other project *Phases* (e.g., spawning capability) indicate that this is a reasonable estimate of survival. Some caveats to be explored include the implications of season and/or water temperature on survival and the health of the fish on collection. For example, fish with large parasite loads, like those collected from the Anacostia River, already have compromised immune function suggesting a greater likelihood of post-operative infection and subsequent mortality.

- *Spawning Capability* — Eight pairs of largemouth bass were maintained in spawning raceways at the Manning Hatchery, Cedarville, MD. These fish were collected regionally (mostly from the Potomac River) over approximately the month of April 2014. Unfortunately, unusually cold and variable spring weather impeded normal spawning of largemouth bass in the region, both in natural surface waters and in the hatchery ponds. While male fish in raceways demonstrated typical spawning behaviors (e.g., nest preparation and coaxing of females), females released no eggs so males released no sperm. Without the natural release of eggs to induce the subsequent release of milt, post-operative spawning capability could not be assessed. As largemouth bass spawning is seasonal and highly

temperature dependent, we were unable to address this research phase during the period of project

- *Field Validation* — In total, nearly 100 male specimens (predominantly largemouth bass and a smaller cohort of smallmouth bass) were field collected for this study. Collections occurred in regional waters as well as from a region in Vermont with a known history of high intersex occurred (June, 2014) and from a similarly impacted lake in Georgia (March, 2015). Final results are pending but appear promising.

Significance

Preliminary results suggest laparoscopic tissue collection has promise as a non-lethal field sampling strategy. Since mortality is generally low (e.g., < 15%) the method appears especially well suited for use in regions where fish populations are already compromised. The method also shows significant adaptability for application to other species of interest, broadening the scope of potential studies. This procedure could provide fishery managers and scientists with an additional tool for monitoring fish populations for intersex both temporally (by repetitive sampling of individual fish) and spatially (by including compromised regions without causing undue impact). Field studies could be designed to correlate observed effects (i.e., TO prevalence and severity) with particular land use practices (e.g., non-point source agricultural runoff, WWTP effluent discharges) in an attempt to identify culprit contaminants and devise management strategies. There also exist the potential to use the method to non-lethally sample other tissues (e.g., liver) for contaminant analysis.

REFERENCES

Blazer VS, Iwanowicz LR, Iwanowicz DD, Smith DR, Young JA, Hedrick JD, Foster SW, Reeser SJ. 2007. Intersex (testicular oocytes) in smallmouth bass Micropterus dolomieu from the Potomac River and selected nearby drainages. *J Aquat Anim Health* 19:242–253.

Blazer VS, Iwanowicz LR, Henderson H, Mazik PM, Jenkins JA, Alverez DA, Young JA. 2012. Reproductive endocrine disruption in smallmouth bass (Micropterus dolomieu) in the Potomac River basin: Spatial and temporal comparisons of biological effects. *Environ Monit Assess* 184:4309–4334.

Hinck JE, Blazer VS, Schmitt CJ, Papoulias DM, Tillitt DE. 2009. Widespread occurrence of intersex in black basses (Micropterus spp.) from U.S. rivers, 1995-2004. *Aquat Toxicol* 95:60–70.

Iwanowicz LR, Blazer VS, Guy CP, Pinkney AE, Mullican JE, Alvarez DA. 2009. Reproductive health of bass in the Potomac, USA drainage: Part 1. Exploring the effects of proximity to wastewater treatment plant discharge. *Environ Toxicol Chem* 28:1072–1083.

Luna LG. 1992. Histopathologic Methods and Color Atlas of Special Stains and Tissue Artifacts. American Histolabs, Gaithersburg, MD, USA.

Matsche, MA. 2013. A portable electro-immobilization and laparoscopy system for sex determination and gonadal biopsy in largemouth bass *Micropterus salmoides. Journal of Fish Biology* 83:1391–1400.

Yonkos LT, Friedel EA and Fisher DJ. 2014. Intersex (Testicular Oocytes) in Largemouth Bass *(Micropterus salmoides)* on the Delmarva Peninsula, USA. *Environ Toxicol Chem* 33:1163–1159.

2. PUBLICATIONS ASSOCIATED WITH THE RESEARCH PROJECT

As the work is on-going, no publications have yet been generated associated with this research. There have been numerous poster and platform presentations including seven abstracts accepted at regional and national professional society meetings.

Accepted abstracts at professional scientific meetings

MacLeod, Alexander; Lance Yonkos. 2015. Non-lethal Detection of Intersex (Testicular Oocytes) from Largemouth Bass *(Micropterus salmoides)* Using Laparoscopy. Society of Environmental Toxicology and Chemistry 35th Annual Meeting. Vancouver, BC. November, 2015 (platform presentation; abstract accepted)

MacLeod, Alexander; Lance Yonkos. 2015. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass *(Micropterus salmoides)* for Non-Lethal Detection of Intersex (Testicular Oocytes). Chesapeake and Potomac Regional Chapter of the Society of Environmental Toxicology and Chemistry Annual Meeting. Columbia, MD. April 24, 2015 (platform presentation).

MacLeod, Alexander; Lance Yonkos. 2015. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass *(Micropterus salmoides)* for Non-Lethal Detection of Intersex (Testicular Oocytes). Association of Mid-Atlantic Aquatic Biologists. Cacapon, WV. March 25, 2015 (poster presentation).

MacLeod, Alexander; Lance Yonkos. 2014. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass *(Micropterus salmoides)* for Non-Lethal Detection of Intersex (Testicular Oocytes). Society of Environmental Toxicology and Chemistry 35th Annual Meeting. Vancouver, BC. November, 2014 (poster presentation).

Yonkos, Lance; Christine Kim; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu. 2014. Testicular oocytes and vitellogenin induction in largemouth bass *(Micropterus salmoides)* exposed to 17β-estradiol and poultry litter at different ages. Society of Environmental Toxicology and Chemistry 35th Annual Meeting. Vancouver, BC. November, 2014 (poster presentation).

MacLeod, Alexander; Lance Yonkos. 2014. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass (*Micropterus salmoides*) for Non-Lethal Detection of Intersex (Testicular Oocytes). Chesapeake and Potomac Regional Chapter of the Society of Environmental Toxicology and Chemistry Annual Meeting. Grasonville, MD. April 28, 2014 (poster presentation).

Yonkos, Lance; Christine Kim; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu. 2014. Poultry Litter-Induced Intersex in Regional Largemouth Bass Populations. Chesapeake and Potomac Regional Chapter of the Society of Environmental Toxicology and Chemistry Annual Meeting. Grasonville, MD. April 28, 2014 (poster presentation).

Public presentations:

MacLeod, Alexander; Lance Yonkos. 2015. Non-lethal Detection of Intersex (Testicular Oocytes) from Largemouth Bass *(Micropterus salmoides)* Using Laparoscopy. MS Exit Seminar - Department of Environmental Science and Technology Seminar Series. April 15, 2015.

MacLeod, Alexander; Lance Yonkos. 2014. Validation of a Laparoscopic Method for Collecting Testis from Largemouth Bass (*Micropterus salmoides*) for Non-Lethal Detection of Intersex (Testicular Oocytes). University of Maryland - Environmental Science and Technology Department Annual Graduate Student Research Day. College Park, MD, May 13, 2014.

Kim, Christine; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu; Lance Yonkos. 2014. Poultry Litter-Induced Intersex in Regional Largemouth Bass Populations. University of Maryland College Park – Undergraduate Research Day. College Park, MD, April 30, 2014.

Kim, Christine; Angela Leasca; Winston Liu; Shivani Patel; Laura Poulsen; Shefali Shah; Taylor Throwe Renuka Tripu; Lance Yonkos. 2014. Poultry Litter-Induced Intersex in Regional Largemouth Bass Populations. University of Maryland College Park. 2014 Maryland Day. College Park, MD, April 26, 2014.

3. STUDENTS SUPPORTED BY RESEARCH PROJECT

One master's student in the University of Maryland - Department of Environmental Science and Technology, Alex MacLeod, focused the majority of his thesis research on validation of laparoscopic tissue collection as a viable means of non-lethal testicular oocyte detection and quantification. By adapting techniques and equipment to provide quality results, he optimized the method for rapid and reproducible biopsy collection on bass testis from fish of various ages/sizes. He also adapted the method for use on other species including the invasive northern snakehead *(Channa argus)*. Additionally, he worked with regional partners to demonstrate the utility of laparoscopy as a sound approach for field monitoring of fish condition. Included were fisheries biologists and environmental toxicologists from MD DNR, USGS Leetown Science Center, US FWS Chesapeake Bay Field Office, the University of Georgia, and elsewhere.

A Gemstone Undergraduate Research Team comprised of eight (8) undergraduate students from various University of Maryland academic departments was been supported in part by this MWRR-funded project. They investigated poultry litter-induced intersex in largemouth bass by exposing batches of fish at various ages to aqueous poultry litter mixtures before growing to maturity for assessment of testis pathology. While they did not actually use the laparoscopic procedure on their test fish, they were trained on the method and assisted Alex MacLeod in his research efforts.

4. NOTABLE ACHIEVEMENTS AND AWARDS

Alex MacLeod was awarded a $5,000 Dean's Fellowship from the College of Agriculture and Natural Resources largely in recognition of the quality of his laparoscopy validation research project. He presented preliminary results of the project at the Society of Environmental Toxicology and Chemistry (SETAC) national (poster; Nov 2014) and regional (platform; Apr 2015) meetings, and will present final results of the research at the 2015 SETAC national meeting in November. Alex will complete and defend his thesis in August and expects to submit a manuscript to *Transactions of the American Fisheries Society* before the end of the year.

Steel Slag-Induced Surface Water and Groundwater Contamination (Graduate Fellowship)

Basic Information

Title:	Steel Slag-Induced Surface Water and Groundwater Contamination (Graduate Fellowship)
Project Number:	2014MD316B
Start Date:	3/1/2014
End Date:	9/30/2014
Funding Source:	104B
Congressional District:	MD-005
Research Category:	Water Quality
Focus Category:	Geochemical Processes, Groundwater, Non Point Pollution
Descriptors:	None
Principal Investigators:	Kaye Lorraine Brubaker

Publications

There are no publications.

MARYLAND WATER RESOURCE RESEARCH CENTER
Summer Graduate Fellowship 2014 Final report

Steel Slag-Induced Surface Water and Groundwater Contamination

Asli YALCIN DAYIOGLU

Advisor: Dr. Ahmet H. AYDILEK

1. INTRODUCTION

The American Society of Civil Engineers estimates that nearly $3 trillion is needed over a five-year period to bring the nation's infrastructure to a good condition. As a result, there is a policy shift at the state level aimed at substantially increasing the use of such materials in geotechnical construction. For instance, steel slag is a material that continues to receive increasing interest in several States, including Maryland. Steel slag, a by-product of steel making and approximately 15% by mass of the original steel output, is produced during the separation of the molten steel from impurities in steel-making furnaces. Steel slag generation in the U.S. is estimated to be in 9-11 million metric tons, and nearly 40% of the total slag is currently being disposed in landfills or lagoons, occupying valuable land space. Use of steel slag in highway structural fills (i.e., embankments) is an efficient way to recycle large amounts of material in engineering applications. Processed steel slag has favorable mechanical properties for use in embankment fills, including its good compactibility and high friction angle.

Even though mechanical properties of the steel slag embankments deemed satisfactory, one key issue that precludes use of steel slag in embankments is the surface and groundwater impacts caused by metals in the steel slag leachate. Steel slag is mainly composed of environmentally benign oxides of calcium, silicon and iron but may also contain low concentrations of potential toxic elements, including chromium and vanadium. However, the high calcium and iron levels create an environment that is generally conducive to sequestering these pollutants. As rainwater percolates through the embankment profile, trace elements leach from the steel slag and migrate downward through the subgrade soils until they reach the groundwater table or they are transported by rivers and streams. Furthermore, the pH conditions and potential for pollutant leaching will be highly influenced by the intended slag application and the possible use of simple additives to mitigate adverse properties.

The objectives of the proposed study were to evaluateand mitigate the surface water and groundwater pollution by steel slag used in highway construction. These objectives were addressed through a combination of laboratory column leach tests and computer modeling.

2. LABORATORY STUDY

A quantitative analysis of the contaminant leaching in the environment was addressed in laboratory column tests. The tests were conducted in fixed-wall columns specially constructed to minimize contaminant losses. After being packed into the columns, the test specimens were leached with 0.02 M NaCl in deionized water to simulate water percolating through a constructed embankment. In order to simulate the flow of leachate through an embankment and the underlying subgrade layers, sequential columns were set up, consisting of the virgin or treated steel slag in the first column and the subgrade soil in the second column.

Two-year field aged steel slag samples used in this study were originated from eastern part of Maryland. Water treatment residual (WTR) obtained from the Rockville Drinking Water Treatment Plant in Potomac, Maryland were used in stabilization of steel slag samples via pH reduction. WTR is an aluminum-based sludge which is a by-product of drinking water treatment that uses alum ($Al_2(SO_4)_3.xH_2O$) as coagulant. This sludge cake is dried and sieved through U.S. #40 sieve, and added to steel slag in different percentages by dry weight. A borrow material ($\gamma_{d\ max}$ = 16.4 kN/m^3, w_{opt} = 26 %) obtained from Maryland State Highway Administration and used for encapsulating by products in embankment construction was also included in the testing program.

A sequential column leach test (SCLT) procedure was followed. The first column was used to house the pure steel slag or WTR-slag mixtures. The effluent exited from this column was directly sent to the next column containing subgrade soil or encapsulting borrow material/subgrade soil combination. Thus, the characterics of the leachate coming the fisrt column was expected to simulate contamination of surface waters while the second column effluent was expected to simulate contaminant transport in groundwater (Figure 1). The leachate samples from both columns were collected in acid-washed plastic bottles and measured for pH and electrical conductivity. The concentrations of metals were determined via inductively coupled plasma spectrophotometry. A photograph of the SCLT setup is shown in Figure 2.

Figure 3a shows the temporal characteristics of effluent pH for both columns. pH stays fairly constant throughout the tests. An increase in WTR content decreases the pH of pure slag material; however, addition of 30% WTR by weight still results in pH>10.5. On the other hand, effluent passing through the encapsulating clay layer for both cases yield pH ~ 7 due to high buffering capacity of the clayey borrow material. Such decrease in pH is can be associated with adsorption of free calcium on negatively charged clay particles (Figure 3b).

Calcium and arsenic metal concentrations for pure steel slag, steel slag+30 WTR mixture, and steel slag+30 WTR/encapsulation clay materials are shown in Figure 3. For both elements, the metal concentrations decreased with addition of 30% WTR by weight and a further decrease in concentations was observed when percolation through the encapsulation layer occurred. It is believed that addition of WTR results in formation of calciumaluminasilicates and decreases the free Ca within the leachate.

The release of arsenic exhibited a first-flush pattern followed by a decrease in concentrations; the first-flush effect was more clearly pronounced for the %100 slag specimen. Most of the metals were leached out at the beginning of the tests, and eventually reached an equilibrium concentration over time (at 30-40 pore volumes of flow). The higher initial pH of the effluent solutions of steel slag and steel slag+30 WTR may have contributed to an increase in the solubility of As due to unavailability of positively charged surfaces.

3. COMPUTER MODELING

The transport of the metals in soil vadose zone and groundwater were simulated using a computer model known as WiscLEACH, a recent and verified algorithm for simulating water and solute movement in two-dimensional variably saturated media (Li et al. 2006, 2007, Cetin et al. 2012, 2014). For transport in surface waters, such as rivers and streams, van Genuchten (2013) solutions were utilized to develop a numerical model to estimate concentration distributions as a function of time and distance.

Figure 4 demonstrates the transport of arsenic in the stream after 20, 50, 100, 200, 500 and 1000 s. The results are obtained based on the assumptions of an instantaneous injection of 1 kg of a solute in the main channel of a river having a cross-section of 10 m^2, an average flow velocity of 1 m/s and a dispersion coefficient of 5 m^2/s, per recommendations of van Genuchten (2013) and De Smedt et al. (2005). As depicted in Figure 5, As concentration decreases significantly with increasing travel time, i.e. distance from the point of contact (corner of the embankment). The data in Figure 5 also suggest that the first-flush effect is minimized by WTR addition to slag. Numerical simulations performed by WiscLEACH showed that there is no metal leaching within 200 years due to low permeability of the subgrade (Figure 5), even though the As concentrations measured in column tests can be above the EPA Maximum Concentration Levels (MCL). A parametric study is needed to investigate the effect of soil subgrade, encapsulation soil type, and WTR addition on leaching characteristics of steel slag.

The laboratory test results obtained in current study suggest that aqueous samples should be collected during the field construction phase, since metal concentrations in leachates that come out of the

mixtures at that stage are expected to be higher. On the other hand, the laboratory test results suggested that metal concentrations are expected to be very low after this first-flush period. A groundwater transport modeling study coupled with the current laboratory data can be useful in estimating the transport of the metals under field conditions. It is likely that concentrations of metals may be greatly reduced far away from the source before reaching the point of compliance due to chemical and biological activity in the field.

Even though the results obtained by simulating the pollutant characteristics of steel slag leachate flowing through the encapsulated embankment and the subgrade are promising, pH dependent leaching tests should be performed in order to understand the influence of field pH excursions on metal leaching.

References

Cetin, B., Aydilek, A.H., and Li, L. (2012). "Experimental and Numerical Study of Metal Leaching from Fly Ash Amended Highway Bases", *Waste Management*, Vol. 32, No. 5, pp. 965-978

Cetin, B., Aydilek, A.H, and Li, L. (2014). "Trace Metal Leaching from Embankment Soils Amended with High Carbon Fly Ash", *Journal of Geotechnical and Geoenvironmental Engineering, ASCE*, Vol. 140, No. 1, in press.

De Smedt, F., Brevis, W., Debels, P., (2005). "Analytical solution for solute transport resulting from instantaneous injection in streams with transient storage." *Jounal of Hydrology*., 315, pp. 25–39.

Li, L., Benson, C., Edil, T., and Hatipoglu, B. (2006) WiscLEACH: "A Model for Predicting Ground Water Impacts from Fly-Ash Stabilized Layers in Roadways." *GeoCongress 2006*: 1-6.

Li, L, Benson, C.H., Edil, T.B., and Hatipoglu, B., (2007). "Groundwater impacts from coal ash in highways." *Waste and Resource Management* 159, 151 – 163.

van Genuchten, M.Th., Leij, F.J., Skaggs, T.H., Toride, N., Bradford, S.A., Pontedeiro, E.M., (2013). "Exact analytical solutions for contaminant transport in rivers. 1. The equilibrium advection-dispersion equation." *J. Hydrol. Hydromech*., 61(2), 146–160.

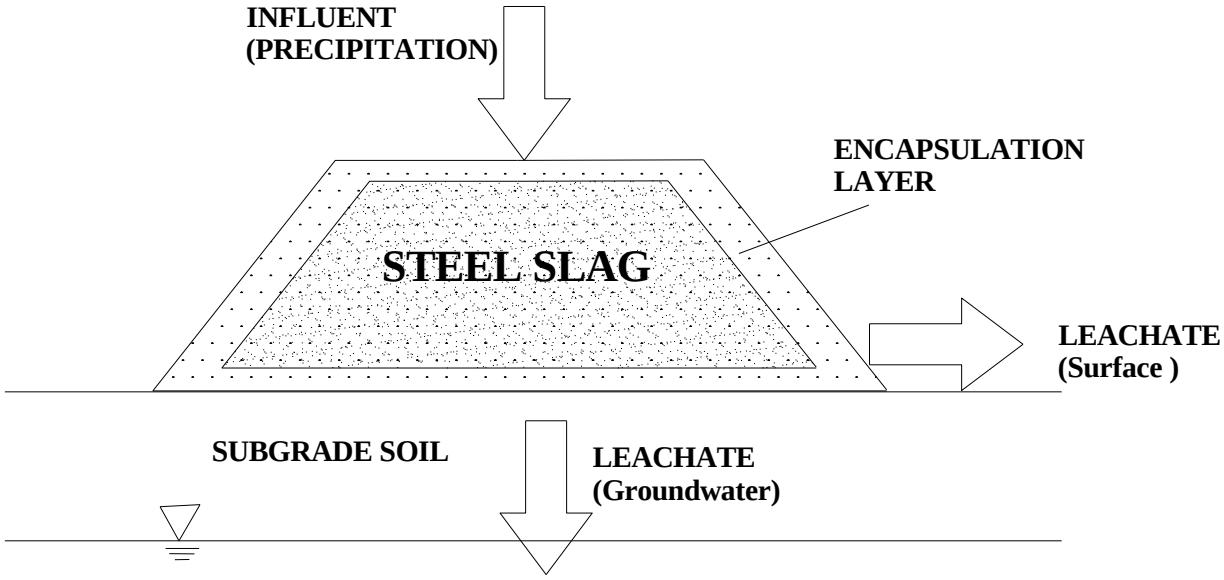


Figure 1. Conceptual Flow Model of Steel Slag Leachate Through Embankment

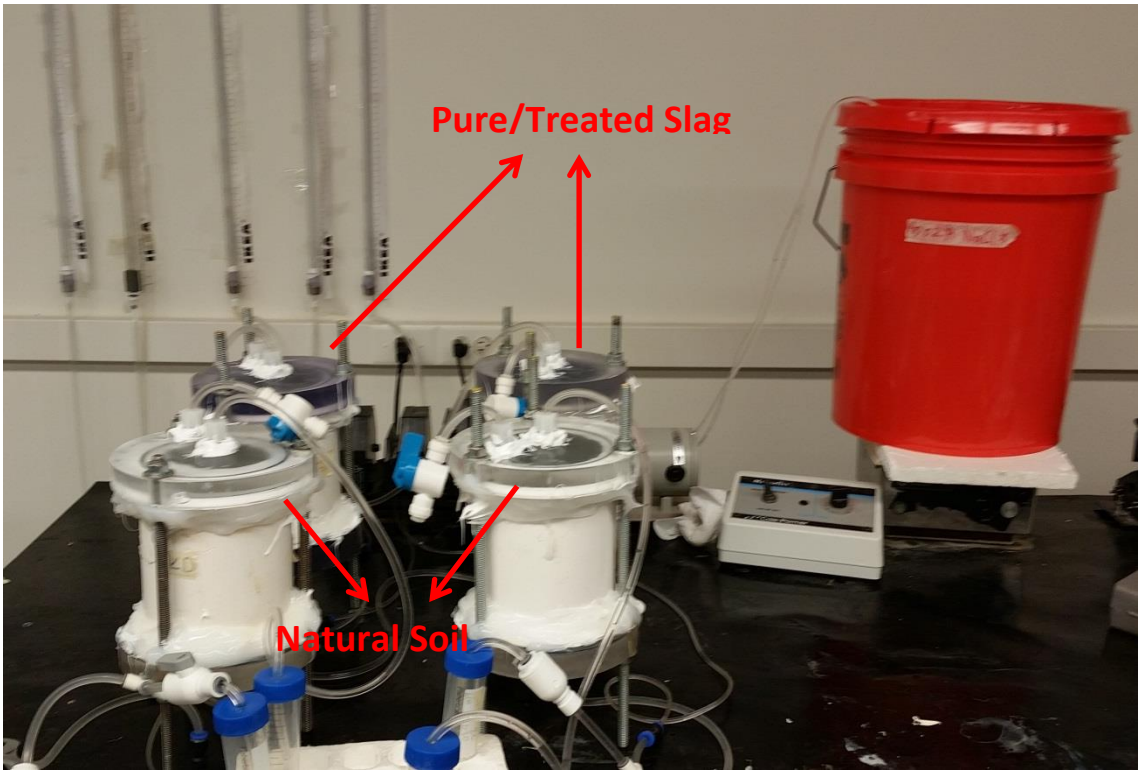
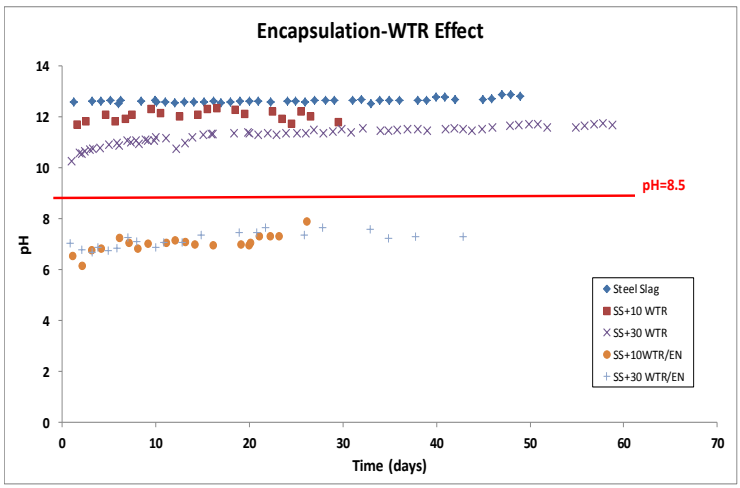
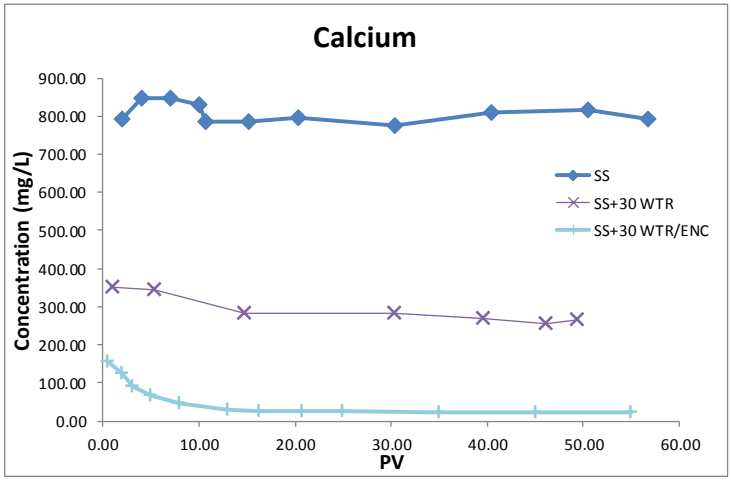


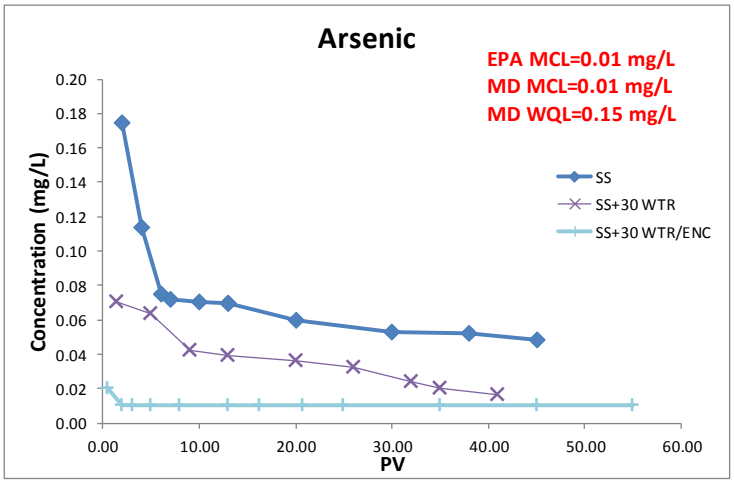
Figure 2. Sequential Column Leach Test (SCLT) Setup



(a)



(b)



(c)

Figure 3. (a) pH of the effluent from SCLT conducted with WTR mixtures, elution curves for (b) Ca, and (c) As in SCLTs (Note: 30 WTR designates the specimen with 30% WTR by weight and 30 WTR/ENC designates the effluent of second column with encapsulation clay inside. MCL:Maximum Contaminant Levels for Drinking Water. WQL: water quality limits for protecting the human health and aquatic life in fresh water.

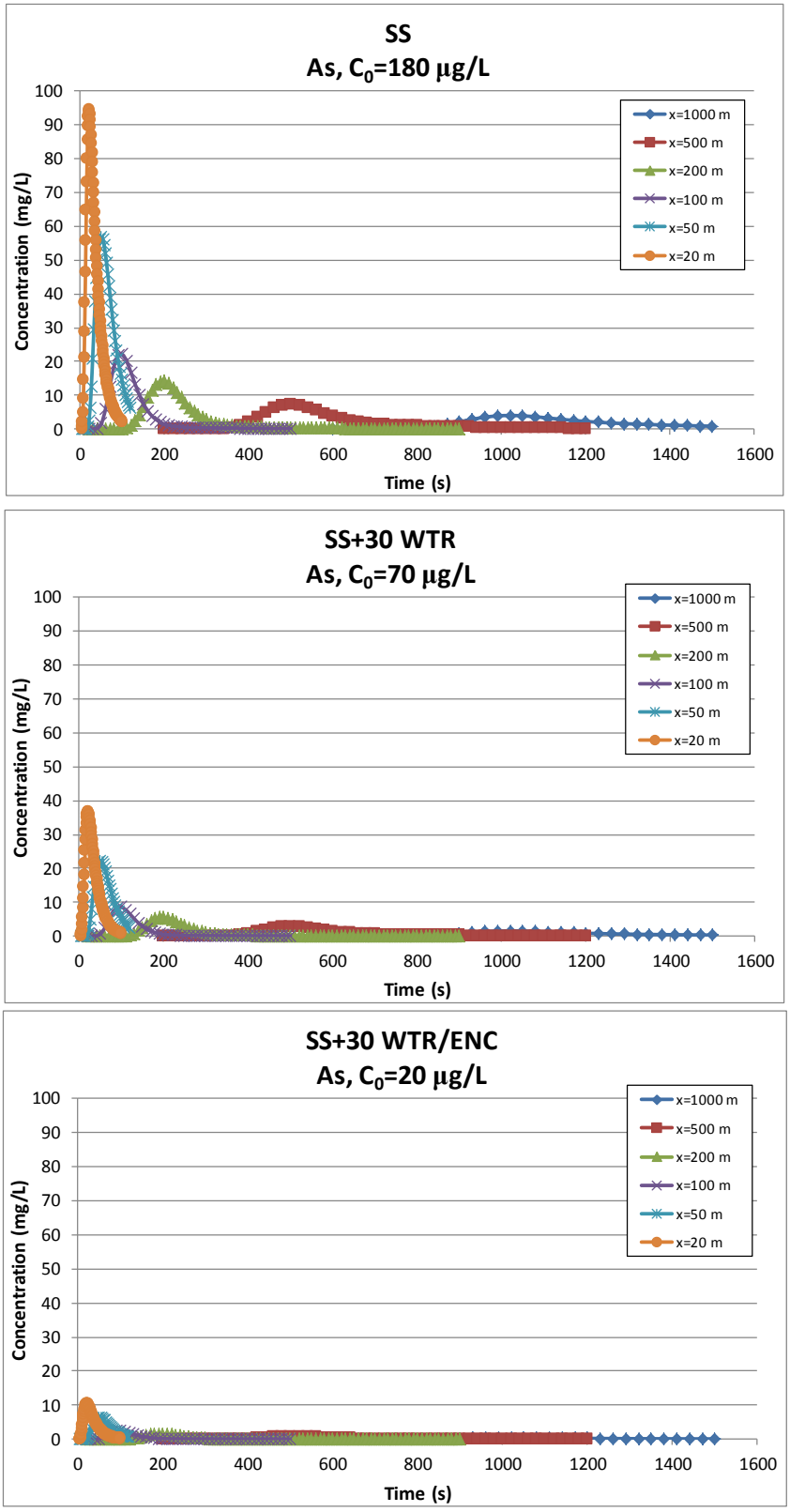


Figure 4. As the solute distributions in the stream after 20, 50, 100, 200, 500 and 1000 s

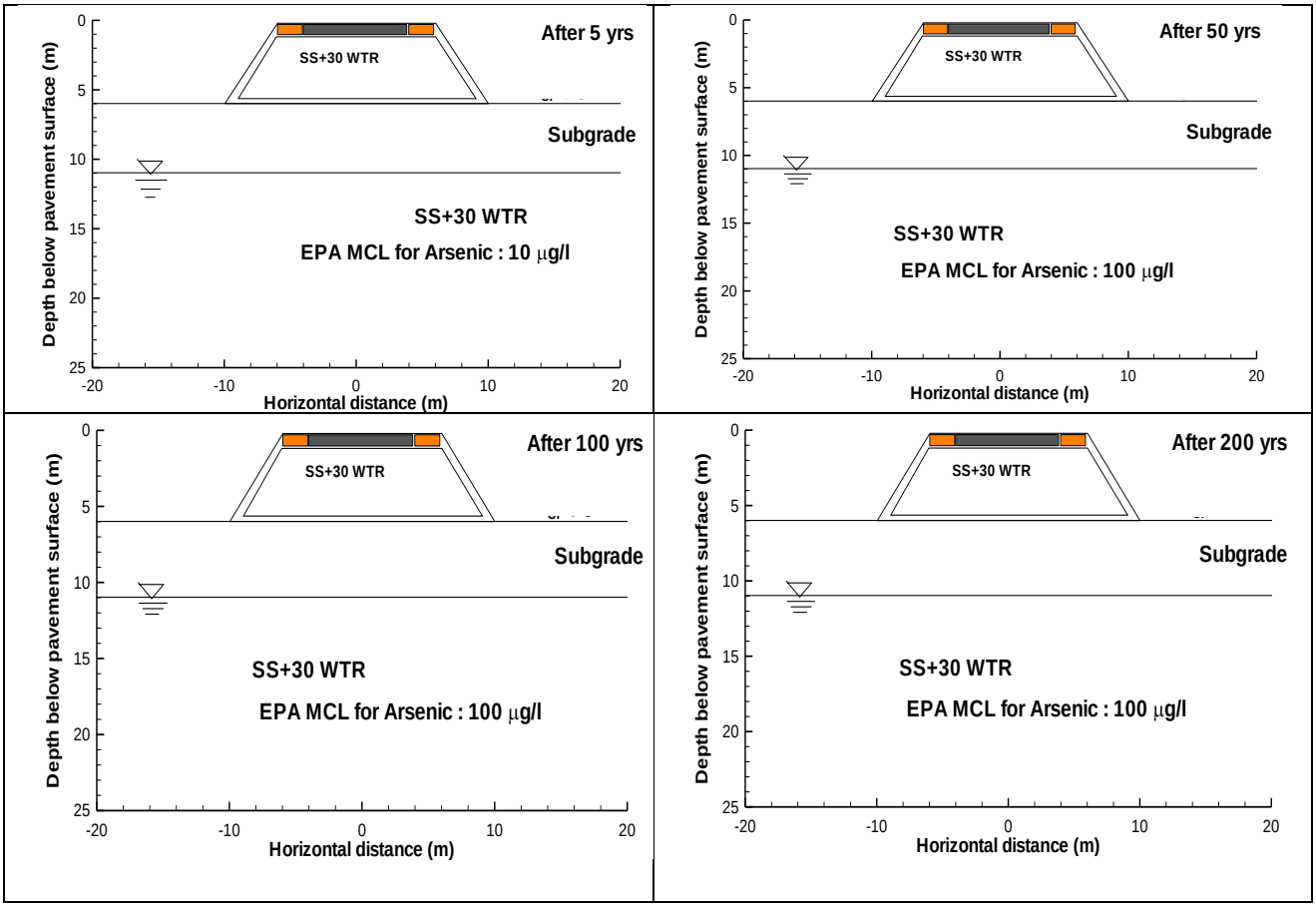


Figure 5. Predicted concentrations of arsenic in vadose zone and groundwater (Note: 30 WTR designate the specimens with 30 % Water Treatment Residual.

Impacts of Stormwater Management on Greenhouse Gas Fluxes from Urban Streams (Graduate Fellowship)

Basic Information

Title:	Impacts of Stormwater Management on Greenhouse Gas Fluxes from Urban Streams (Graduate Fellowship)
Project Number:	2014MD317B
Start Date:	3/1/2014
End Date:	9/30/2014
Funding Source:	104B
Congressional District:	MD-005
Research Category:	Climate and Hydrologic Processes
Focus Category:	Surface Water, Nutrients, Geochemical Processes
Descriptors:	None
Principal Investigators:	Kaye Lorraine Brubaker

Publications

There are no publications.

Impacts of Stormwater Management on Greenhouse Gas Fluxes from Urban Streams

Rose Smith
Department of Geology
University of Maryland College Park

Report to the Maryland Water Resources Research Center
2014 Fellowship progress report
June 14, 2015

1. Background and Objectives

Streams are potential hot spots for GHG emissions in watersheds. Water quality and watershed land use are important factors regulating greenhouse gas (GHG) production and emissions. Several studies have shown that GHG production is greater in nitrogen-loaded streams. In urban streams, specific management practices such as created stormwater wetlands, and septic systems are likely important contributing factors. This study explored the hypothesis that stormwater management strategies could significantly reduce nitrogen loads and GHG emissions in urban watersheds of Chesapeake Bay. Specifically, this study seeks to determine 1) whether stormwater management significantly affects GHG emissions from headwater streams, 2) whether there is a significant interaction between watershed management and temporal variability GHG fluxes from streams, and 3) whether there is a significant interaction between watershed management and the spatial variability of GHG fluxes along the urban stream continuum.

This project examined GHG and nitrogen dynamics in two sub-basins of Gwynns Falls in Baltimore County, MD. Dead Run was developed earlier has prominent stream burial with several retrofitted stormwater management areas, while Red Run has more recent (early 2000's) development and comprehensive stormwater management. Eight headwater streams were selected as study sites and gaged with continuous flow monitoring equipment based on a gradient of stormwater management and urban development age. Of these eight suburban first order watersheds, two are in Dead Run and unmanaged (Old/Degraded), two in Dead Run are stormwater retrofits (Old/Managed), two in Red Run have comprehensive stormwater management (New/ Managed), and two in red run have septic infrastructure (New/Septic).

2. Methods and Results

During summer 2014, I completed two years of sampling dissolved nitrogen and carbon concentrations, and one year of bi-weekly dissolved GHG concentrations at eight headwater stream gage sites. I found that CO_2, N_2O and CH_4 were net sources to the atmosphere at all on all sampling dates. Saturation ratios (measured / equilibrium concentrations) were also significantly ($p<0.01$) different between the four watershed management types (old vs. new/ managed vs. degraded). N_2O saturation ratio varied from 1.1 – 47 during this study. I found very high N_2O concentrations in streams draining

diffuse septic systems and high total dissolved nitrogen (TDN) concentrations, and the lowest N_2O in streams with connected floodplains and low TDN. CO_2 was highly correlated with N_2O and across all sites and dates (r2=0.84) as well as with CH_4, although the relationship varied between sites (Figure 1). CO_2 saturation ratio varied from 1.1 – 73. CH_4 concentrations were always super-saturated by a factor of 3.0 - 2157. The highest CH_4 concentrations were measured the New/Managed streams, which were characterized by adjacent stormwater management wetlands and low concentrations of nitrate and TDN (Figure 2).

Seasonal variability differed between gases and with management. CO_2 and N_2O concentrations did not show seasonal variation at any sites. By contrast, CH_4 was similar year-round for all 'Degraded' sites and peak in late fall and minimum in early spring in all 'Managed' sites (Figure 3).

In order to assess the spatial variability in GHG and solute fluxes at a larger watershed-scale, I conducted longitudinal surveys of dissolved GHG and solute concentrations and solute were performed in Red Run and Dead Run in March 2014 and December 2014. Samples for dissolved GHG's, DOC, TDN, and NO_3^- were collected along with discharge measurements approximately every 500m along the main stem of each stream. Longitudinal sampling in spring and fall showed consistently super-saturated concentrations of all three gases throughout both watersheds (Figure 4).

3. Next Steps

The next steps in this project include modeling gaseous and solute fluxes and completing statistical analyses, and submitting the manuscript to a peer reviewed journal. Gas flux estimations will be conduced using empirical relationships between gas transfer velocity and gas flux measurements in nearby streams. Solute flux estimates will be carried out using the USGS- LOADEST model as soon as rating curves are completed for the headwater stream gages.

4. Figures

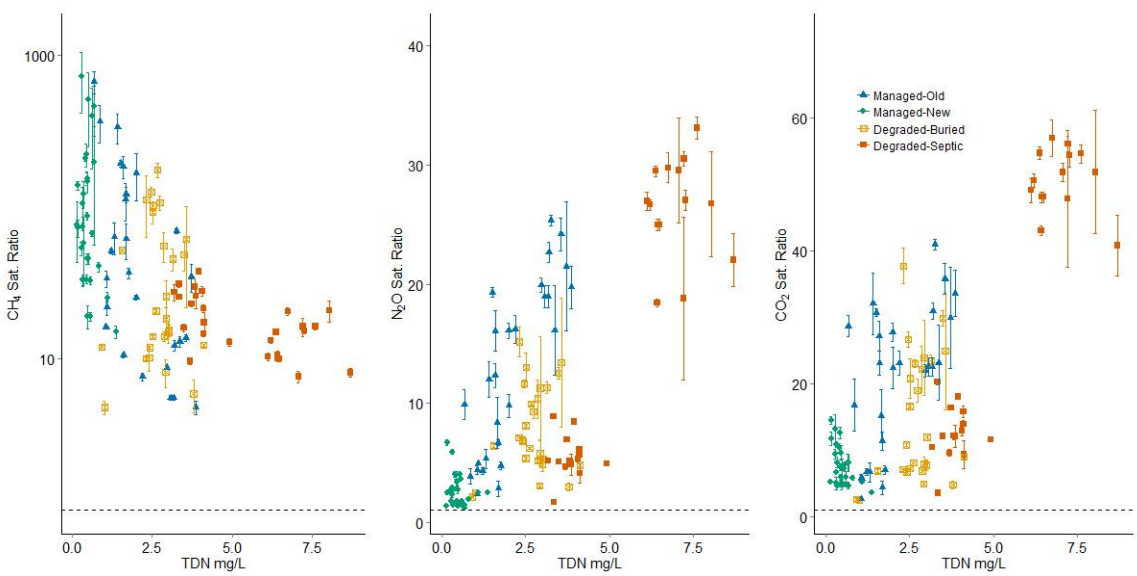


Figure 1. Relationships between concentrations of TDN and saturation ratios for all three GHG's. TDN is positively correlated with CO_2 and TDN, and negatively correlated with CH_4.

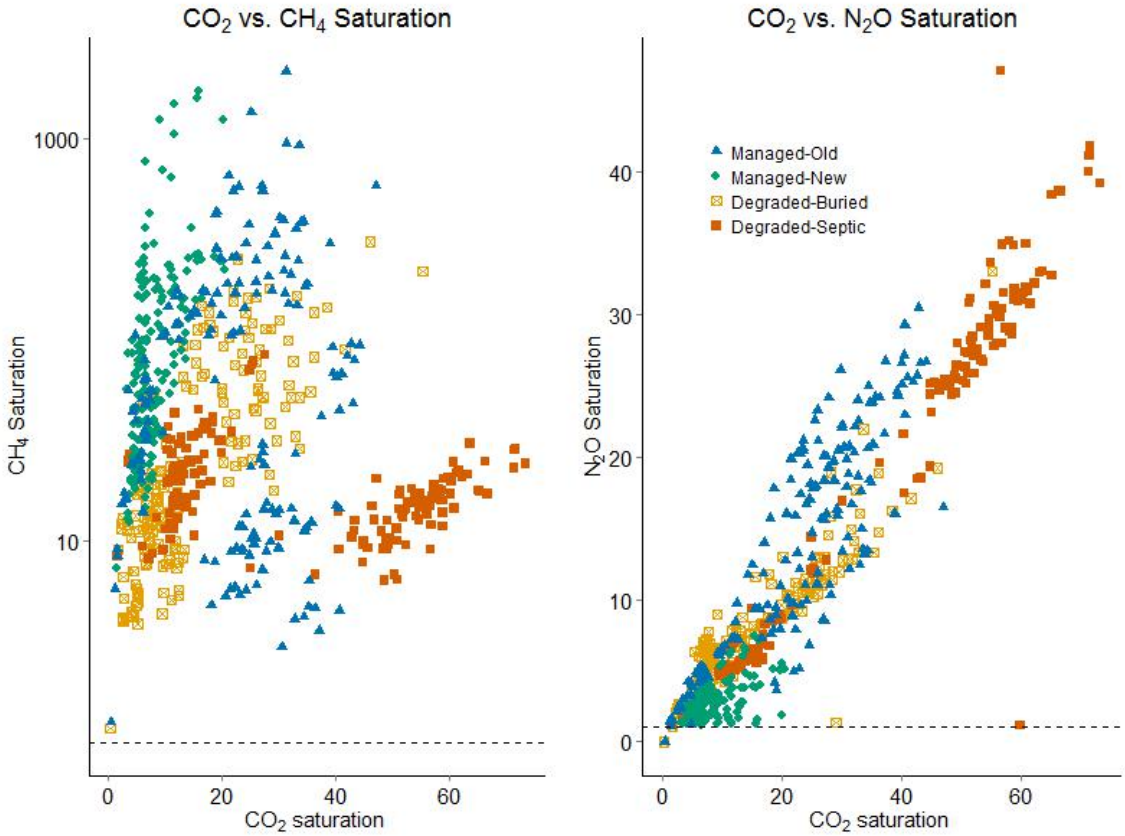


Figure 2. This figure shows correlations between CO_2 and the other two (CH_4 and N_2O) GHG's. The relationship between CO_2 and CH_4 differences between sites, however the CO_2: N_2O ratio is more constant.

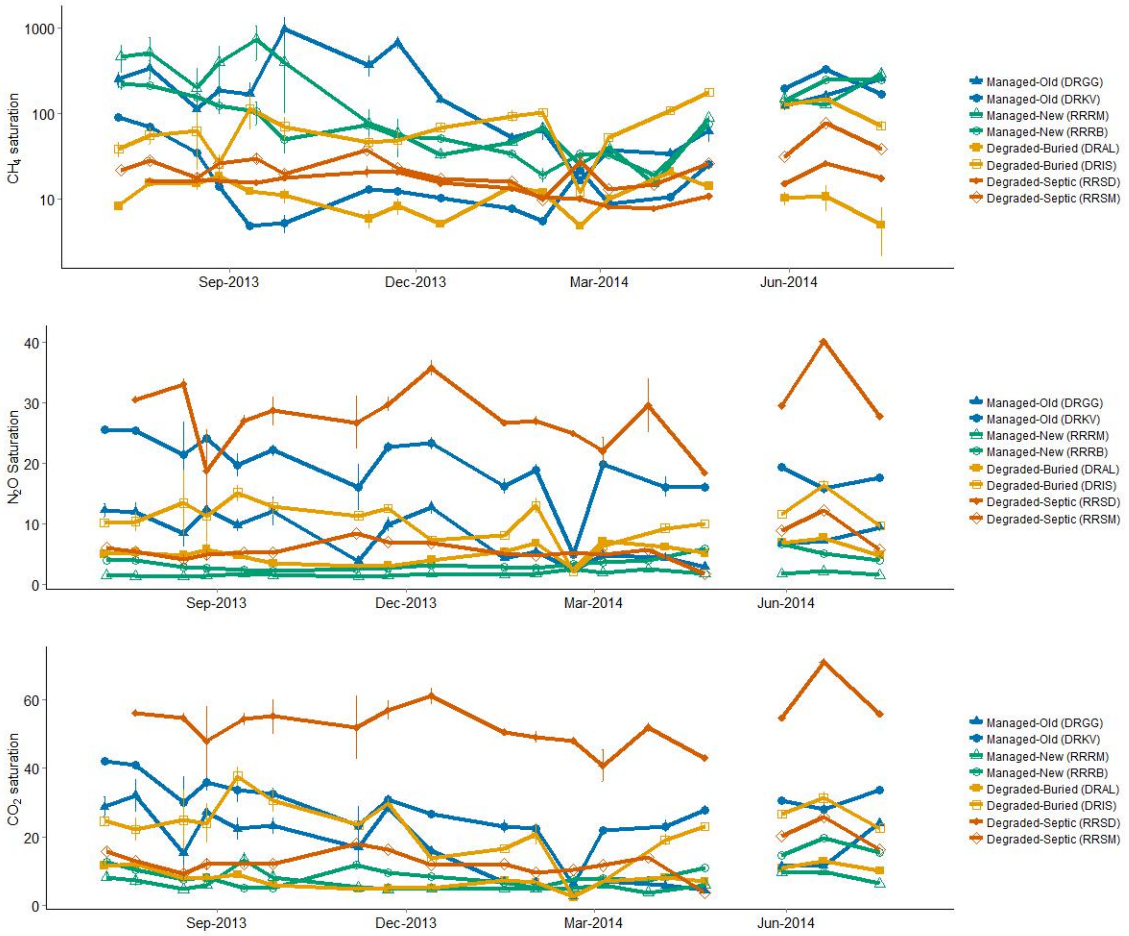


Figure 3. Time-series of GHG saturation ratios. CO_2 and N_2O show similar pattern- no seasonal differences. CH_4 shows seasonal pattern at managed (blue) sites and no seasonal pattern in degraded/ septic and degraded/buried sites.

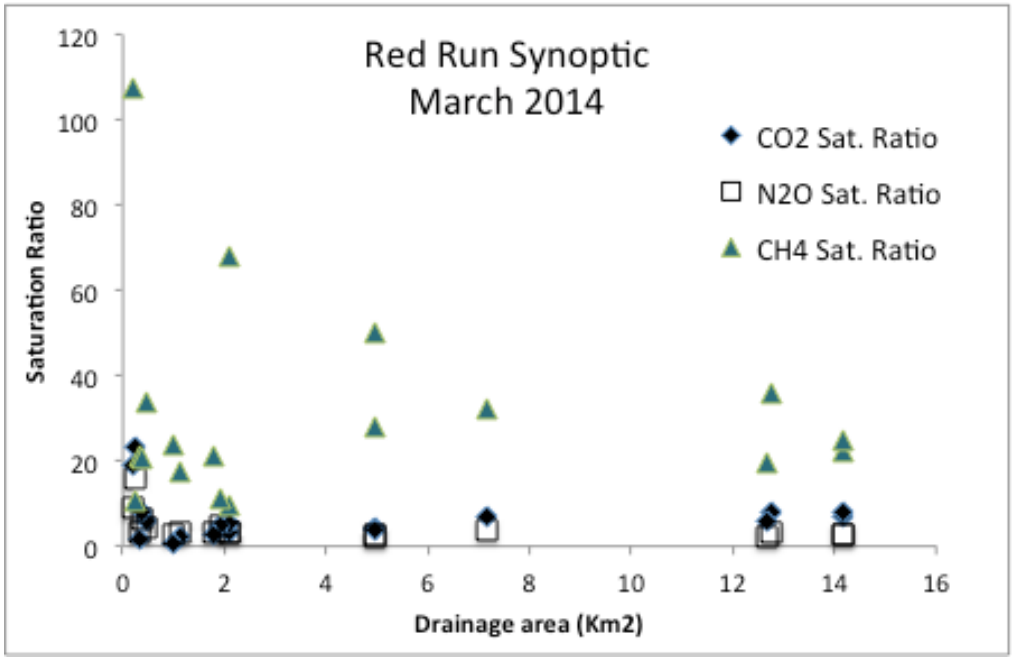


Figure 3. Synoptic sampling results from Red Run in March, 2014. This example of longitudinal sampling data displays the consistently super-saturated conditions for all three GHG's in Red Run.

Photo of R. Smith at one of the old/degraded sites in July, 2014.

Kinetics of the Iodination of Natural Organic Matter and Iodinated Disinfection Byproduct Formation in Drinking Water Treatment (Graduate Fellowship)

Basic Information

Title:	Kinetics of the Iodination of Natural Organic Matter and Iodinated Disinfection Byproduct Formation in Drinking Water Treatment (Graduate Fellowship)
Project Number:	2014MD318B
Start Date:	3/1/2014
End Date:	9/30/2014
Funding Source:	104B
Congressional District:	MD-007
Research Category:	Water Quality
Focus Category:	Toxic Substances, Water Supply, Water Quality
Descriptors:	None
Principal Investigators:	Kaye Lorraine Brubaker

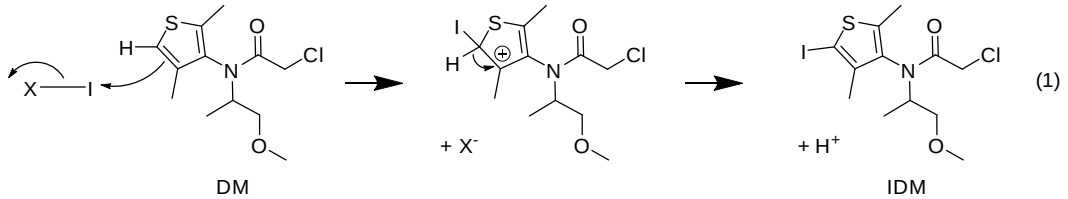
Publications

There are no publications.

Michael Rose
November 15, 2014

As a 2014 Maryland Water Resources Research Center Summer Graduate Fellow, I continued building my foundation to investigate environmental aqueous reactions between iodine and natural organic matter (NOM). My PhD research focuses on toxic iodine-containing organic compounds formed during drinking water treatment, iodinated disinfection byproducts (I-DBP). I-DBP production begins by formation of a bond between iodine and NOM (iodination reaction). For the initial phase of my dissertation, I will investigate NOM iodination kinetics under water treatment conditions utilizing adsorbable organic iodine (AOI) measurements. This summer I developed my experimental technique on the iodination kinetics of dimethenamid (DM), a common herbicide, and searched chemical literature to find an optimal analytical method to quantify iodide (I^-) for AOI measurements. Both endeavors have deepened my appreciation for the complexity of aqueous iodine speciation and reactivity.

Kinetic experimentation is a fundamental tool in physical organic chemistry to quantify reaction rate and elucidate reaction mechanism. Kinetic data facilitates water quality modelling and allows more reliable comparison across studies than arbitrary formation potential tests. DM offers several advantages as a starting point for developing experimental technique: DM is experimentally convenient for halogenation reactions (e.g., no ionizable moieties and only one halogenation product), the data can be compared to prior work in our lab on the chlorination[1] and bromination[2] rates of DM, and the analytical method was readily available from the previous work[1]. The reaction mechanism for DM halogenation is consistent with electrophilic aromatic substitution (EAS, Rxn 1).[1]



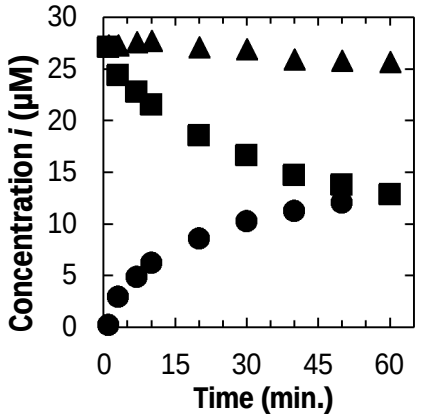


Fig 1. DM iodination time course plot. Square = DM, Circle = IDM, and triangle = mass balance. Solution conditions: pH = 6.0, T = 22°C, [free available chlorine] and [I^-] = 66 mM, $[DM]_0$ = 30 μM, I = 0.1 M.

Figure 1 is an example time course plot produced from a DM iodination kinetic experiment. Samples are collected from the reactor at regular time intervals to analyze DM and the iodination product, iododimethenamid (IDM), by gas chromatography-electron capture detection (GC-ECD). GC-ECD analysis requires an authentic chemical standard for reasonable certainty in analyte identification. The DM analytical standard is commercially available but not for IDM; so, I synthesized it. Initially, I tried the synthesis procedure previously developed for bromo-DM[1], which used an aqueous solution of hypochlorous acid (HOCl) and bromide (Br^-) to generate the brominating agents hypobromous acid (HOBr), aqueous bromine (Br_2), and bromine monochloride (BrCl). The analogous iodination reaction slowed greatly far from completion, despite great excess of equimolar HOCl and I^-, while the bromination reaction proceeded to completion in minutes. Hypoiodous acid (HOI) disproportionation occurs rapidly compared to other hypohalous acids and eventually produces iodate (IO_3^-), a non-reactive sink.[3] HOI disproportionation appears to out-compete iodination in this case, which reinforced the lesson that trends in chlorine and bromine reactivity cannot necessarily be applied to iodine.

Iodination reactions are relatively novel in DBP research but are common in synthetic organic chemistry. My recent Advanced Organic Chemistry class taught me the language and literature to access the information needed to design an effective iodination scheme. An approach[4] using N-iodosuccinimide and catalytic trifluoroacetic acid in acetonitrile worked wonderfully. The synthesized

IDM was characterized by GC-mass spectrometry (MS) and ^{1}H nuclear magnetic resonance (NMR) spectroscopy. The progress in my DM iodination work has increased considerably with the confidence afforded by an analytical standard of IDM. The process of synthesizing IDM taught me valuable lessons about iodine chemistry and chemical synthesis that will aid future work with NOM model compounds.

Researching an optimal aqueous iodine analytical technique was another major effort of summer 2014. The dominant forms of iodine in fresh water are I^-, IO_3^-, and organic iodine which is measured as adsorbable organic iodine (AOI).[5] The AOI method converts organic iodine to I^-. Our lab has a Mitsubishi TOX-100 Total Organic Halogen Analyzer that can be modified to enable AOI measurements. IO_3^- is quantitatively reduced to I^- by a variety of reductants (e.g., metabisulfite[6]). Using sample pretreatment and multiple analytical runs, all target analytes can be measured as I^- and calculated by difference. An ideal analytical method could measure I^- and IO_3^- in a single run for efficiency, but I^- is the crucial species to measure.

Ambient iodine occurs at low concentration in most fresh water. A survey of 36 North American rivers found total iodine concentrations between 0.5 and 212 µg/L (4 and 1,670 nM; detection limit = 0.1 µg/L [1 nM]).[5] Analytical sensitivity of ~1 nM would allow observation of iodine speciation in most fresh waters. Selective detectors mitigate the inevitable interference of more abundant halogens, Cl and Br. The ability to separate analytes in a complex mixture (e.g., chromatography) reduces interference in the detection of trace constituents in complex mixtures, such as natural waters. The basic criteria for an aqueous iodine analytical method are high sensitivity (1 nM), selective detection, and analyte separation capability. Table I provides a summary of the most promising methods our lab is considering.

Table I. Methods of Aqueous Iodine Detection

Ref	Method		I^- Quantitation Range (nM)	
	Separation/Concentration	Detection		
7	Ion Chromatography (IC)	Inductively Coupled Plasma Mass Spectrometry (ICP-MS)	1	394
8	IC	UV-Vis after post-column rxn	1.6	7,880
9	Anion exchange chromatography (AEC)	Pulsed Amperometric Detection (PAD)	0	394
10	High Performance Liquid Chromatography (HPLC)	Ultra-violet/Visible Spectrophotometry (UV-Vis)	0.44	300
11	Gas chromatography (GC) of generated I_2	Electron capture detector (ECD)	5.9	15,760
6	GC after derivitization w/ N,N-dimethylaniline	Mass spectrometry (MS)	0.8	126
12	None	UV-Vis of triangular gold nanoparticles (tAg-NPs)	1	1,001

Our department owns a Perkins Elmer ELAN II ICP-MS system coupled to an HPLC interface, which meets the basic criteria. Unfortunately, the cost per sample is prohibitive for routine analysis due to consumable and maintenance costs. This summer, I conducted preliminary trials of a GC-MS method[6] using N,N-dimethylaniline derivatization. The results are promising, but the method requires considerable sample preparation and the derivatization may be subject to interference in a natural matrix. Our department recently purchased a Thermo/Dionex ICS-2100 IC system, which I look forward to testing. Additional detectors (UV-vis and PAD) could greatly increase sensitivity, making this method attractive due to minimal sample preparation.

I am hopeful to find an ionophoric chemosensor that can be detected by UV-vis absorbance, fluorescence, or ion-selective electrode, similar to ref 12. My Advanced Mechanistic Organic Chemistry class exposed me to recent developments in supramolecular chemistry that may offer a sensitive analytical method at a reasonable cost that could be replicated in any typical wet laboratory. This investment in developing an analytical technique should pay-off throughout my dissertation work and hopefully will help other laboratories investigating aqueous iodine reactions in the environment.

References

1. Sivey, J. D.; McCullough, C. E.; Roberts, A. L., Chlorine Monoxide (Cl2O) and Molecular Chlorine (Cl2) as Active Chlorinating Agents in Reaction of Dimethenamid with Aqueous Free Chlorine. *Environ. Sci. Technol.* **2010,** *44* (9), 3357-3362.
2. Sivey, J. D.; Arey, J. S.; Tentscher, P. R.; Roberts, A. L., Reactivity of BrCl, Br(2), BrOCl, Br(2)O, and HOBr toward dimethenamid in solutions of bromide + aqueous free chlorine. *Environmental science & technology* **2013,** *47* (3), 1330-8.
3. Bichsel, Y.; von Gunten, U., Hypoiodous acid: kinetics of the buffer-catalyzed disproportionation. *Water Res.* **2000,** *34* (12), 3197-3203.
4. Castanet, A.-S.; Colobert, F.; Broutin, P.-E., Mild and regioselective iodination of electron-rich aromatics with N-iodosuccinimide and catalytic trifluoroacetic acid. *Tetrahedron Lett.* **2002,** *43* (29), 5047-5048.
5. Moran, J. E.; Oktay, S. D.; Santschi, P. H., Sources of iodine and iodine 129 in rivers. *Water Resources Research* **2002,** *38* (8).
6. Zhang, S.; Schwehr, K. A.; Ho, Y. F.; Xu, C.; Roberts, K. A.; Kaplan, D. I.; Brinkmeyer, R.; Yeager, C.; Santschi, P. H., A novel approach for the simultaneous determination of iodide, iodate and organo-iodide for ^{127}I and ^{129}I in environmental samples using gas chromatography-mass spectrometry. *Environmental science & technology* **2010,** *44* (23), 9042-9048.
7. Wang, K.-e.; Jiang, S.-J., Determination of iodine and bromine compounds by ion chromatography/dynamic reaction cell inductively coupled plasma mass spectrometry. *Anal. Sci.* **2008,** *24* (4), 509-514.
8. Bichsel, Y.; von Gunten, U., Determination of iodide and iodate by ion chromatography with postcolumn reaction and UV/visible detection. *Analytical chemistry* **1999,** *71* (1), 34-38.
9. Bruggink, C.; van Rossum, W. J.; Spijkerman, E.; van Beelen, E. S., Iodide analysis by anion-exchange chromatography and pulsed amperometric detection in surface water and adsorbable organic iodide. *Journal of chromatography. A* **2007,** *1144* (2), 170-4.
10. Schwehr, K. A.; Santschi, P. H., Sensitive determination of iodine species, including organo-iodine, for freshwater and seawater samples using high performance liquid chromatography and spectrophotometric detection. *Anal. Chim. Acta* **2003,** *482* (1), 59-71.
11. Akhoundzadeh, J.; Chamsaz, M.; Yazdinezhad, S. R.; Arbaz-zavvar, M. H., Head-space single drop microextraction combined with gas chromatography with an electron capture detector for determination of iodine in infant formulas. *Analytical Methods* **2013,** *5* (3), 778.
12. Hou, X.; Chen, S.; Tang, J.; Xiong, Y.; Long, Y., Silver nanoplates-based colorimetric iodide recognition and sensing using sodium thiosulfate as a sensitizer. *Anal Chim Acta* **2014,** *825*, 57-62.

Environmental Concentrations and Exposure Effects of Environmental Gestagens on a Sentinel Teleost Fish

Basic Information

Title:	Environmental Concentrations and Exposure Effects of Environmental Gestagens on a Sentinel Teleost Fish
Project Number:	2014MD321G
USGS Grant Number:	G14AS00014
Start Date:	9/1/2014
End Date:	8/31/2017
Funding Source:	104G
Congressional District:	MD-005
Research Category:	Water Quality
Focus Category:	Water Quality, Toxic Substances, Wastewater
Descriptors:	None
Principal Investigators:	Edward F Orlando, Michael T Meyer, Patrick Phillips

Publications

There are no publications.

Department of the Interior, United States Geological Survey, National Institutes for Water Resources

PROGRESS REPORT

September 1, 2014 – February 28, 2015

1. **Title of Project and Grant ID:** Environmental Concentrations and Exposure Effects of Environmental Gestagens on a Sentinel Teleost Fish (2014MD321G)
2. **Principal Investigator and Organization:** Edward F. Orlando, University of Maryland College Park
3. **Date of Report:** June 12, 2015
4. **Overview of Progress.**

   In the first six months of our study, we have completed (a) setting up the fish exposure system, (b) testing the exposure system and capability of our lab group by doing an ethanol exposure, (c) executing our first gestagen exposure experiment, and working out gestagen analysis methods. We have interesting data from both the ethanol solvent exposure and the first gestagen exposure experiments. We are off to a strong start, and I am pleased with our progress.

5. **Project Hypothesis, Objectives, and Progress Update.**

   A. *Summarize the hypothesis and objectives of your project.*

   We hypothesize that gestagen exposure will negatively affect the reproductive health of fathead minnows and that progesterone and at least some of the progestins will be measured in ng/L (biologically active concentrations) in environmental samples from the Southeast Stream Quality Analysis (SESQA) study.

   Objective 1: We will assess the exposure effects of gestagens on fathead minnow reproductive health using a combination of behavioral and physiological endpoints to determine the most effective gestagen and the most sensitive endpoints.

   Objective 2: We will augment an existing 41 hormone method, which already includes progesterone and one progestin norethindrone, developed by Dr. Mike Meyer (Co-PI), with three additional progestins that are not in the method but that are required for this study: levonorgestrel, gestodene, and drospireone.

   Objective 3: We will determine the concentration of gestagens in water, POCIS, and sediment samples from the SESQA study sites. Results from this objective will provide the first comprehensive examination of the occurrence of environmental gestagens in stream systems in the United States. We will also quantify concentration of gestagens in the lab exposure studies of Objectives 1 and 4.

   Objective 4: We will integrate the findings from Objective 1-3 to determine environmentally relevant exposure concentrations for individual gestagens and also test one gestagen mixture to verify and compliment the results of the earlier objectives, and to initiate the identification of biomarkers of gestagen exposure, which currently are not known.

   B. *Which objectives have you completed to date?*

   We have substantially begun Objectives 1 and 2 (please see preliminary results below). Results and monthly progress is discussed in a group conference call on the 2nd Tuesday of each month with Drs. Michael Meyer (co-PI), Dana Kolpin (USGS, Lawrence, KS), and David Alvarez (USGS, Columbia, MO).

C. What work is planned during the next 6-12 months?

We will complete the first and second objectives and make substantive progress on the third objective. We will present preliminary results at the North American Conference of Comparative Endocrinology, Ottawa Canada, June 21-25, 2015. We will have our first paper in review from the research supported by this grant by the end of first year and will be working on a second manuscript. Our first face-to-face group meeting (same individuals as in 5B. above) will occur at the Society of Environmental Toxicology and Chemistry annual meeting, Salt Lake City, UT, November 1-5, 2015.

6. Is there anything the DOI/USGS/NIWR needs to be notified of relevant to the study's progress?

Progress is on target or greater than our projected schedule.

7. Preliminary results.

Dr. Mike Meyer has initial results suggesting that he will have no difficulty measuring the gestagens in this project at the required ng/L concentrations. He is currently working on maximizing sensitivity of the assays and validating those methods.

In the Orlando Lab's first experiment, we have tested the effects of different concentrations of ethanol (0, 0.001, 0.0001, and 0.00001% EtOH) on the reproductive behavior and fecundity of fathead minnows. We find no published papers that have systematically studied the effects of ethanol on these ecologically critical endpoints in fish. This was very surprising to us. We chose our ethanol concentrations based on the literature and made sure we were at or below the concentrations used by others. We found a negative impact on numbers of eggs produced by fish at the two highest concentrations with no difference seen in the 0.00001% EtOH and water only control.

In our second experiment, we exposed fish to water only, water plus 0.0000095% EtOH, 10 ng/L, and 100 ng/L gestodene (in 0.0000095% EtOH). Gestodene is a synthetic gestagen or progestin that is used in oral contraceptives. In previous research by my lab, we found that gestodene did not activate the *nuclear progesterone receptor* but was a strong activator of the *nuclear androgen receptor* of fathead minnows (doi: 10.1021/es501428u). In the current study, gestodene exposure of reproductive adult fathead minnows was associated with a dramatic decrease in egg production (fecundity) and masculinization of secondary sex characteristics (male phenotype including nuptial tubercles and finspot, induced in female fish). Preliminary analysis suggests that behavior of both females and males is altered and may be contributing to the decrease in spawned eggs. *The importance of these data is that decreased egg production and altered reproductive behavior have strong links to effects on population (recruitment to the next generation) and, by extension, aquatic ecosystem health.*

Funding from the DOI/USGS/NIWR is enabling the training of one PhD student and approximately six undergraduate research assistants. Two of these undergraduates are considering research careers in part because of this experience.

Information Transfer Program Introduction

The Center's major Information Transfer effort (beyond publications and presentations by the PIs themselves) is the annual Maryland Water Symposium. This year's Symposium was the 12th such event. The Center is also making an effort to reach out to broader audiences with educational events.

Maryland Water 2014 - Symposium

Basic Information

Title:	Maryland Water 2014 - Symposium
Project Number:	2014MD319B
Start Date:	3/1/2014
End Date:	2/28/2015
Funding Source:	104B
Congressional District:	MD-005
Research Category:	Not Applicable
Focus Category:	None, None, None
Descriptors:	None
Principal Investigators:	Kaye Lorraine Brubaker

Publications

There are no publications.

Maryland Water Resources Symposium 2014

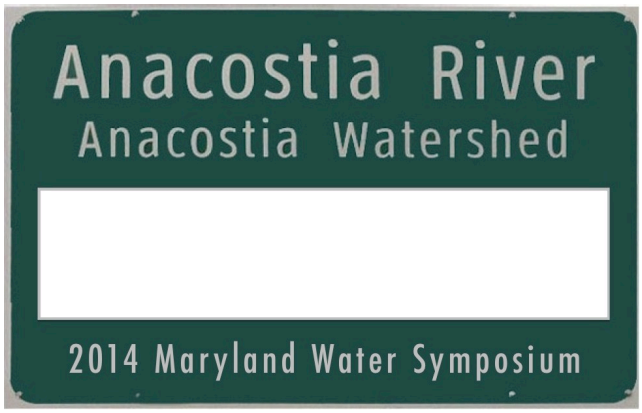
The Maryland Water Resources Research Center hosted "Anacostia: River and Watershed," the Maryland Water 2014 Symposium on Nov. 20, 2014. This was the 12th one-day symposium hosted by the Center.

The Anacostia River flows into the Potomac River in the Nation's Capital; its watershed (176 sq mi) lies in Montgomery and Prince George's County, Maryland, and Washington, D.C. Since European settlement, the watershed has been transformed by cultivation and urbanization, and the river's water quality has been severely degraded. Governments and citizens have joined to preserve and restore the chemical, physical and biological integrity of the river. This one-day symposium reviewed the Anacostia's history, the forces that have shaped it, the community's investment in its revival, and current research in, on, and about the river and its watershed.

The day's agenda is included in this report. Over 70 attendees signed in; however, an unspecified number of participants did not sign in (estimated 20). Participants included University research faculty, graduate and undergraduate students, agency scientists (including a good USGS representation), and community members. Our keynote speaker was the president of the Anacostia Watershed Society (AWS), and the pre-symposiu outreach focused on the AWS community.

A novel addition to this year's Symposium was the inclusion of two walking tours following the conclusion of the research presentations. Dr. Karen Prestegaard (Geology, University of Maryland, College Park) led a tour to reasonably intact headwater wetlands located in the forested area of the UM campus, adjacent to the University Golf Course. Dr. Allen Davis (Civil & Environmental Engineering, University of Maryland, College Park) toured Low Impact Development / Storm Water Management installations on the UMCP campus.

Responses indicate that this Symposium was successful in reaching new audiences for MWRRC: undergraduate students and non-University-affiliated neighbors.



Symposium nametag inspired by local road signs identifying Anacostia tributaries.

The Anacostia
River & Watershed
2014 Maryland Water Resources Symposium

Thursday, Nov. 20, 2014
8:30 a.m. – 3:30 p.m.

Adele H. Stamp Student Union
Charles Carroll Room
University of Maryland, College Park

8:30 a.m.	**Gathering and Refreshments**
9:00 a.m.	**Welcome and Introduction** Kaye L. Brubaker, Director, Maryland Water Resources Research Center
9:10 a.m.	**Then, Now, and Beyond: Celebrating 25 Years of the Anacostia Watershed Society (Keynote)** James Foster, President, Anacostia Watershed Society
9:50 a.m.	**Reconstructing the Pre-Urban Hydrology of the Anacostia River** Mallori McDowell, University of Maryland, College Park & U.S. Geological Survey
10:20 a.m.	***Break***
10:30 a.m.	**Anacostia Geomorphology: Water, Soil, Plants, Sediments** Karen Prestegaard, University of Maryland, College Park
11:00 a.m.	**Dredging in the Anacostia River** Robert Blama, U.S. Army Corps of Engineers, Baltimore District
11:30 a.m.	**Locating Sources of Pollutants in the Anacostia River using Active Biomonitoring with *Corbicula fluminea*** Harriette Phelps, University of the District of Columbia
12 noon	***Lunch Break***
1:00 p.m.	**Water Quality in the Anacostia - How Are We Doing?** Joseph Bell, U.S. Geological Survey
1:30 p.m.	**Novel Stormwater Management in the Anacostia Watershed** Allen Davis, University of Maryland, College Park
2:00 p.m	**Adjourn; Depart for Walking Tours**

Sponsored by

MARYLAND Water Resources Research Center

USGS science for a changing world

www.waterresources.umd.edu

Maryland Water Issues - Educational Event

Basic Information

Title:	Maryland Water Issues - Educational Event
Project Number:	2014MD320B
Start Date:	3/1/2014
End Date:	2/29/2016
Funding Source:	104B
Congressional District:	MD-001
Research Category:	Not Applicable
Focus Category:	Education, None, None
Descriptors:	None
Principal Investigators:	Kaye Lorraine Brubaker

Publications

There are no publications.

2014 Maryland Water Education Event: Progress Report

The 2014 proposal envisioned an educational/outreach event on Sea Level Rise to be held on the Eastern Shore of Maryland (e.g., Salisbury). This event will include screening of a recent independent documentary film about coastal communities and sea level rise, and an expert speaker or panel of experts.

The event was not scheduled during 2014-15; it has been postponed to the 2015-16 academic year. The Director attended an event "The Future Is Now: Dealing with Rising Seas on Maryland's Shores," sponsored by the League of Women Voters and the Center for Leadership in Environmental Education (CLEEn) at Chesapeake College in Wye Mills, Md., on the Eastern Shore on Feb. 21, 2015. At that event, Dr. Brubaker made connections with partners including CLEEn and the Eastern Shore Land Conservancy, with whom she plans to work to organize the proposed MWRRC Eastern Shore educational event.

USGS Summer Intern Program

None.

Student Support					
Category	**Section 104 Base Grant**	**Section 104 NCGP Award**	**NIWR-USGS Internship**	**Supplemental Awards**	**Total**
Undergraduate	23	6	0	0	29
Masters	6	0	0	0	6
Ph.D.	5	1	0	0	6
Post-Doc.	0	0	0	0	0
Total	34	7	0	0	41

Notable Awards and Achievements

Two PIs at the University of Maryland leveraged their 104(B) projects to include undergraduate students through linkages with established curricular structures. A. Baldwin (2014MD314B) partially supported a team of 7 students to develop a two-semester senior Capstone project related to the research. L. Yonkos (2014MD315B) partially supported a Gemstone Undergraduate Research Team comprised of eight (8) undergraduate students from various University of Maryland academic departments; they investigated poultry litter-induced intersex in largemouth bass by exposing batches of fish at various ages to aqueous poultry litter mixtures before growing to maturity for assessment of testis pathology.

Alex MacLeod (M.S. student, 2014MD315B) was awarded a $5,000 Dean's Fellowship from the College of Agriculture and Natural Resources, University of Maryland, College Park, largely in recognition of the quality of his laparoscopy validation research project.

The sensor deployment originally proposed in MD2014313B is being built upon through funds applied for, and received from, the School of Emerging Technology at Towson University. The $31,725 in funds was used to purchase additional conductivity and pressure sensors as well as Isco instruments for measuring pipe flow in culverts. PI Joel Moore is collaborating with Michael McGuire, a computer scientist specializing in spatio-temporal data mining algorithms, on this project. Additionally, a computer science graduate student is being supported by the Towson University funds in the summer of 2015 to apply spatio-temporal data mining techniques and develop ways for visualizing the sensor datasets collected as part of the work supported by this grant.

Joel Moore (MD2014313B) applied for, and was awarded, a three-year endowed chair position awarded to pre-tenure faculty (Jess and Mildred Fisher Endowed Chair of Geological Sciences) at Towson University in part to expand on the work originally proposed in the MWRRC grant.

The work supported by 2014MD313B has been well received in Maryland and beyond. Dr. Moore gave the following talks related to this research:

- Research seminars
  - Sept. 2014: A high salt diet: How stormwater management ponds affect road salt impacts on soils, groundwater, and streams. Department of Geology Seminar, Kent State University.
  - Dec. 2014: Groundwater that looks like seawater: Stormwater management basins & road salt loading into suburban watersheds. Maryland-DC-Delaware Water Science Center, US Geological Survey.
  - Mar. 2015: Groundwater that looks like seawater: Stormwater management basins & road salt loading into suburban watersheds. Department of Geography & Environmental Planning, Towson University.
- Talks to local and regional governmental groups
  - Mar. 2015: Groundwater that looks like seawater: stormwater management basins & road salt loading into suburban watersheds. Department of Environmental Protection & Sustainability, Baltimore County, MD.
  - Apr. 2015: Effects of road salt on dissolved sodium and chloride in suburban Baltimore groundwater and streams. Reservoir Technical Group, Baltimore Metropolitan Council.

Additionally, in part related to the work supported by this proposal, Dr. Moore was nominated and elected to serve a three-year term on the Maryland Water Monitoring Council (starting January 2015).

Publications from Prior Years

1. 2009MD193B ("Low-cost Anaerobic Digesters for Dairy Manure Treatment and Renewable Energy Production") - Articles in Refereed Scientific Journals - Lansing, S.A., K.H. Klavon, W.W. Mulbry, and A.R. Moss, 2015. Design And Validation Of Field-Scale Anaerobic Digesters Treating Dairy Manure For Small Farms, Transactions of the ASABE 52(2):441-449.
2. 2010MD229B ("Occupational and Community Exposure to Antimicrobial-Resistant Bacteria and Antimicrobials Present in Reclaimed Wastewater ") - Articles in Refereed Scientific Journals - Goldstein, R.E.R., S.A. Micallef, S.G. Gibbs, X. He, A. George, A. Sapkota, S.W. Joseph, and A.R. Sapkota, 2014, Occupational Exposure to Staphylococcus aureus and Enterococcus spp. among Spray Irrigation Workers Using Reclaimed Water, International Journal Of Environmental Research And Public Health 11(4):4340-4355, DOI 10.3390/ijerph110404340.
3. 2009MD193B ("Low-cost Anaerobic Digesters for Dairy Manure Treatment and Renewable Energy Production") - Articles in Refereed Scientific Journals - Moss, A.R., S.A. Lansing, D.R. Tilley, K.H. Klavon, 2014, Assessing the sustainability of small-scale anaerobic digestion systems with the introduction of the emergy efficiency index (EEI) and adjusted yield ratio (AYR), Ecological Engineering 64:391-407, DOI 10.1016/j.ecoleng.2013.12.008.
4. 2012MD262B ("Quantifying remobilization rates of legacy sediment from Maryland Piedmont floodplains") - Articles in Refereed Scientific Journals - Donovan, M., A. Miller, M. Baker, and A. Gellis, 2015, Sediment contributions from floodplains and legacy sediments to Piedmont streams of Baltimore County, Maryland, Geomorphology 235:85-105, DOI 10.1016/j.geomorph.2015.01.025.
5. 2013MD304B ("Bromide as a Precursor of Potent Brominating Agents During Drinking Water Chlorination") - Articles in Refereed Scientific Journals - Sivey, J.D., M.A. Bickley, and D.A. Victor, 2015, Contributions of BrCl, Br-2, BrOCl, Br2O, and HOBr to Regiospecific Bromination Rates of Anisole and Bromoanisoles in Aqueous Solution, Environmental Science & Technology 49(8):4937-4945, DOI 10.1021/acs.est.5b00205.