SAVANNAH RIVER ECOLOGY LABORATORY

ANNUAL TECHNICAL PROGRESS REPORT OF ECOLOGICAL RESEARCH FOR FY19

Final Report: Submitted January, 2020

Supported under Cooperative Agreement DE-EM0004391

between The University of Georgia *and* The U.S. Department of Energy *for the period of* 1 October 2018 – 30 September 2019

> Dr. Olin E. Rhodes, Jr. Director

Prepared by Savannah River Ecology Laboratory P. O. Drawer E Aiken, SC 29802 This report is provided for information only and is not to be considered formally published literature. We request that no citations be made of information contained herein without the express consent of the investigator.

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TASK 6.	SREL will serve as the point-of-contact for the "DOE Research Set-Aside" areas that are protected from site impacts so that they are available for environmental research and can serve to establish representative standards for comparison to impacted areas on the SRS. Currently SRS has 30 'set-aside" areas. SREL will also continue to promote the role of the SRS as a National Environmental Research Park
TASK 7.	Through general research and public outreach programs, SREL will increase scientific understanding in the general areas of environmental characterization, ecological risk assessment, and environmental remediation and restoration. This will require research on topics such as terrestrial and aquatic ecology, environmental chemistry, molecular ecology and genetics, microbial ecology, radiation ecology, and ecotoxicology. SREL will also continue to communicate and coordinate with SRS contractors and the public on these issues
TASK 8.	SREL will continue to serve as a regional resource for scientific expertise and environmental research. SREL staff scientists will continue to provide special technical assistance to other site contractors, area stakeholders, other researchers, and the public. SREL will also continue to collaborate with scientists from other institutions

- TASK 11. The participant will operate and maintain the Savannah River Ecology Laboratory (SREL)

 facilities on the Savannah River Site (SRS) to efficiently and successfully perform the

 research, education and outreach programs described in this project description (See Appendix

 A of the Cooperative Agreement for List of Facilities).

TASK 12. <u>U</u>	niversity of Georgia Research Foundation (UGARF) will be responsible for management
ar	nd engineering services for the planning, design, and construction of approved projects as
<u>m</u>	ay be required to repair, modify or upgrade existing facilities or construct new facilities, not
<u>to</u>	include line item projects, necessary to support the University of Georgia Research
Fo	oundation (UGARF) scope of work, as approved by the Contracting Officer and appropriate
D	OE program personnel. Funding for major repairs and new construction will be provided
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SECTION I: Savannah River Ecology Laboratory – FY19 Overview of Achievements

The Savannah River Ecology Laboratory (SREL) is a research unit of The University of Georgia (UGA). SREL has been conducting ecological research on the Savannah River Site (SRS) near Aiken, South Carolina for over 68 years. The overall mission of the laboratory is to enhance our understanding of the environment by acquiring and communicating knowledge of ecological processes and principles that contribute to sound environmental stewardship. In addition, as directed in the Cooperative Agreement with the U.S. Department of Energy (DOE), SREL will provide the public with an independent evaluation of the ecological effects of SRS operations on the environment. Toward these goals, SREL conducts fundamental and applied ecological research, as well as education and outreach programs.

The laboratory's research mission during FY19 was fulfilled with the publication of 95 journal articles, proceedings articles, and book chapters by faculty, technical staff, students, and visiting scientists. One book was also authored by SREL faculty and staff. Additional journal articles and books have been submitted or are in press. Significantly, SREL conducted over 541 outreach events reaching over 63,000 people of all ages. Other noteworthy events took place as faculty members, staff, and graduate students received awards for the quality of their research. These are described in Section IX *Special Accomplishments*.

The vision, structure, and operations of SREL continue to evolve since changes in funding structure were instituted in FY07. However, the five-year Cooperative Agreement between the University of Georgia's Research Foundation and the Department of Energy for support of the Savannah River Ecology Laboratory has allowed funding from the DOE and other SRS tenants to fund SREL to meet the specific needs of DOE Environmental Management (EM) and DOE National Nuclear Safety Administration (NNSA) on the Savannah River Site. The current funding model for SREL is entrepreneurial and interdisciplinary, and seeks to pursue funding strategies that are competitive, responsive to sponsors' requirements, and based on a diverse and sustainable foundation. This model has required restructuring of research and supporting infrastructure at the laboratory.

Today, a leaner, but robust SREL presence continues to operate on the SRS. SREL's total employment in FY19 was approximately 167 faculty, technicians, students, and support staff. Although the number of employees and level of funding has been reduced relative to a decade ago, SREL continues progress toward stated objectives and does not compromise safety and security. New and continuing partnerships and collaborations with the Athens campus (Warnell School of Forestry and Natural Resources, UGA Complex Carbohydrates Center, Odum School of Ecology, College of Public Health, College of Agriculture and Environmental Sciences), other universities (University of South Carolina – Aiken, University of South Carolina – Upstate, Georgia Reagents University) and other agencies (US Department of Agriculture, US Army Corps of Engineers, US Department of Defense, Federal Aviation Administration) continue to be explored and developed in order to maximize the use of SREL assets. Graduate student programs have continued with funding provided by DOE, external grants, UGA, or the student's host university.

During FY19, DOE-SR funding was leveraged to acquire approximately 1 million dollars in salary and infrastructure investments from the University of Georgia, in addition to the 20% cost share negotiated under the terms of UGA's Cooperative Agreement with DOE. DOE funding also is being used to leverage cost-shared faculty positions with UGA units on the main campus, resulting in three tenure track faculty lines at SREL and a portion of three tenure track faculty lines on the main UGA campus that will contribute to the SREL mission on the SRS during the coming years.

SREL faculty have responded to the revised funding structure for the laboratory and have sought financial support from multiple external funding agencies, DOE-EM, DOE-NNSA, Savannah River Nuclear

Solutions-Area Closure Projects (SRNS-ACP), Savannah River Remediation (SRR) and UGA. In addition, DOE-EM has provided additional infrastructure support to SREL to help revitalize aging facilities and meet safety standards for our working environment. Establishment of a new Cooperative Agreement with DOE allows SREL/UGA access to the SRS through 30 September 2021. The SREL continues to work closely with local community groups, local schools, and other area stakeholders on a number of research, environmental monitoring, education, and outreach activities.

During FY19, SREL has continued to optimize its research programs to address DOE and SRS concerns, maintain staff in critical research disciplines, and attract new personnel. SREL researchers are vigorously pursuing additional funding sources to leverage existing research funds, while continuing to focus the laboratory's research efforts on projects of interest to the SRS. In addition, personnel from SREL have been actively engaged in furthering DOE's SRS missions in Radioecology, Environmental Stewardship, Next Generation Cleanup Technologies and Renewable Energy.

Researchers at SREL received funding from 48 new and continuing external grants during FY19. Sources of grant awards range from private foundations to federal and state agencies, including the US Department of Interior, the US Department of Agriculture, the National Science Foundation, and the Department of Defense.

SREL faculty members hold positions in various departments at UGA. Several SREL faculty members (and emeritus faculty) have adjunct status at other colleges and universities. Faculty, staff, and students are active in providing outreach and service to the scientific community. Representatives from SREL hold editorial or committee positions in national groups and organizations and serve on several UGA academic and administrative committees. SREL faculty members continue to make scientific presentations, contribute posters to scientific meetings, and present seminars at colleges and universities.

SREL's Education Program continues to be successful with SREL faculty and staff mentoring over 87 graduate students from numerous colleges and universities in the United States during FY19.

The SREL Outreach Program communicates scientific awareness to area schools and the general public, an audience which differs significantly from science professionals. During the past year, SREL presented over 442 talks, 36 tours, 34 exhibits, and 36 *Ecologist for a Day* programs reaching a total of over 63,000 people. Topics for these presentations included ecological studies of reptiles and amphibians, southeastern plants and habitats, long-term research, safety, biodiversity, local wetlands and watersheds, conservation, and careers in ecology and research. In the past year, SREL participates by providing presentations on the history and research of the lab as well as a "show and tell" session featuring research animals native to the SRS.

Continuing in FY19, SREL's DOE-funded Radiological Education, Monitoring and Outreach Program has been focused on increasing public awareness and education on the topic of radiation in the environment and radiological risks to the general public. The outreach coordinator for this program has been providing monthly talks in the local Waynesboro, GA community as well as ad hoc presentations to a variety of local audiences. This program began to conduct limited environmental monitoring in FY19 and will begin providing presentations to the local community later in FY20 using these data to explain and inform the public about monitoring programs that currently provide data to the region.

The UGA Conference Center continues to be a valuable asset to SREL and other entities on the SRS. SREL used the facility to host numerous meetings and environmental education programs for students, teachers, and other organizations this past year. The facility is also used by DOE, the USDA Forest Service, and other site tenants when available.

In summary, it is important to note that the important roles that SREL plays on the SRS unfold prominently in several strategic areas. Such efforts by SREL staff play a critical role in helping the DOE and other SRS tenant organizations reduce costs and continue with their missions on the SRS by assisting them to maintain regulatory compliance, validating remediation efforts, providing basic research for the development of new technologies, promoting sound environmental stewardship of natural resources on the SRS, serving as an independent source of scientific expertise for reviews of technical data and monitoring programs, educating the next generation of radioecologists and nuclear biogeochemists, and conducting outreach efforts to educate local communities about the SRS, its missions, and environmental health. For example, as a critical source of scientific expertise for the Department of Energy on the SRS, SREL provides state of the art scientific support to both DOE-EM and DOE-NNSA. Examples include research on biogeochemical cycling and biological impacts of copper associated with the H-02 mitigation wetlands that provide data needed to validate regulatory compliance for the DOE-NNSA's Tritium mission on the SRS, research on the ecological impacts and potential options for recovery of function of the U-8 stream drainage associated with DOE-NNSA's construction efforts for the MOX fabrication facility on the SRS, research on the organismal effects (proteome and glycome) to aquatic organisms exposed to chronic levels of low dose radiation, research into the extent and impact of biovectors of contaminants on the SRS, and development of strategic management plans for Set Asides on the SRS to maintain the SRS designation as DOE's first National Environmental Research Park.

SREL also serves as a source of critical scientific expertise for other SRS tenant organizations, providing analytical and ecological expertise to assist these organizations with issues ranging from regulatory compliance to creative new technologies for remediation of contaminants on the SRS. Examples include research conducted in support of the SRR mission on the SRS to provide accurate and precise data for use in parameterization of models employed to comply with NRC requirements concerning long-term contaminant exposure risk from stored nuclear materials, research conducted in support of SRR and SRNS to assess biological exposure risks from leakage events involving stored nuclear materials into soil and groundwater, research on the design and efficiency of strategies for bioremediation of tritium in SRS groundwater for SRNS, and research conducted in support of SRNS ACP's remediation mission to assess radionuclide and heavy metal accumulation in long-lived vertebrates and game species to inform regulatory compliance and risk assessment issues associated with ecosystem and human health.

As a source of regional and national scientific expertise, SREL scientists attract external funding to conduct research that not only contributes to areas of national research priority, but also helps contribute to DOE's nuclear and environmental missions on the site. Examples include research on development of technologies for control of feral swine (funded by the US Department of Agriculture), which makes the SRS both a national focal point for feral swine research as well as a recipient of the technologies and strategies as they are developed, research on conservation and management of threatened and endangered species across the nation (funded by the US Department of Defense and the US Fish and Wildlife Service), which contributes to the strategies and tools available for environmental stewardship applications on the SRS, research on avian dispersal technologies (funded by the Federal Aviation Administration and the City of Augusta), which contributes to the ability of SRS tenants to manage nuisance wildlife populations, research on scavenging ecology (funded by the US Department of Agriculture and the Department of Defense), which provides insights into the role of scavengers in recycling of energy and contaminants in the environment and the potential transfer of contaminants off of SRS, and research on risk assessment models for heavy metals in avian species (funded by the US Army Corps of Engineers), which adds to the set of risk assessment modeling expertise available for study of fauna on the SRS.

SECTION II. Cooperative Agreement Key Tasks

TASK 1. SREL will assess the impact of Site operations on the environment, and will continue to provide the public and DOE with an independent view of the environmental management of the SRS

Through a Cooperative Agreement between the Department of Energy and the University of Georgia Research Foundation, SREL provides an independent evaluation of the ecological effects of SRS operations through a program of ecological research, education, and public outreach. This program involves basic and applied environmental research, with emphasis upon expanding the understanding of ecological processes and principles, and upon evaluating the impacts of industrial and land use activities on the environment.

This is accomplished through a broad-based program of field and laboratory research conducted on the SRS and published in the peer-reviewed scientific literature; by providing education and research training for undergraduate and graduate students from colleges and universities throughout the United States and abroad; and by engaging in community outreach activities and service to professional organizations.

The quality of research conducted by SREL scientists is facilitated by their unique expertise in environmental sciences and ecology, the unparalleled field research opportunities at the SRS, and the long-term data sets, research tools, and capabilities that SREL has developed over the last 65 years.

The FY19 SREL research plan can be divided into three critical research areas:

Environmental Characterization

Characterization is a necessary first step in determining environmental and health risks and in devising appropriate remediation and restoration strategies. Environmental information is also needed to make informed decisions about long-term stewardship and land management, and is a critical component of NEPA (National Environmental Policy Act) reports, Records of Decision (ROD), and other regulatory documents. Environmental characterization is more than simply measuring contaminant concentrations in biota or other media, or reporting the presence of organisms at various locations. It includes developing an understanding of the processes that control distributions of contaminants, chemical forms, and their bioavailability. Characterization is also necessary to construct models of how natural and engineered systems function, both in the presence and absence of environmental contamination.

Ecological Risks and Effects

Estimated risks and effects determine the need for remediation and restoration efforts, while perceived risks and effects determine the public's acceptance and support of DOE policies and actions. Estimating ecological risks and effects on the basis of sound science helps to ensure that good decisions are made by reducing uncertainties associated with complex environmental processes. A 1999 report from the National Academy of Sciences stated that *"Ecological risks are better characterized at the Savannah River Site than at any other DOE installation, due in part to the designation of the site as a National Environmental Research Park and the presence of the Savannah River Ecology Laboratory."*

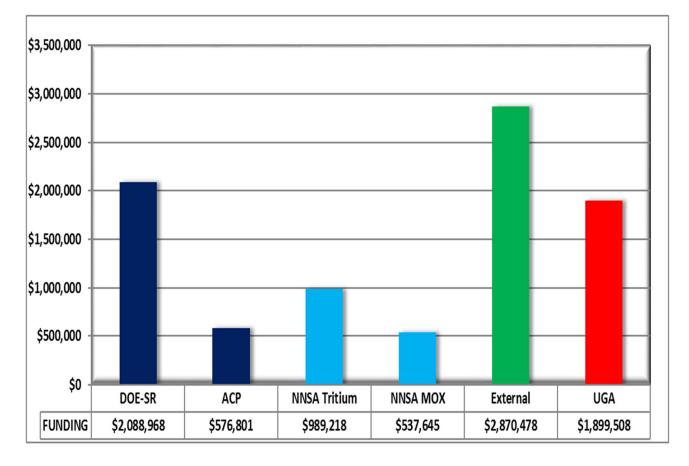
Remediation and Restoration

The knowledge and expertise at SREL are ideally suited to address the remediation and restoration of large land areas contaminated with relatively low levels of metals, organics, and radionuclides. SREL conducts multidisciplinary research designed to assist in the development, evaluation and stakeholder acceptance of remediation and restoration efforts that protect human and ecosystem health. Fundamental to the success of various bioremediation, natural attenuation, and *in situ* remediation applications is an understanding of the underlying scientific principles on which they are based.

TASK 2. <u>SREL will continue basic and applied environmental research with emphasis upon expanding</u> the understanding of ecological processes and principles, and upon evaluating the impacts of site activities, new mission, and land use practices on the environment

In FY19, the Savannah River Ecology Laboratory received approximately 9 million dollars in funding from a variety of sources (Figure 2.1). These funds supported approximately 169 faculty, staff, and students conducting basic and applied environmental research for at least some portion of FY19 (Table 2.2). In total, University of Georgia funding (both direct and indirect funds returned to the lab) and external dollars received from non-SRS sources were responsible for approximately 52% of the laboratories budget. Laboratory personnel were productive and successful in attracting external funding to the site, and very active in graduate student education and service to their communities and professions (Table 2.2). SREL continues to be a productive, independent partner to DOE on the SRS and an excellent value to both stakeholders on the SRS and taxpayers.

Figure 2.1. Overview of funding received by SREL in FY19. Acronyms are as follows: University of Georgia (UGA), Savannah River Site Office of Department of Energy (DOE-SR), all combined sources of funding received from sources external to the Savannah River Site (External), Department of Energy National Nuclear Security Administration's Mixed Oxide Fuel Production Facility (NNSA-MOX), Department of Energy National Nuclear Security Administration's Tritium Facility (NNSA-Tritium), and Savannah River Nuclear Solutions Area Closures Project (ACP).



FY19 SREL FUNDING

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Table 2.2. SREL organizational structure for FY19. This table includes all research faculty, classified staff and Emeritus faculty in residence at the Savannah River Ecology Laboratory for any portion of the FY19 fiscal year.

SREL ORGANIZATIONAL CHART – FY19					
Director Dr. Olin E. Rhodes, Jr.					
Assistant Director Research	Assistant Director Budget and Facilities				
Dr. T. DeVault	C. McBride				
<u>Research Faculty</u>	<u>Safety</u>				
Dr. J. Seaman	D. Mosser				
Dr. S. Lance	R. Beasley				
Dr. Xiaoyu Xu	<u>Computer Service and GIS Lab Manager</u>				
Dr. T. Tuberville	W. Taylor				
Dr. G. Dharmarajan	Property Management				
Tenure Track Faculty	B. Morton				
Dr. J. Beasley	Outreach Program Staff				
Dr. D. Aubrey	P. Perea				
Dr. J. Martin	V. Sutton-Jackson				
Dr. K. Capps	H. DeVault				
Dr. B. Parrott	M. Winzler				
Emeritus Faculty in Residence	J. Green-McLeod				
Dr. D. Adriano	S. Poppy				
Dr. I. Brisbin, Jr.	A. Tucker				
Dr. J.W. Gibbons	A. Hurst				
Dr. K. McLeod	N. Herrington				
Post Docs	Research and Facilities Technical Services				
Dr. F. Coutelot	R. Christie D. Kling				
Dr. X. Xu	M. Edwards M. Squires				
Dr. A. Ferreia	C. Cooper P. Carroll				
<u>Research Prof/Asst/Tech</u>	D. Fraser				
Dr. K. Buhlmann	Administrative Services				
A. Bryan M. Mason	L. LopezdeVictoria				
D. Fletcher L. Lee	M. Roberts				
R. Kennamer D. Scott	J. Scott-Phillips				
A. Lindell P. Stankus	C. Summer				
A. Riggs M. Hamilton	M. Wilburn				
C. Fulghum M. Baker	V. Taylor				
A. Rakowski M. Shapiro	M. Wead				
K. McCallie E. Peck	Temp. Research Technicians				
J. Lott E. Spivey	40 Temp. Techs were employed at SREL				
K. Wilms M. Strassburg	Graduate Students				
T. Mills J. Helton	56 Graduate Students advised by SREL faculty				
J. Obryhim M. Chapman					
F. Toledo					
	(As of 10/1/2019)* Excludes Students				

Peer Reviewed Journal ArticlesBook and Book ChaptersProceedings ArticlesPrimer or Other Scientific NotesNon-Peer reviewed ArticlesArticles In PressArticles In ReviewPeer Review of Manuscripts ConductedExternal Funding (non-SRS)External Grants Submitted as PI or CoPIExternal Grant Funding Submitted as PI or CoPI\$16,	<u>otal</u> 84 4
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External Grants Funded as PL or CoPI ⁺	300,749
	48
	129,536
	otal
MS Graduate Students Chaired	41
•	11
PhD Graduate Students Chaired	15
PhD Graduate Students Completed	1
Graduate Student Committee Memberships	54
Graduate Students Hosted at SREL	8
Post Docs Supervised	5
Presentations T	otal
Invited Presentations	40
Professional Oral Presentations	93
Professional Poster Presentations	57
Extension Presentations	26
Extension Publications	6
Other T	7-4-1
Awards or Honors	otal
Professional Society Committee Memberships	22
K A	
Technical Research Consultations	22

Table 2.3. Summary of professional activities and accomplishments by Savannah River EcologyLaboratory research faculty, research professionals, postdocs and students in FY19.

 1 – includes new grants and contracts, renewals and continuations associated with funding sources external to DOE. Total includes multi-year funding commitments received in FY19 and to be received in future fiscal years.

TASK 3. <u>SREL will use the information collected in the environmental research to develop and test</u> hypotheses that will contribute to the scientific foundation necessary to conduct meaningful ecological risk assessments and to understand the environmental consequences of energy technologies, remediation efforts, and other SRS activities

In FY19 SREL faculty, staff, and students conducted and completed a diversity of environmental research projects on the SRS in support of the missions of SRNS, SRR, and DOE-SR, specifically in the areas of risk assessment and elucidation of the environmental consequences of energy technologies, legacy contamination, and remediation activities on the SRS. Much of this work was funded through support to the SRNS Area Closures Project, through subcontracts from SRR to perform specialized sampling or analyses, and through commitment of funds received through SREL's Cooperative Agreement with the Department of Energy to address research topics of importance to the continuing missions of DOE on the SRS. The details of these projects are outlined below:

Research Support to SRNS Area Closure Projects, SRR, and DOE-SR

Examination of Selected SRS Aquatic Habitats for Metal Contamination and Bioaccumulation

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> April 2018; \$20,000 <u>SREL PI and Co-PI's</u> Larry Bryan, Dean Fletcher, and Dr. Stacey Lance Objectives

Objectives

The primary objectives of this study were to fill in data gaps relative to metal contamination in drainages and/or aquatic systems on the SRS for SRNS-ACP. Specifically, we (1) analyzed archived fish samples from Fourmile Branch and Beaver Dam Creek for metals and Radiocesium, (2) collected and analyzed samples from the upper tributaries of Fourmile Branch, areas with environmental conditions potentially conducive or indicative of increased metals availability, and (3) examined environmental factors (e.g., hydroperiod, size, depth, water chemistry)that influences background levels of mercury in Carolina Bays, wetlands presumably contaminated by atmospheric deposition. This is a second installment of funding for this project.

Summary of Research Activities

We analyzed additional archived fish samples from Fourmile Branch for metals and Radiocesium. We also analyzed sediment (N=4), biofilm (N=12), aquatic invertebrate (N=12), amphibian (N=3), and fish (N-57) samples from 4 sites in the upper tributaries of Fourmile Branch for a suite of metals. We analyzed THg in sediment (96) and biofilm (36) samples from three locations within 12 ephemeral wetlands. We built off of a previous study examining THg in amphibians and examined additional samples. In total we now have measured THg in southern leopard frogs (103), marbled salamanders (104) and mole salamanders (114). For a subset of amphibian samples we have measured methyl Hg (meHg). We are currently analyzing the sediment samples for meHg. Lastly, we analyzed trace elements and Hg in archived fish (36 species, N=566) and invertebrates (5 genera, N=30). We now have analyzed a total of 772 fish samples distributed across 43 species and 265 invertebrate composite samples distributed across 15 invertebrate genera (total N=1037). We have additionally analyzed liver and ovarian tissue from select fish species.

Conclusions

1) Trace element accumulation varies between and within fish and invertebrate families; feeding strategy and habitat use appear to be factors in some taxa, but patterns tended to be element and species specific. Mobility influences spatial avariability. The amount of THg in sediments and

biofilms was not related to wetland hydroperiod, but there is significant variation among the wetlands.

- 2) The amount of THg in amphibians is significantly related to wetland hydroperiod with larvae developing in shorter hydroperiod wetlands accumulating more Hg.
- 3) The among of THg accumulated in amphibian tissues differs among species with southern leopard frogs having the highest body burdens. However, levels of meHg were similar across species.

Major Impact(s) of Research

- 1) We are providing better understanding of metal accumulation in aquatic biota from two SRS drainages to aid in assessment of risk from legacy contaminants.
- 2) Combined with previous work, a solid baseline of D Area pre-closure contaminant accumulation in Beaver Dam Creek has been established.
- 3) For the Fourmile headwaters sites, we will determine if conditions presumed conducive to metal bioavailability actually resulted in higher metal concentrations.
- 4) When completed, we will have a better understanding of what drivers influence variation in background mercury concentrations in ephemeral wetlands. As initially predicted, amphibians inhabiting short hydroperiod wetlands are at a higher risk of accumulating levels of THg that may cause sublethal effects. More data are needed to determine what is driving patterns of THg deposition on the wetlands.

Other Project Personnel

Cara Love, PhD Student - SREL

Demetrius Calloway, Research Assistant - SREL

David Scott, Research Professional - SREL

Christina Fulghum, Research Technician - SREL

Brooke Lindell, Research Technician - SREL

Paul Stankus, Research Professional – SREL

Michaela Day, Research Assistant – SREL

Alexis Korotasz, Research Assistant - SREL

External Collaborators

NA

Products

- Fletcher, D.E., C.M. Fulghum, B.E. Lindell, A.H. Lindell, P.T. Stankus, and JV. McArthur. 2019. Trace element accumulation from coal combustion waste contamination differs among aquatic invertivores in a coastal plain stream. Annual Meeting of the Society of Freshwater Science, Salt Lake City, UT, May 2019 [poster].
- Fletcher, D.E., B. E. Lindell, C.M. Fulghum, A.H. Lindell, P.T. Stankus, and J.V. McArthur. 2018. Variability of trace element accumulation among invertivorous fishes from a coastal plain stream contaminated by coal combustion waste. Annual Meeting of the Society of Environmental Toxicology and Chemistry, Sacramento, CA, November 2018 [poster].
- Lance, S.L, D.C. Calloway, X. Xu, C. Love, M. Winzeler, A. Coleman, C. Tapia, R. Beasley, P. Walkup, and D.E. Scott. 2018. Distribution of total and methyl mercury in sediment and amphibians from ephemeral wetlands on the Savannah River Site. South Carolina Water Resources Conference, Columbia, SC.
- Calloway, D., C. Love, D. Scott, and S. Lance. 2018. The fate of atmospheric mercury (Hg) in ephemeral wetlands. Emerging Researchers National Conference. Washington, DC. Won 3rd place for best undergraduate platform presentation.
- Calloway, D., C. Love, D. Scott, and S. Lance. 2018. The fate of atmospheric mercury (Hg) in ephemeral wetlands. Minorities in Agriculture Natural Resources and Related Sciences. Greensboro, NC. Won 2nd place for best undergraduate platform presentation.
- Calloway, D., C. Love, D. Scott, and S. Lance. 2018. The fate of atmospheric mercury (Hg) in ephemeral wetlands. Society of Toxicology. San Antonio, TX.

Population Modeling of Contaminant Uptake in Aquatic Turtles on the Savannah River Site

Funding Entity

SRNS Area Closures Projects <u>Start Date and Funding Amount</u> March 2018; \$129,980 <u>SREL PI and Co-PI's</u> G. Dharmarajan and Tracey Tuberville

Objectives

Understanding how the effects of contaminants at the individual-level scales up to affect population-level responses is critical because most environmental directives aim to protect local populations, rather than individuals. Individual based models can effectively link individual and population level processes within a mechanistic framework, and are thus increasingly being recognized as powerful tools for risk-based management of contaminated sites. In this study we propose to improve our understanding of contaminant exposure risk in aquatic turtles inhabiting the Savannah River Site using a Markov-Chain Monte-Carlo simulation approach in conjunction with Bayesian inference. To this end we will develop a spatially explicit, individual-based Monte-Carlo simulation model that incorporates prior information on the spatial heterogeneity and trophic transfer of contaminants, as well as parameters associated with turtle ecology that likely influence exposure risk (e.g., habitat utilization and demography)

Summary of Research Activities

As of this report, we have collated and collected electronic copies of data from Dr. Tuberville's laboratory on contaminant levels in turtles sampled from the Savannah River Site. These data originate from collections made during previous ACP funding of Long-Lived Reptiles scopes during 2010-2011, 2014-2015. We have also collated life history and demography data based on SREL's long-term mark-recapture database for the species and published literature to parametrize our models. We have completed the first submodel (Spatial submodel) and generated preliminary results. We have modeled the landscape factors that affect landscape resistance to turtle movement using a novel Monte-Carlo simulation combined with a random forests algorithm, a common machine-learning approach. The landscape factors we used included those related to soil type, the dominant landcover (e.g., vegetation type, water and industrial areas), terrain (e.g., slope and roughness) and distance to various landscape features (e.g., roads, powerlines and Carolina bays). The preliminary spatial maps generated by our analyses reveal that the SRS generally consists of habitat that is not conducive to turtle movement. We found that hydric soils that are close to Carolina bays increase landscape permeability to turtle movement, while proximity to powerlines reduces turtle movement. It was not surprising that we also found that forested areas (both pine and hardwood cover) increased landscape permeability. Interestingly however, distance to roads and/or industrial areas had minimal effects on landscape permeability. We also have formulated the second submodel (*Bioenergetics submodel*), but the finalization of the model is awaiting results from the last submodel (see below). We have run into some data-related issues with regard to parametrization of the last submodel (Contaminant submodel). We have received spatial (e.g., ArcGIS file format) data from DOE on environmental contaminant levels on the focal metals (primarily V, Cr, Mn, Ni, Cu, Zn, As, Se, Sr, Cd, and Pb). However, one major issue is related to the fact that detailed environmental contaminant data for the SRS is primarily available for the IOUs, and thus many of the ephemeral wetlands inhabited by turtles has few data. While machine-learning algorithms trained on the IOU data can be used to extrapolate to the ephemeral wetlands there is no easy way to validate these results without observed data. To address this issue we are currently trying to parametrize a hierarchical machine-learning approach to model jointly the effects of landscape-level and individual-level factors on contaminant uptake **Conclusions**

The models and data analyses are being completed and no conclusions can be drawn at this time.

Major Impact(s) of Research

- 1) This study aims to produce ecologically relevant models of contaminant exposure in yellow bellied sliders wherein exposure is integrated over individual life-span taking into consideration age-specific bioenergetics and spatio-temporal heterogeneity contaminant exposure.
- 2) We hope to also identify parameters that critically impact model performance and help in identifying important deficiencies in existing data, thus helping to make future experimental research more focused.

Other Project Personnel

Amelia Russel, Research Technician – SREL Austin Coleman, Research Technician – SREL External Collaborators

NA

Products

No presentations or publications are associated with this project currently

Literature Review of Radionuclide Levels in SRS Fauna

Funding Entity

SRNS Area Closure Project

Start Date and Funding Amount

April 2016; \$78,000

SREL PI and Co-PI's

Larry Bryan, Dr. John Seaman, and Dr. I. Lehr Brisbin, Jr.

Objectives

The overall goal of the project is to compile the known published info1mation of radionuclides by SRS fauna, primarily from peer-reviewed literature, these/disse1tations and SREL technical/final project reports.

Summary of Research Activities

To date, over 270 published a1ticles, technical reports, theses/dissertations have been complied. Summarization of radionuclide concentrations in SRS has been completed for some elements (e.g., Pu, Sr) but remains on-going for others, including ¹³⁷Cs, the primary radionuclide found in fauna on the SRS.

Conclusions

We have generally completed the search but continue to add findings from recent publications. We will finalize report in early 2020.

Major Impact of Research

When completed, information pertaining to radionuclide concentrations on SRS fauna from 1960 through 2019 will be available in one document.

Other Project Personnel

Alexis Korotasz, Research Technician - SREL

External Collaborators

NA

Products

None at this time.

Hydrological Characterization of Beaver Dam Creek

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> May 2019; \$25,000 <u>SREL PI and co-PI's</u> Dean E. Fletcher

Objectives

Establish the hydrologic extent of Beaver Dam Creek after pumping has ceased, determine whether runoff from D Area is reaching perennial waters, and map primary runoff routes.

Summary of Research Activities

For decades, water flows in Beaver Dam Creek were augmented by Savannah River water pumped through the D-Area coal-fired power plant. After water pumping from the powerhouse ceased, the hydrology of Beaver Dam Creek was poorly known. We are establishing the extent of perennial stream in the historical Beaver Dam Creek channel. Maps were created from LiDAR imagery and aerial photos for use in field surveys. Field surveys have found that under current stream-flow levels and D Area activities, surface water from the D Area Discharge Canal is continuous through Beaver Dam Creek (BDC) to the Savannah River, however, complex flow patterns exist. The upper 2.5 km of BDC tends to be shallow with relatively low discharge, but is continually flowing. In the braided section about 1.6 km downstream of D Area, water presently only flows through the west channel at base flow. The east channel has flowed during storm events indicating considerably higher flows during storm events. BDC joins an abandoned river channel about 2.5 km downstream of D Area. Water will also flow from the Savannah River into this area when the river floods. Springs appear to enter BDC from this channel. BDC is larger and slower below this point. This area represents a substantial depositional area with much deeper and softer sediments. Sediments could be accumulating from upstream in BDC or through the Savannah River overflow channel. Trace element analyses of these sediments will be required to establish their source. BDC water continues to flow toward the Savannah River below this point with variable water velocities. BDC stream flow becomes relatively still as it approaches the Savannah River. At a point about 400 m from the Savannah River the water direction reverses and water began to flow from the river into BDC. This directionality will depend upon factors such as river level, water flowing though secondary floodplain channels, and stormflows from D Area. Regarding the distribution of surface water between D Area and the river, presence of fish throughout the BDC channel suggests that that the observed surface water distribution has been continual and not the result of a recent events. Fieldwork will continue to examine variability of the BDC flow patterns.

Conclusions

Conclusions are presently preliminary.

Major Impact of Research

Establishing runoff routes and hydrologic state of this area will provide a better understanding of the risk of offsite transport of contaminants from this system.

Other Project Personnel

Christina Fulghum, Research Technician – SREL

Paul Stankus, Research Professional – SREL

Erin Spivey, Research Technician – SREL

Jennifer Dirks, Research Technician - SREL

External Collaborators

NA

Products

Preliminary GIS maps are being assembled.

Hydrological Characterization Around D Area

<u>Funding Entity</u> SRNS Area Closure Project <u>Start Date and Funding Amount</u> May 2019; \$25,000

SREL PI and co-PI's

Dean Fletcher

Objectives

Establish runoff patterns from D Area into the Savannah River Swamp. Identify, map, and monitor water quality of springs and seeps down slope from D Area.

Summary of Research Activities

The ground water below D Area has a reduced pH as the result of operation of a coal-fired power plant. Future remediation will increase ground water levels under and downslope of D-Area. Surface water runoff patterns and areas where ground water reaches the surface are being established. A series of maps were created from LiDAR imagery and aerial photos for use in field surveys. Field reconnaissance is mapping runoff routes from D Area onto and across fluvial terraces between D Area and the Savannah River. Most springs and seeps were dry during drought conditions, but based on geomorphology and vegetation, locations of potential spring runs and seeps are being identified for future evaluation during wetter conditions. A portable pH/conductivity meter was purchased to monitor water quality of springs/seeps in this area.

Conclusions

1) Conclusions are presently preliminary.

Major Impact(s) of Research

1) Establishing runoff routes and hydrologic state of this area will provide a better understanding of the risk of offsite transport of contaminants from this system.

Other Project Personnel

Christina Fulghum, Research Technician - SREL

Paul Stankus, Research Professional – SREL

Erin Spivey, Research Technician – SREL

Jennifer Dirks, Research Technician - SREL

External Collaborators

NA

Products

Preliminary GIS maps are being assembled.

Controlling the spread of radioactive contamination by biotic vectors on the Savannah River Site

Funding Entity DOE-SR **Start Date and Funding Amount** October 2018; \$198,771

SREL PI and Co-PI's

Dr. O. E. Rhodes, Jr., Dr. G. Dharmarajan, A. L. Bryan, L. Lee, and Dr. T. DeVault

Objectives

During routine surveys at the H-Area Tank Farm (HTF), SRS, high levels of radioactivity (>100,000 disintegrations per minute (dpm) beta-gamma activity) were detected in bird feces in December 2017. Additional surveys by SRNS revealed at least three more instances of contaminated bird feces in the HTF complex (40,000-240,000 dpm beta-gamma activity). The overarching goal of this project is to identify the source(s) of radioactive materials that is (are) contributing to the contamination of facilities and other structures on the SRS so that these risks can be mitigated. This study had four objectives: (1) Identify the community of bird vectors that pose a high risk of exposure to radionuclide contaminants at the HTF and characterize the levels of contamination extant in this community; (2) Quantify spatial risk associated with the accumulation and transport of radionuclide contaminants by bird vectors within and outside the HTF; (3) Produce a spatial risk map and identify the distribution of potential radionuclide contaminant sources (soil and/or plants) in and around the HTF; (4) Evaluate mice as a potential vector of radionuclide contamination within the HTF and identify potential sources of contamination for mice within the facility

Summary of Research Activities

During the study period we captured a total of 506 individuals belonging to 50 species. We quantified whole-body radionuclide contamination levels in 225 birds belonging to 45 species using Gamma Well counter with a NaI (TI) gamma detector. We found that 3 of the 225 individuals counted (1.3%) had counts > 5000 dpm. All birds with counts > 5000 dpm and a subset of birds below the threshold were euthanized and their tissue analyzed for radionuclide contaminants using an auto-gamma counter. Importantly, no bird captured or collected showed radiation levels greater than background when scanned with Ludlum probes in the field/lab. To characterize bird habitat utilization and home range we deployed VHF transmitters on 70 individuals belonging to 9 species. We have collected over 2500 radiotelemetry locations on these individuals and data collection is ongoing. Analyses of these data will be carried out at the end of the Fall field season. To characterize potential radionuclide contaminant sources we have carried out intensive sampling of vegetation in and around the HTF area. We have collected 588 vegetation samples, of which 273 have been analyzed so far for radionuclide contaminants using an autogamma counter. So far only one vegetation sample has shown radiation levels above background. This was a sample of the plant pokeweed (*Phytolacca americana*), and this sample had a count of 2500dpm (27Bq/g dry). Several more samples of pokeweed have been collected from controlled areas across the HTF and these remain to be analyzed. Evaluate rodents as a potential vector of radionuclide contamination within the HTF we also trapped mice and rats. To date we have trapped and whole-body counted 75 individual rodents belonging to three species. Of the individuals counted only one individual had counts > 5000 dpm. The mouse with counts > 5000 dpm and a subset of rodents below the threshold were euthanized and their tissue analyzed for radionuclide contaminants using an auto-gamma counter. Importantly, no mouse captured or collected showed radiation levels greater than background when scanned with Ludlum probes in the field/lab.

Conclusions

Data collection and analyses are still ongoing and no conclusions are available at this time.

Major Impact(s) of Research

- 1) To characterize the identity and spatial scale of movement of bird (and rodent) vectors that could accumulate and transport radionuclide contaminants
- 2) To characterize and identify the spatial distribution of radionuclide contamination sources (e.g., vegetation) and identify biotic species that accumulate radionuclides

- 3) To determine whether remediation strategies can be developed to mitigate risks associated with biotic accumulation and transport of radionuclides within or into the HTF
- 4) To determine the occurrence (or lack thereof) and concentrations of radionuclide contaminants in biotic vectors commonly collected in areas adjoining the HTF, and to assess the likelihood of public exposure to these contaminants through these sources.

Other Project Personnel

Mary Chapman, Research Technician – SREL Abigail Riggs, Research Technician – SREL Tyler Walters, Research Technician – SREL

Kaitlin Wilms, Research Technician - SREL

Elizabeth Butler, Research Technician – SREL

External Collaborators

NA

Products

Chapman, M., A. Riggs, K. Wilms, T. Walters, J. Garabedian, L. Lee, A. L. Bryan, G. Dharmarajan, O. E. Rhodes, Jr. (2019) Avian and Small Mammal Habitat Use Within a Nuclear Storage Facility on the Savannah River Site. SREL Graduate Student Symposium. July 19, 2019. Aiken, SC

Examination of Mercury/Methylmercury in Aquatic Biota Associated with Fourmile Branch

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> April, 2018; \$20,000 <u>SREL PI and Co-PI's</u> Larry Bryan, Dr. Xiaoyu Xu and Dr. Gary Mills

Objectives

We received the final year of five years of funding in Febmary of 2018. During the initial year of the study, we perfolmed a preliminary smvey of total mercmy (THg) in biota in the three areas of concern (H-Area seepline, F-Area seepline, Savannah River swamp system [SRSS]) along the gradient of Fournile Branch (FMB). In each of the three subsequent years, we examined THg in environmental samples (sediments, biofilms, aquatic inveltebrates and fish) in one of the SRSS, H-Area, and F-Area sites, and analyzed a subset of the samples for methymercury (MHg). In this the final year of study, we filled data gaps from the previous years and initiated data summary and write-up.

Summary of Research Activities

Year 1 was a general survey of THg in biota in the three areas of concern. In Year 2, within the SRSS, we compared environmental samples from sites in the area associated with Fourmile to samples collected in sites in Crackerneck (up-river from Fourmile). We found THg concentrations were generally higher in Fourmile SRSS samples than samples from Crackerneck, although not consistently so, demonstrating a pattern of a patchy distribution of Hg in the SRSS. H-Area environmental samples were consistently higher in THg than similar samples collected from a control stream, Mill Creek (Year 3), but THg in F-Area samples (Year 4) were considerably lower than those H-Area. We also deployed Diffusive Gradients in Thin Films (DGT) in SRSS, FMB, and Mill Creek. Mill Creek exhibited constant concentrations of DGT measured Hg (DGT-Hg) through selected sediment depth intervals, but profiles at contaminated sites showed sharp increases at the sediment-water interface (SWI), demonstrating the impoltance of SWI as it indicated the accumulation and/or generation of DGT-Hg in the pore water gradient. In this the final year of study, we filled data gaps from the Year -4 monitoring (e.g., MHg analyses), acquired supplemental data (e.g., stable isotope values of selected samples) to enhance and/or confirm our understanding of mercury bioavailability. The food web magnification factor calculated with sample MHg concentrations and &15N values in FMB is 3.5, meaning MHg concentration increased approximately 3.5-fold for each trophic level. We continue to summalize selected aspects of our findings. Meanwhile, passive samplers (peepers) were deployed to measure concentrations of labile Hg species, major anions (chloride and sulfide), dissolved organic carbon in the sediment pore waters. We have completed the analysis of anions and part of the Hg measurements. Once all analysis was completed, the veltical profile of anions and organic matter in pore waters can be explored, which will provide implications on Hg biogeochemistiv in selected sites, especially the anaerobic methylation and demethylation of Hg.

Conclusions

Mercury concentrations in the SRSS associated with Fourmile Branch were elevated, yet "hot spots" were patchily distributed. Mercury concentrations associated with biota in the H and F-area seeplines were elevated above a control stream, but H-Area biota concentrations was> F-Area biota concentrations. Research based on DGTs indicated increases in Hg concentrations at the sediment-water interface (SWI), demonstrating the importance of SWI in the accumulation and/or generation of DGT-Hg in the pore water gradient. The food web magnification factors of MHg, based on trophic data (biofilm, inve1tebrates, and fish) were 9.6 (95% CI: 4.0 - 23.4) and 4.4 (95% CI: 2.5 - 7.7) for the Fourmile and the control stream, respectively.

Major Impact(s) of Research

- 1) DGT-Hg concentrations in the water were positively correlated to biofilm Hg concentrations, which can be used to generate a modified biomagnification model. Therefore, MHg accumulation at different trophic positions can be estimated with DGT-Hg concentrations in the water.
- 2) We provide here a better understanding of abiotic and biotic interactions related to THg and MHg bioavailability within the Fourmile Branch system.
- 3) DGT-Hg concentrations in the water were positively correlated to biofilm Hg concentrations, which can be used to generate a modified biomagnification model. Therefore, MHg accumulation at different trophic positions can be estimated with DGT-Hg concentrations in the water.
- 4) We provide here a better understanding of abiotic and biotic interactions related to THg and MHg bioavailability within the Fourmile Branch system.

Other Project Personnel

Alexis Korotasz, Research Technician – SREL David Haskins, Graduate Student – SREL

External Collaborators

NA

Products

- Xu, X., A.L. Bryan, G. L. Mills, and A.M. Korotasz. 2019. Mercury speciation, bioavailability, and biomagnification in contaminated stream systems on the Savannah River Site (S.C., U.S.A.). Science of the Total Environment 668:261-270.
- Haskins, D.L., A.M. Korotasz, and A.L. Bryan. 2019. Mercury concentrations in the two-toed amphiuma (*Amphiuma means*) and lesser siren (*Siren intermedia*): Validating non-lethal sampling methods in southeastern aquatic salamanders. Archives of Environmental Contamination and Toxicology 77:330-335.

Tracking Sources of Mercury in Contaminated Aquatic Systems on the Savannah River Site

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> April 2017; \$31,954 <u>SREL PI and Co-PI's</u> Dr. Xiaoyu Xu and Larry Bryan

Objectives

The Savannah River Site (SRS) has received mercury (Hg) contamination from multiple sources, including atmospheric deposition (local and non-local), off-site pollution (Savannah River – Olin Canal) and on-site activities. Accumulation of Hg by SRS biota has been documented frequently, occasionally at levels of regulatory concern, but the sources of Hg remain unclear. Our objective is to utilize a relatively new technique, Hg stable isotope analysis, in an attempt to confirm sources of Hg found in SRS biota. The isotope analysis will occur at the Center for Applied Isotope Studies at UGA after custom-fabricated accessories are installed on their existing instrumentation.

Summary of Research Activities

We collected sediment samples from the Savannah River (above and below Olin Canal) and Fourmile Branch, and their total Hg and MeHg (methylmercury) concentrations were analyzed. We also collected biota samples from Fourmile Branch in FY2017, and their total Hg and MeHg (methylmercury) concentrations were analyzed. We collected pore water samples from Fourmile Branch in FY2018, and their total Hg and MeHg (methylmercury) concentrations were analyzed. The stable carbon and nitrogen isotope signatures were determined for all samples, and the δC^{13} and δN^{15} ratios in the sediment were statistically different than that in the biota samples. All sediment and biota samples were digested with mixed acid (HCL and HNO₃) and prepared for the future Hg isotope analysis. We had the needed accessories for Hg isotope analysis fabricated, they have been installed on the existing instrumentation, and the system has been calibrated and tested. But the accuracy of the system still needs to be improved and the QCQA of a new standard reference material also needs to be certified. We hired a research professional who is an expert in Hg stable isotope and will start on January 6th, 2019. We anticipate the instrument will be adjusted well and the samples can be analyzed in FY20.

Conclusions

We collected the sediment and biota samples and customized the instrumentation to analyze for Hg isotopes. Sample analysis is pending.

Major Impact(s) of Research

- 1) Impacts cannot be determined prior to the analyses of the samples.
- 2) Assuming we can isotopically differentiate among the various sources of Hg, when completed, we will have a better understanding of Hg sources in biota found on the SRS biota.

Other Project Personnel

Sarah Jantzi – UGA

External Collaborators

NA

Products

- Xu, X., A.L. Bryan, G.L. Mills, and A.M. Korotasz. 2019. Mercury speciation, bioavailability, and biomagnification in contaminated stream systems on the Savannah River Site (S.C., U.S.A.). Science of the Total Environment 668:261-270.
- X. Xu. Poster. Mercury Speciation, Bioavailability, and Biomagnification in Contaminated Streams on the Savannah River Site (SC, USA). July 2019. SREL Graduate Symposium.

Contaminants in Wild Pigs and Eastern Wild Turkeys

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> October 2017; \$176,290 <u>SREL PI and Co-PI's</u> Dr. Jim Beasley, Larry Bryan, and Dr. James Martin Objectives

Objectives

The primary objective of this research is to quantify levels of Hg, trace elements, and radiocesium in wild turkeys and wild pigs on the SRS, and compare contaminant burdens in these species on the SRS to populations off-site to assess potential risks to hunters.

Summary of Research Activities

Wild Turkeys –We have collected samples from 48 turkeys harvested during the Ultimate Turkey Hunts held on the SRS, as well as from an additional 17 turkeys near areas of known contamination on the SRS, for a total of 65 birds. Off-site samples (17) were collected from hunters in South Carolina and Georgia. Samples taken include; breast and leg muscle, liver, and feathers. Feathers were collected from a subset of individuals that we will use to determine whether turkey feathers can be used as a non-lethal predictor of contaminant burdens within muscle or liver tissue. We are currently analyzing trace element concentrations in the final samples, and are beginning to analyze the data and prepare a manuscript for publication stemming from these data.

Wild Pigs – From 2018-2019 we collected wild pig muscle and liver samples from the SRS as well as control samples collected from the Congaree National Park. We have collected a total of 70 pigs from various locations on site, as well as additional from the Congaree. All SRS samples have been collected proximal to areas of known contamination, and include D-area, the Savannah River Swamp, L-Lake Dam, Dunbarton, Par Pond, P-area, R-area, Tim's Branch, H-area, and Hunt Unit 25, which borders several SRS facilities. These samples have been freeze-dried, and we are currently in the process of prepping the samples for radiocesium and trace element testing.

Conclusions

Data analyses are incomplete; thus there are no conclusions at this time.

Major Impact(s) of Research

- 1) This research will provide the first assessment of contaminant burdens in wild turkeys on the SRS, data necessary to assess potential risks to hunters consuming birds harvested during annual hunts on site.
- 2) Spatial analyses will be used to determine whether spatial variability in contaminant burdens exists among wild turkeys inhabiting various locations on the SRS.
- Additional data on Hg, trace elements, and radiocesium in wild pigs will be combined with data from an earlier study to provide an updated assessment of potential risks to hunters consuming pigs collected on/near the SRS.

Other Project Personnel

Kaitlin Wilms, Research Technician – SREL Sara Cheatham, Research Technician – SREL Chris Leaphart, Ph.D. Student – SREL Cody Tisdale, M.S. Student – SREL Allison Rakowski, Research Professional – SREL **External Collaborators** Robert Byrd (USDA-APHIS-WS)

Products Tisdale, C.A., J.A. Martin, and J.C. Beasley. 2019. Lead concentrations in muscle tissue of wild turkeys harvested with lead and copper-plated lead shot. Savannah River Ecology Laboratory Graduate Student Symposium. Aiken, SC.

Contaminants in Resident Waterfowl on the Savannah River Site

Funding Entity

SRNS Area Closure Project

Start Date and Funding Amount

October 2019; \$227,063 **PI and Co-PI's**

Dr. Jim Beasley

Objectives

This research seeks to improve our understanding of the bioavailability and effects of various contaminants on waterfowl species inhabiting the SRS. The primary objectives of this research are to: 1) quantify contemporary contaminant levels in wood ducks on the SRS, 2) determine the influence of prenatal mercury contamination on wood duck duckling survival, and 3) determine the influence of fine-scale habitat use on radiocesium burdens in waterfowl on the SRS.

Summary of Research Activities

Due to the arrival of funds after the migratory season for the species being studied, we had to delay collection of data on contaminant burdens in SRS wood ducks by several months, as these samples must be collected in fall/winter. We will begin data collection during fall/winter 2019/2020 once we can begin sampling free-ranging wood ducks. During FY19 we were able to establish and test our protocol for dosing duck eggs with methylmercury II chloride dissolved in corn oil. We conducted these pilot studies over the summer (FY19) and have all protocols and materials in place to initiate these studies in spring 2020.

Conclusions

Data analyses are incomplete; thus there are no conclusions at this time.

Major Impact(s) of Research

- 1) This research will provide the most comprehensive estimates of the levels of various contaminants (e.g., mercury) in wood ducks inhabiting the SRS to date.
- 2) Spatial analyses will be used to determine how fine-scale space use influences contaminant burdens in waterfowl utilizing wetlands on the SRS.
- 3) These findings will produce novel data on the extent to which mercury exposure could contribute to detrimental effects on wood ducks.

Other Project Personnel

Chris Leaphart, Ph.D. Student – SREL

External Collaborators

NA

Products

Leaphart, C.L., and J.C. Beasley. 2019. Effects of prenatal methylmercury exposure on waterfowl hatchling phenotypic plasticity and survival. Savannah River Ecology Laboratory, Graduate Student Symposium. Aiken, SC.

Tritium Distribution at the Tritiated Water Management Facility - Southwest Plume Interim Measures

Funding Entity

SRNS Area Closure Project Start Date and Funding Amount

October 2018; \$127,000

SREL PI and co-PI's Dr. John C. Seaman

Objective

Evaluate the efficacy of ongoing remediation efforts at the SRS Mixed Waste Management Facility (MWMF) to address the tritium and 1, 4 dioxane plume originating from the Old rad Waste Burial Ground.

Summary of Research Activities

In FY 2019, SREL worked collaboratively with the SRS-US Forest Service, SRNS-ACP and DOE to complete the following activities: (1) collect, process and analyze 22 soil cores collected to a depth of \approx 3 m to evaluate tritiated water (³H₂O) distribution as an estimate of irrigation efficiency (\approx 220 samples annually); and (2) maintain and update the Cornell Model for estimating water-use efficiency.

Conclusions

The estimated tritium evapo-transpiration efficiency for individual irrigation plots through the end of calendar year 2018 based on soil core samples ranged from ≈ 69.8 to 99.1% when accounting for leaching below the root zone, with lower tritium use efficiencies generally reflecting the lower vegetative cover associated with the Western Expansion Area. Monthly efficiency results derived from the Cornell 1D model ranged from 65.8 to 88.9% between plots, with lower efficiencies observed for the Western Expansion Area plots, i.e., plots 45v and 48. The average efficiency for all plots was approximately 86.7 ± 1.2 % for the original plots, 78.6 ± 2.1 % for the EEA plots, and 68.0 ± 1.4 % for the WEA plots, resulting in an overall of annual efficiency of 81.1 ± 7.3 % for all 11 monitored plots.

Other Project Personnel

Matt Baker, Research Professional – SREL Christina Logan, Research Professional – SREL Morgan Shapiro, Lab Technician – SREL Jeffery Lott, Lab Technician – SREL Liyun Zhang, Grad Assistant – SREL

External Collaborators

NA

Products

Baker, M.R., and J.C. Seaman. 2019. Estimating Evapo-Transpiration Losses for Tritium at the MWMF: 2018 End of Year Summary Report. SREL Doc.: R-19-0002. Submitted to SRNS-ACP March 27, 2019.

Selective Sorbents for the In Situ Immobilization of ¹²⁹I and ⁹⁹Tc at the Four-Mile Branch Seepline

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> August 2019; \$100,000 <u>SREL PI and co-PI's</u> Dr. John C. Seaman

Objective

The primary objective is to evaluate selective sorbents for the in situ immobilization of both ¹²⁹I and ⁹⁹Tc in an effort to enhance the natural attenuation capacity at the Four-Mile Branch seepline.

Summary of Research Activities

Based on recent literature, a number of potential sorbents were identified for initial lab evaluation, with an emphasis on in situ methods that don't require removal and subsequent disposal. The proposed sorbents can be placed in one of two general categories: metal-based and carbon-based sorbents. The metal-based sorbents included a Porous Iron Composite (PIC) materials that has proven to be superior to other zero valent iron (ZVI) materials for immobilizing ⁹⁹Tc and uranium (U), even in the presence of O₂ and alternate electron acceptors, such as NO₃⁻, a major component of the F-Area plume. Two additional PIC materials were evaluated that incorporate copper (Cu) and silver (Ag) into the materials, referred to as Cu-PIC and Ag-PIC, respectively, as a potential means of increasing the effectiveness of the PIC material at immobilizing I⁻ through precipitation. Reagent grade ZVI and two commercial Fe-oxide based sorbents were included for comparison with the novel materials. The carbon-based materials included granular activated carbon (GAC) plus similar materials that have been modified to include Ag and Cu (i.e., Ag-AC and Cu-AC), similar to the PIC sorbents discussed above.

The effectiveness of various sorbent materials is highly dependent on the chemical conditions of the system in which they are applied. Factors such as pH, dissolved organic matter (DOM) content, redox potential and the presence of other electron donors, such as nitrate, greatly impact the initial partitioning and the long-term effectiveness of sorbent in immobilizing contaminants. In consultation with ACP, SREL established a limited set of test conditions that reflect specific groundwater parameters that reflect important phases in the remediation process, i.e., active remediation (sorption) and post-remediation monitoring (desportion). And finally, batch sorption experiments were conducted using stable Iodine (¹²⁷I) and Rhenium (Re as perrhenate, ReO₄⁻) as analogues for ¹²⁹I and ⁹⁹Tc, respectively, while monitoring other important components, i.e., total Fe, Fe(II), NO₃⁻, NH₄⁺, etc. Care was taken to also evaluate if the proposed remediation strategies exceed regulatory limits for Ag and Cu. **Conclusions**

Based on initial batch tests, the number of potential sorbents was reduced from over twenty to four that will be evaluated in subsequent experiments. Batch tests using ¹²⁹I and ⁹⁹Tc are currently ongoing.

Major Impact(s) of Research

1) Identify in situ treatment options that reduce the need for expensive, invasive remediation efforts along the Four-Mile Branch stream.

Other Project Personnel

Fanny Coutelot, Post-Doctoral Research Associate – SREL Matt Baker, Research Professional – SREL Christina Logan, Research Professional – SREL Morgan Shapiro, Lab Technician – SREL Jeffery Lott, Lab Technician – SREL **External Collaborators** Dr. D. Kaplan (SRNL)

Dr. D. Li (SRNL)

Products

- Li, D., D.I. Kaplan, K.A. Price, J.C. Seaman, K. Roberts, C. Xu, P. Lin, W. Xing, K. Schwehr, P.H. Santschi. 2019. Iodine immobilization by silver-impregnated granular activated carbon in cementitious systems. Journal of Environmental Radioactivity. 208-209. doi.org/10.1016/j.jenvrad.2019.106017.
- Coutelot, F.M., R.J. Thomas, and J.C. Seaman. 2019. Using Porous Iron Composite (PIC) Material to Immobilize Rhenium as an Analogue for Technetium. Environment International. 128; 379-389. doi.org/10.1016/j.envint.2019.05.001.
- Li, D., J.C. Seaman, S.E. Murph, D.I. Kaplan, K. Taylor-Pashow, R. Feng, H. Chang, M. Tandukar. 2019. Porous iron material for TcO₄⁻ and ReO₄⁻ sequestration from groundwater under ambient oxic conditions. J. Hazardous Materials. 374:177-185. doi.org/10.1016/j.jhazmat.2019.04.030.
- Kaplan, D.I., K.A. Price, C. Xu, D. Li, P. Lin, W. Xing, R. Nichols, K. Schwehr, J.C. Seaman, T. Ohnuki, N. Chen, P.H. Santschi. 2019. Iodine Speciation in a Silver-Amended Cementitious System. Environment International. 126:576-584. doi.org/10.1016/j.envint.2019.02.070.
- Li, D., J.C. Seaman, D.I. Kaplan, S.M. Heald, and C. Sun. C. 2019. Pertechnetate (TcO₄⁻ sequestration from groundwater by cost-effective organoclays and granular activated carbon under oxic environmental conditions. Chemical Engineering Journal, 360, 1-9. DOI: 10.1016/j.cej.2018.11.146.

<u>Technical Support Provided by the Savannah River Ecology Laboratory (SREL) for R&D of</u> <u>Cementitious-Type Materials (i.e., Saltstone) and Soils</u>

Funding Entity

Savannah River Remediation (SRR) <u>Start Date and Funding Amount</u> October 2018; \$337,000 <u>SREL PI and co-PI's</u> Dr. John C. Seaman <u>Objectives</u>

At the Department of Energy's Savannah River Site (SRS) a mix of cementitious dry feed materials (i.e., portland cement, blast furnace slag (BFS) and fly ash) are combined with low-level radioactive saltwaste solutions to form a cementitious material known as saltstone, which is then deposited in a series of concrete vaults for long-term disposal at the Saltstone Disposal Facility (SDF). The objectives for FY19 include ongoing evaluations of contaminant leaching properties (⁹⁹Tc, ¹²⁹I, and ¹³⁷Cs) of saltstone simulants and additional evaluations of the proposed "cement free" saltstone formulations, both in terms of physicochemical properties (i.e., heat capacity, heat of hydration and thermal conductivity) and contaminant retention of intact monoliths. SREL also started testing proposed liner materials for SDU 7. **Summary of Research Activities**

For FY19, ⁹⁹Tc/¹²⁹I spiked "cement free" saltstone simulants were tested using SREL's Dynamic Leaching Method (DLM) and EPA 1315. In the DLM method a flexible-wall permeameter cell is used to achieve saturated leaching through the intact monolith under an elevated hydraulic gradient in an effort to evaluate the persistence of reductive capacity and subsequent changes in contaminant partitioning that occur within intact saltstone monoliths. The composition of the chemical leachates from both tests are then analyzed in an effort to identify potential critical reactions and solid phases controlling contaminant partitioning through geochemical modeling.

DLM tests continue using both spiked saltstone simulants and actual saltstone collected from SDU2A. In FY19, SREL expanded the analysis of the mineralogical and elemental composition of the saltstone dry feed materials using an x-ray diffraction and x-ray fluorescence. This information is relevant in predicting the observed variations in contaminant partitioning associated with changes in dry feed batches. SREL also continued to evaluate the thermal properties of various saltstone formulations, as impacted by compositional changes in the dry feed materials (e.g., cement free materials) and water-to-dry feed material ratios. These results were used by SRR in the development of a thermal model to predict heating within SDU vault 5A. Higher curing temperatures may increase the volatility of organic compounds contained within the salt waste, and potentially impact grout quality. The ability to predict SDU temperatures provides significant advantages with respect to safety and operational planning, and ensuring that grout performance is not compromised.

Conclusions

Contaminant leaching from the "cement free" saltstone formulations were similar to the current 45:45:10 formulation. DLM leaching results are generally consistent with EPA 1315 tests in terms of the degree of contaminant retention within the saltstone grout. In general, the hydraulic conductivity (K_{sat}) values obtained by common test methods are higher than the values observed by the DLM testing.

Major Impact(s) of Research

- First DLM and EPA 1315 data confirming the performance of cement free saltstone in retaining both ¹²⁹I and ⁹⁹Tc
- 2) Provided thermal data used in the development and validation of a thermal model for SDU performance
- 3) Initial dry feed characterization illustrating batch to batch variability

Other Project Personnel

F.M. Coutelot, Postdoc – SREL M.B. Baker, Research Professional – SREL C. Logan, Research Professional – SREL S. Simner, Research Associate – SREL **External Collaborators**

Dr. D. Kaplan (SRNL)

Dr. D. Li (SRNL)

Dr. J. Mangold (SRR)

Products

- Simner, S.P., J.C. Seaman and M. Baker. 2019. Thermal Properties of SDU 5A Saltstone Simulants. SREL DOC No. R-19-0002 Rev. 1.0. Submitted to SRR July 17, 2019.
- Seaman, J.C., S.P. Simner, M.R. Baker, and C. Logan. 2019. Contaminant Leaching from Saltstone Simulants: Summary of EPA 1315 and Dynamic Leaching Method Results for FY2019. SREL DOC No. R-20-0002 Rev. 1.0. Submitted to SRR October 14, 2019.
- Simner, S.P. J.C. Seaman. 2019. Batch-to-Batch Characterization of Dry Feed Materials Used in Saltstone Production. SREL DOC No. R-20-0001 Rev. 2.0. Submitted to SRR October 17, 2019.
- Coutelot, F.M., R.J. Thomas, and J.C. Seaman. 2019. Using Porous Iron Composite (PIC) Material to Immobilize Rhenium as an Analogue for Technetium. Environment International. 128; 379-389. doi.org/10.1016/j.envint.2019.05.001.
- Li, D., J.C. Seaman, S.E. Murph, D.I. Kaplan, K. Taylor-Pashow, R. Feng, H. Chang, M. Tandukar. 2019. Porous iron material for TcO4- and ReO4- sequestration from groundwater under ambient oxic conditions. J. Hazardous Materials. 374:177-185. doi.org/10.1016/j.jhazmat.2019.04.030.
- Kaplan, D.I., K.A. Price, C. Xu, D. Li, P. Lin, W. Xing, R. Nichols, K. Schwehr, J.C. Seaman, T. Ohnuki, N. Chen, P.H. Santschi. 2019. Iodine Speciation in a Silver-Amended Cementitious System. Environment International. 126:576-584. doi.org/10.1016/j.envint.2019.02.070.
- Li, D., J.C. Seaman, D.I. Kaplan, S.M. Heald, and C. Sun. C. 2019. Pertechnetate (TcO4–) sequestration from groundwater by cost-effective organoclays and granular activated carbon under oxic environmental conditions. Chemical Engineering Journal, 360, 1-9. DOI: 10.1016/j.cej.2018.11.146.

SREL also assisted in completing the following documents:

Cement-Free Formulation Down-Select Report. SRR-CWDA-2019-00003 rev. 0. February 2019. SDU Thermal Model Inputs: July 2019, X-ESR-Z-00045, Rev. 0. July 2019.

Aqueous and Solid Phase Characterization of Potential Tank Fill Materials

Funding Entity

Savannah River Remediation (SRR) Start Date and Funding Amount

June 2019; \$109,000

SREL PI and co-PI's

Dr. John C. Seaman

Objectives

SREL is conducting a series experimental tests evaluating the dynamic interactions of Savannah River Site (SRS) soil/vadose zone pore-water solutions in contact with Tank Closure Grout (TCG). Radionuclide release from residual tank waste is largely controlled by tank grout degradation in contact with water that has passed through the closure cap. Infiltration passes through the tanks and drives changes in grout mineralogy/composition, with the emerging fluid reflecting such interactions that vary over time. The current project focuses on identifying changes in aqueous chemistry (i.e., Eh, pH, DO, alkalinity, solution components, etc.) that occur in both open and closed (i.e., oxic and anoxic systems) environmental systems with extended leaching.

Summary of Research Activities

Dynamic column experiments and extended batch equilibrations are underway to evaluate the interactions of soil backfill with cementitious materials (i.e., TCG) in a simulated leaching environment that mimics potential leachate chemistries contacting various SRS waste disposal units. In addition to monitoring eluate chemistry, changes in the properties and solid phase composition of the soil and test cement materials are extensively characterized using appropriate analytical techniques (i.e., x-ray diffraction (XRD) spectroscopy, x-ray fluorescence (XRF) spectroscopy).

Major Impact(s) of Research

1) Batch and column results will be used in updating a flow and transport model to describe radionuclide release from residual tank waste based on component solubility as controlled by tank grout degradation in contact with water that has passed through the closure cap.

Other Project Personnel

C. Logan, Research Professional – SREL

M. Shapiro, Research Technician – SREL

J. Lott, Research Technician – SREL

External Collaborators

Dr. J. Mangold (SRR)

Dr. G. Flach (SRR)

Products

Seaman, J.C., and M.R. Baker. 2019. SREL Test Plan: Aqueous and Solid Phase Characterization of Potential Tank Fill Materials. SREL. Doc. R-19-0004, submitted to SRR June 4, 2019.

Watersnakes as ecological receptors for mercury contamination and bioaccumulation on the SRS

Funding Entity

SRNS Area Closure Project <u>Start Date and Funding Amount</u> January 2018; \$105,000 <u>SREL PI and co-PI's</u> Dr. Tracey D. Tuberville

Objectives

The overarching objective is to explore the value of watersnakes (*Nerodia* spp.) as receptor species for mercury (Hg) contamination across the Savannah River Site (SRS) and in the Savannah River area. We are targeting three primary aquatic system types: 1) the Savannah River and its tributaries, in which the brown watersnake (*N. taxispilota*) is the dominant snake species; and 2) former nuclear cooling reservoirs in which the green watersnake (*N. floridana*) is the dominant species; and 3) isolated ephemeral wetlands (i.e., Carolina bays) in which the banded watersnake (*N. fasciata*) is the dominant species. All three species are strictly carnivorous but they vary in the degree of specialization on fish vs amphibians – some of which may be due to prey composition in their aquatic habitat. These three systems also vary in their source and degree of Hg contamination, which will also influence Hg bioaccumulation in our target species. We will compare Hg body burdens in *Nerodia spp*. across these three system types and evaluate the effects of system type, species, body size, and sex on Hg concentrations.

Summary of Research Activities

This fiscal year focused on collection of aquatic snakes from the Savannah River system and isolated Carolina bay wetlands. We completed analysis of Hg concentrations in tail tips and blood samples from *N. taxispilota* collected from the Savannah River and tails tips from *N. fasciata* and *N. floridana* collected from reference wetlands. We also had the opportunity to compare Hg concentrations in current samples from Lower Three Runs Creek and those collected from same locality in 1980s. Two graduate students are using data from this project to form the basis of their thesis or dissertation.

Conclusions

We have only begun preliminary analysis of samples and associated data. However, based on these preliminary analyses, the following patterns have emerged:

- 1) Hg concentration in both blood and tail tips is positively and significantly correlated with individual body size in both *N. taxispilota* from Savannah River and *N. floridana* and *N. fasciata* from Carolina bays.
- 2) Hg concentrations in tail tips are higher than in blood samples collected from the same individuals.
- 3) Tail tip Hg concentrations in *N. taxispilota* varies spatially along the Savannah River, with highest concentrations near Lower Three Runs Landing, followed by Steel Creek Landing and Ellenton Bay Landing (which were similar), and finally the Jackson Landing upstream of the SRS.
- 4) Tail tip Hg concentrations of snakes collected from Carolina bays is low relative to published consumption limits and for values reported in literature for watersnakes from contaminated sites.
- 5) Historical Hg tail tip concentrations in *N. taxispilota* collected from Lower Three Runs Creek were four times higher than contemporary samples collected from the same area.

Major Impact(s) of Research

Collectively, our research to date indicates that Hg levels in high trophic feeding watersnakes overall have low concentrations relative to recognized consumption limits and no adverse effects were noted in snakes we collected. In at least one watersnake species, Hg levels were actually higher in individuals collected from isolated Carolina bays than from sites with known Hg contamination (i.e., former nuclear cooling reservoirs) – likely reflecting both Hg cycling and bioavailability in isolated temporary wetlands and dietary differences of snakes in the two major habitat types. Watersnakes, due to their propensity to biaccumulate and their small home range sizes, appear to be excellent indicators of local contamination.

Other Project Personnel

David Haskins, PhD Student – SREL M. Kyle Brown, MS Student – SREL Louise McCallie, Research Technician – SREL

External Collaborators

Dr. Robert Gogal (UGA)

Dr. Melissa Pilgrim (University of South Carolina-Upstate)

Products

- Haskins, D.L., R.M. Gogal, and T.D. Tuberville. 2019. Snakes as novel biomarkers of mercury contamination: a review. Reviews of Environmental Contamination and Toxicology 249:133-152.
- Brown, M.K. 2019. Bioaccumulation of mercury and radiocesium in *Nerodia floridana*: standard metabolic rate, hemoparasite infection, and interspecific comparisons to *Nerodia fasciata*. M.S. Thesis, University of Georgia.
- Haskins, D.L., M.K. Brown, and T.D. Tuberville. Brown watersnakes (*Nerodia taxispilota*) as bioindicators for mercury in the Savannah River. UGA's Interdisciplinary Toxicology Program Graduate Student Symposium, Athens, GA. April 2019. Platform presentation.
- Brown, M.K., D.L. Haskins, M.A. Pilgrim, and T.D. Tuberville. Interspecific comparisons of mercury bioaccumulation in *Nerodia floridana* and *Nerodia fasciata* from the Savannah River Site, SC. Warnell Graduate Student Symposium, UGA, Athens, GA. February 2019. Platform presentation.
- Haskins, D.L., M.K. Brown, M.A. Pilgrim, K. Meichner, R.M. Gogal, and T.D. Tuberville. Mercury bioaccumulation and body size influence peripheral blood leukograms in the brown watersnake (*Nerodia taxispilota*). Society of Environmental Toxicology and Chemistry, Sacramento California. November 2018. Poster.
- Brown, M.K., D.L. Haskins, M.A. Pilgrim, and T.D. Tuberville. Interspecific comparisons of mercury bioaccumulation in *Nerodia floridana* and *Nerodia fasciata* from the Savannah River Site, SC. Southeastern Partners in Amphibian and Reptile Conservation, Black Mountain, NC. February 2019. Poster.
- Lambert, M., K. Brown, D. Haskins, A. Russell, M. Pilgrim, and T. Tuberville. Sublethal effects of ¹³⁷Cs and Hg on Florida green watersnake (*Nerodia floridana*). Southeastern Partners in Amphibian and Reptile Conservation, Black Mountain, NC. February 2019. Poster.
- Cain, K.L., M.K. Brown, D.L. Haskins, M.A. Pilgrim, and T.D. Tuberville. ¹³⁷Cs whole body burdens and plasma biochemistry profiles of *Nerodia floridana* occupying a former nuclear cooling reservoir. 2018 Council on Undergraduate Research Meeting, Arlington, VA. October 2018. Poster.
- Research featured on "Hidden Biodiversity" episode of Coastal Kingdom environmental education television show. Full show available for viewing at: <u>http://coastalkingdom.com/episodes/hidden-biodiversity</u>

TASK 4. SREL public outreach and communication programs will focus on the SRS environment and ecological research to increase the public's understanding of scientific issues affecting the Site and to increase general ecological awareness

SREL Outreach Activities in FY19

SREL's public outreach and communication programs focus on habitats and environments on the SRS and the ecological research that is conducted by SREL, with the purpose of increasing public understanding of scientific issues affecting the site and bringing general ecological awareness to the general public. Historically, the program's mission has been to educate the public about ecological research and environmental issues. SREL has also worked with the Citizens Advisory Board, various onsite organizations, state and federal regulatory authorities, and other stakeholder groups to raise awareness of the SRS and of regional ecological issues and opportunities for environmental stewardship. The program highlights SREL's ecological research on the SRS through oral presentations, exhibits, tours, and various electronic media. Outreach programs facilitate and encourage participation by students, regional teachers, resident and visiting faculty and training programs. SREL actively communicates information to the media via UGA Public Affairs and local and regional media outlets. To accomplish these goals and provide an overall educational outreach program, SREL has focused on the following specific objectives since its inception:

A. Publish articles on environmental issues and ecological research in popular press outlets including newspaper columns, popular magazines, University of Georgia publications, Department of Energy publications, encyclopedias, special publications such as alumni magazines and ancillary publications of scientific societies.

B. Provide news releases to newspapers and other appropriate media that relate to environmental activities of SREL, with particular emphasis on the SRS.

C. Develop and present an on-site tour program that focuses on the environments of the SRS and the ecological projects of SREL —conveying SREL's role as an independent evaluator.

D. Give presentations to the public, including schools, civic groups, and other organizations that focus on environments of the SRS region and on SREL's ecological projects.

E. Develop portable and permanent exhibits appropriate for use at special presentations at SREL, schools, other organizations, and special events.

F. Develop video and multimedia shows for presentations to groups or for use by onsite organizations, emphasizing SREL environmental programs and projects on the SRS.

G. Investigate opportunities for broadcast programs that focus on environmental issues, SREL's ecological research, and ecological projects on the SRS.

H. Develop and distribute brochures and publications that are informative the public and on-site tenants of SREL's ecological research, and the environments on the SRS.

I. Develop and establish displays of SREL research projects in appropriate areas of the SREL facilities.

J. Publish an internal newsletter (*The GrapeVine*) as a means of enhancing internal communications—promoting individual as well as organizational achievement.

K. Develop the UGA conference center as a focal site for environmental education.

L. Establish a photograph collection that tells SREL's story, is informative of plants, animals, and habitats of the SRS region, and that emphasizes current ecological projects of SREL.

M. Maintain a collection of live plants and animals that can be used to educate the public about environmental issues and ecological research.

N. Maintain an area of the website for education on wildlife native to the SRS to include identification of regional species and information on wildlife safety.

O. Develop and present SRS wildlife safety talks for site tenants and visitors.

In accomplishing the goal of communicating ecological information to non-scientists, the Outreach program has provided on-site training and services to demonstrate the potential sources of injury from animals and plants found on the SRS and the CSRA that could occur to remote workers engaged in field activities or to employees and their families at home. The Outreach program has conducted workshops and training sessions and has attended SRS monthly safety meetings to deliver PowerPoint presentations and introduce live animals and native plants. The Outreach program has also developed and distributed safety materials (protocol badge cards and safety fact sheets) to SRS employees, and has managed an educational section on the SREL website. While the primary focus of most of these wildlife safety programs has been on snakes and alligators, the programs have also provided information on plants, insects, spiders, snapping turtles, and mammals of concern. During the 2019 fiscal year, the Outreach program hosted 10 safety talks for SRS employees and contractors.

The Outreach Program has been a participant in SRS's outreach to the general public via the *SRNS Public Tours* program, with SREL providing a 45 to 60-minute presentation bimonthly year-round (24 scheduled and up to 20 additional lab tours and impromptu presentations). These presentations provide a general introduction about the history and ongoing mission of SREL and the lab's involvement with research, teaching, and community service. The programs conclude with a question and answer period for participants on wildlife identification, site environmental research programs, safety, and other ecological matters of public interest. During the 2019 fiscal year, the Outreach Program hosted 27 tours, including 18 SRNS public tours and 9 lab tours and presentations for SRS partners.

SREL also hosts a series of scientific seminars, which are open to SRS employees, on a variety of research and educational topics which are pertinent to the SRS mission. Speakers include SREL research scientists, invited scientists from other university or agency programs, and graduate students who are conducting research on the SRS. During the 2019 fiscal year, the Outreach Program hosted 18 seminars, featuring current research from the SRS to the latest research in genetics, toxicology, herpetology, invasive species management, environmental monitoring, and radioecology.

Other programs in which Outreach personnel participate include: **REMOP**, a community outreach program designed to educate Burke County, Georgia, residents about environmental monitoring, including metals and radioactive elements; the *Let's Grow Together* pollinator program, a collaboration with the USDA Forest Service that provides pollinator gardens and education programs to local schools as well as encourage families to participate in outdoor adventures at National Parks, Historic Sites and federal properties through the *Every Kid in a Park* free pass program; *Ecotalk*, an opportunity for students to have nature brought into their classroom for a face-to-face lesson on a variety of live animals found in local habitats; the *Ecologist for a Day* program allows students to spend the day in the field gaining hands-on knowledge of the plants and animals of the unique Upper Three Runs Creek area at the off-site UGA Conference Center. The conference center also hosts civic group presentations and ecological tours. All

school programs incorporate science standards and curricula for particular school districts. Most of these programs provide an opportunity for participants to work with SREL staff as they catch, mark, and measure various species of reptiles, amphibians, fish, small mammals, and invertebrates. In addition, Outreach offers an annual free program, *Touch an Animal Day*, to the local and regional community at the H. Odell Weeks Activity Center, which allows individuals of all ages to interact with live animals and plant species, to meet site researchers, and to learn more about SRS efforts, including our research and education components. Lastly, the Outreach Program offers tours of SREL facilities, as well as exhibits and workshops for the general public as well as onsite personnel.

During the 2019 fiscal year, the REMOP program held 59 stakeholder, education and general public events in Burke County. The program brought together the Burke County community, Georgia Power, Savannah River Site scientists, local educators, and special interest groups. The Let's Grow Together program created four pollinator gardens and curriculum at local schools in areas that are traditionally underrepresented in STEM education. In addition, the Outreach program distributed more than 3,500 Every Kid in a Park passes in the CSRA region. The outreach staff presented 299 Ecotalks in area classrooms, hosted 36 Ecologist for a Day programs and participated in 26 CSRA Summer Library Programs. The annual Touch an Animal Day brought more than 1,583 visitors to the H. Odell Weeks Activity Center. The educational programs reached more than 63,000 students, teachers and members of the public in the CSRA region. During fiscal year 2019, SREL Educator Sean Poppy was honored with the South Carolina Environmental Awareness Award for his 20-years of service to the community.

The communications program has worked to increase the visibility of the SREL and its role on the SRS through traditional media such as newspapers, magazines, and regional/local TV and radio outlets. In addition, the communications program has greatly increased its digital presence on social media such as Facebook, Twitter, and other emerging social media platforms. During the 2019 fiscal year, the SREL outreach programs, press releases, media advisories, and staff appeared in 59 newspapers, magazines, television shows, and radio programs. The media coverage spanned local, regional, national and international outlets reaching millions of people. The SREL Facebook community grew to more than 1,353 followers and had more than 79,000 views of its content. The SREL Twitter community grew to 295 followers and had more than 569,900 views of its content. The SREL Instagram community grew to more than 500 followers and generated more than 56,000 impressions of its content.

The Outreach section of the SREL website, *https://srel.us.edu*, receives many visitors, as it has links to the popular *Ecologist for a Day* program, Outreach fact sheets and educational products, the *Ecoviews* weekly newspaper column. It also invites questions about wildlife native to the SRS that are answered by the Outreach personnel. This website is frequented by educators, bloggers, researchers and news outlets from all over the world, who use the materials in their classrooms, blogs, research and news platforms. SREL distributes thousands of educational products and materials worldwide to schools, organizations, and the general public. The website is consistently ranked in the top 10 among UGA's research affiliated websites.

TASK 5.SREL will maintain ecological data bases for use by the public, SRS, governmental,
academic, and private organizations. These databases incorporate more than 60 years of data
collection on the SRS and provide a resource for understanding changes impacting
ecosystems on the SRS and elsewhere in the southeastern United States

SREL Data Management Activities in FY19

IT Infrastructure

The goal of the Savannah River Ecology Laboratory (SREL) IT department is to provide an effective, reliable computer network that enables our employees to carry out their duties efficiently and at the same time protect their collective data. Over the past year, SREL conducted two major initiatives to improve our IT systems to meet the above stated goal.

1. Expanded use of cloud service Dropbox.

Historically SREL maintained a network of servers to store and secure data pertaining to SREL. This network was expensive and time consuming to operate, servers had relatively short effective service lives, and loss of data was a constant threat due to network issues or hardware failure. In an effort to mitigate these issues we began using Dropbox as a data storage alternative two years ago. This approach provided increased data security, the ability for SREL employees to access desktop and network data from anywhere in the world, and the cost per employee for data storage was less that using our own hardware constructed system. While there are some SREL data needs that will always require our own network servers, GIS data for example, our previous experience with Dropbox has led us to move the vast majority of our data storage needs to this cloud system. To implement this transition, we added another 50 Dropbox licenses to our current inventory this past fiscal year to give us the required storage capabilities.

2. Desktop computer replacement.

This past year at SREL we did an audit of employee desktop computers and this revealed that we had 48 employee computers that were between 5 to 8 years old. We were able to allocate funding in FY 19 to replace these 48 computers and provide these employees with new more reliable units. All of these units are warrantied for 5 years and acquiring these units is another step in ensuring an up-to-date, efficient, IT network.

SREL has taken steps in the last few years to build a secure and effective IT network that can be operated on a cost effective basis with limited IT support personnel. This year's accomplishments help us to continue to take steps in that direction.

Database Management

Responsible management of research data plays an important role in preserving SREL's institutional memory. Data archiving supports DOE's mission, contributes to future research ecological research, and is now often required by funding agencies. SREL's current challenge is to rebuild the technological and policy infrastructure to support an active archiving program, as well as to address existing legacy data.

During the course of FY19, the manuscript acceptance notification process established the previous year has proven to be a useful tool for gathering data archive submissions, complementing the checkout process. These files continue to be formatted and documented via the stand-alone data archiving template, until the data submission tool is complete, at which time they will be added to the searchable data portal. At present, the metadata entry tool remains in testing phase.

During FY19 SREL also continued work on legacy data in the existing archive. Manual checking of the most complete sets (403 projects) is complete. The unparsed, original files have also been added to each project as a zip archive folder. Where applicable, study locations were pulled from the datasets and added to the database. Reprint numbers were added to the bibliography database and linked to the matching data sets. The remaining sets, which were deemed incompletely documented (72 projects) or essentially undocumented (23) are now under review. The decision was made to not include taxonomic ID information for legacy data sets because the taxonomic concepts used by the original researchers are usually not known. However, the taxonomic database will be available in the metadata entry tool for use in future studies.

During FY19 SREL increased efforts to recover legacy data that may not have been (or are not yet) included in the legacy archive, including hiring a temporary data technician to assist with these tasks. The 1998 CD discovered in FY17 contained a large number of other directories as well as multiple copies of the archive; we have now been able to compare these files to the existing archive, finding several hundred that did not appear to be part of the archive. They are of various formats and essentially undocumented. Future work will determine whether headers can be recovered for those that do not have headers, and whether the files can be associated with existing SREL publications. Some initial efforts have also been made to examine stored paper files for important legacy datasets, although many of the items examined have proven to be the programs or program outputs rather than input data. In FY19 we were also able to collate database versions and digitize the bulk of locations for the SREL snake database, one of our well-known historic datasets that has not been archived. We are currently processing the remaining unknown locations. As a part for the process we have also been able to recover and digitize most of the sampling stations used in the Freeman herp survey, another famous SRS dataset. We plan to digitize and archive the full list of sampling stations, which were used for a variety of biological surveys conducted during the very early years of the SRP.

In support of one of the SREL Data Management Committee's longer-term goals, the SREL data manager been in discussion with two interested researchers, regarding proactive ways to promote good data-keeping practices within their programs.

TASK 6. <u>SREL will serve as the point-of-contact for the "DOE Research Set-Aside" areas that are</u> protected from site impacts so that they are available for environmental research and can serve to establish representative standards for comparison to impacted areas on the SRS. Currently SRS has 30 "set-aside" areas. SREL will also continue to promote the role of the SRS as a National Environmental Research Park.

SREL Set Aside and National Environmental Research Park Activities in FY19

The SRS's Set-Aside Program began in the 1960s when the Atomic Energy Commission (AEC) established 10 relatively small SREL Reserve Areas to represent the various habitats on what was formerly known as the Savannah River Plant and to secure study sites for conducting long-term ecological research. The program was expanded in the 1980s to 30 DOE Research Set-Aside Areas to better protect sensitive species habitats, preserve the biological integrity of Upper Three Runs Creek, and to buffer SREL's long-term research sites from forest management activities. These areas are a significant component of the SRS landscape (7% of SRS, totaling 14,560 acres/5,892 ha) and are found in 43 of the site's 89 timber resource compartments. There are approximately 275 miles (443 km) of posted boundary line. Set-Asides provide sites for long-term research, habitat for sensitive species, and protection for several archaeological sites.

<u>Administration and Management of the Set-Aside Areas</u> – SREL serves as the point of contact for the 30 Set-Asides and provides custodial oversight of the SRS Set-Aside Program. SREL chairs the DOE's Set-Aside Task Group, which approves management prescriptions, evaluates proposed ecological research, and ensures protection from onsite land use activities. SREL serves as the representative for the Set-Aside program in the SRS Site Use process and in the military training coordination meetings, reviewing activities in both venues for potential impacts.

<u>Set-Aside Oversight</u> – In recent years SREL has taken a more active approach to managing these areas, with wider application of prescribed fire, as well as some timber management. Management is conducted with an adaptive approach that gives the flexibility to address changing environmental conditions as well as research needs.

- Thinning has been prescribed for the two longleaf stands in the UGA Old Laboratory Site, which will be implemented as part of the Upper Three Runs North prescription. Though an active research site in the 1960s and 1970s, conditions have changed greatly since then. These stands are now too dense to support much groundcover, and the trees have a high incidence of fusiform rust.
- The last remaining peripheral drift fences associated with the Ginger's Bay Set-Aside Area were removed in January. These partial fences had been used to study the terrestrial distribution of salamanders around Ginger's Bay, important data for determining appropriate buffers around geographically isolated wetlands. (SREL reprint #3260.)
- Compartment 24 section of the E. P. Odum Wetland Set-Aside was burned.
- The Sandhills Fire Site SA, Mona Bay and Woodward Bay SA, and the Craig's Pond and Sarracenia Bay SA have all been placed on the burn schedule for the upcoming dormant season.
- USFS will repair the approach to Kennedy's Pond bridge. No issues are anticipated.
- Stream researchers from SREL and University of Kentucky met with the Set-Aside Task Group to discuss a possible stream restoration project in the E. P. Odum Wetland Set-Aside.

Current research in SRS Set-Asides

- SRARP continues to catalog artifacts previously recovered from Flamingo Bay SA. No additional excavations have occurred there this FY. Long term archaeological research at Flamingo Bay has provided a wealth of information on early inhabitants of the CSRA, as well as information on bay formation.
- Research on habitat use of state-endangered gopher frogs (*Lithobates capito*) continues at Craig's Pond and Sarracenia Bay SA.
- Researchers from SREL, USFS-SR, and the University of Kentucky continue stream characterization in the E.P. Odum Wetland Set-Aside. This research will be used to inform future DOE restoration and mitigation efforts.
- A tributary in the E.P. Odum Wetland Set-Aside was used as a reference site to assess effects of excessive stormwater runoff on aquatic insect emergence.
- Dragonfly nymphs from Flamingo Bay were used in studies assessing the effects of contaminated constructed wetlands on aquatic insects.
- Research examining trace element accumulation in beaver wetlands, which included two sites in the Ruth Patrick-Meyers Branch Set-Aside, has been published.
- Researchers collected aquatic insects from the E. P. Odum Wetland Set-Aside for a stream ecology station at SREL's annual lab-wide outreach event, Touch an Animal Day.
- The E. P. Odum Wetland, Organic Soils, Beech-Hardwood, Mature Hardwood, and Flamingo Bay Set-Asides provided field sites for a study of ecological factors affecting the success of rabies elimination in the southeastern US.
- Craig's Pond, Sarracenia Bay, Mona Bay, and Thunder Bay, as well as 9 non-SA wetlands in the central and northeast regions of the SRS, continue to be monitored as egg-laying sites for the stateendangered gopher frog, *Lithobates capito*, and as part of a regional southeastern phylogeographic study. Egg masses were only observed at Bay 58 in spring of 2018.
- Rainbow Bay, Ellenton Bay, Ginger's Bay, and Flamingo Bay continue to serve as reference sites for several amphibian ecotoxicology studies, including effects of copper in the Tritium Facility's H-02 Treatment Wetlands and metals uptake in the D-Area Ash Basin system.
- The amphibian community at the Rainbow Bay Amphibian Reserve Set-Aside has been monitored for 41 consecutive years, during which time local species extinctions, colonizations, and dramatic population fluctuations have occurred. Researchers are currently investigating how amphibian community changes and hydroperiod fluctuations over time have influenced nutrient fluxes between the wetland and upland habitats.
- SREL researchers continue collecting amphibian tissue samples from multiple wetlands for studies of amphibian landscape genetics and effects of future climate change. Samples from eight species have been collected from approximately 42 isolated wetlands across the SRS, including the following Set-Asides: Rainbow Bay Amphibian Reserve, Cypress Bay, Dry Bay, Ellenton Bay, Mona Bay and

Woodward Bay, Flamingo Bay, Thunder Bay, Craig's Pond and Sarracenia Bay, Ginger's Bay, and Road 6 Bay.

- Amphibian species in bay Set-Asides and other site wetlands are being monitored for two amphibian diseases of concern, chytrid and ranavirus, to determine disease prevalence on the SRS and possible relationships to contaminant distributions.
- Eastern green watersnakes and banded watersnakes were captured in the Craig's Pond and Sarracenia Bay Set-Aside to compare mercury, radiocesium (Cs-137), and blood parasite loads to levels in the same watersnake species captured in SRS cooling reservoirs.

<u>National Environmental Research Park Support</u> - SREL serves as the official SRS point of contact for the DOE National Environmental Research Park System. In its role as a point of contact, SREL conducts a variety of functions, one of which is the improvement and archiving of critical historical research data on the SRS. For more information on this subject, see the Data Management section elsewhere in this report.

In FY19, DOE-SR responded to SRS CAB Recommendation 362, which addresses support for the SRS NERP and Set-Aside Program. DOE-SR accepted the subpart addressing Set-Aside signage and boundary marking. DOE-SR partially accepted the subpart requesting updated biological inventories.

TASK 7.Through general research and public outreach programs, SREL will increase scientific
understanding in the general areas of environmental characterization, ecological risk
assessment, and environmental remediation and restoration. This will require research on
topics such as terrestrial and aquatic ecology, environmental chemistry, molecular ecology
and genetics, microbial ecology, radiation ecology, and ecotoxicology. SREL will also
continue to communicate and coordinate with SRS contractors and the public on these issues

In FY19 SREL faculty, staff, and students conducted and completed a diversity of outreach and education programs for the public and environmental research projects on the SRS in support of the missions of DOE (EM & NNSA) on the site. Specifically, outreach programs were conducted for local community residents on behalf of DOE as part of ongoing community education programs to increase environmental awareness of citizens and provide independent information to community residents relative to the activities of site tenants. In addition, specific research programs were conducted for NNSA to assess the environmental consequences of the Mixed Oxide Fuel Fabrication Facility on local stream quality and function as well as to assess the function, performance, and environmental consequences of constructed wetland treatment systems for metal sequestration associated with the NNSA Tritium facility on the SRS. These programs were funded by NNSA and the details of these projects are outlined below:

Research Support to DOE National Nuclear Security Agency

H-02 Constructed Wetland Studies— Biogeochemical Processes

<u>Funding Entity</u> NNSA - Tritium <u>Start Date and Funding Amount</u> October 2018; \$293,240 <u>SREL PI and co-PI's</u> Dr. Xiaoyu Xu **Objectives**

The goal of this research is to support, assess and improve operations of the NNSA constructed wetlands to maintain treatment efficiency and ensure long-term sustainability. Our primary objectives are to study: (1) the removal efficiency of metals for the studied wetland system, (2) the chemical speciation and bioavailability of metals in surface waters, (3) the stability of sediment accumulated metals and the potential for remobilizing sediment metals, (4) the overall biogeochemistry of metals in the sediment and water, and (5) the relative environmental impacts.

Summary of Research Activities

Metal biogeochemistry in the water

Water samples were collected and Diffusive Gradients in Thin Films (DGT) were deployed monthly in FY19 in the primary discharge pipes, retention basin, influent, effluent, both wetland cells, and the discharge stream that carries the effluent to Upper Three Runs. Water quality parameters (temperature, pH, oxidation-reduction potential (ORP), alkalinity, dissolved organic carbon (DOC), sulfate, and chloride) and concentrations of total, dissolved, and labile metals (Cu and Zn) were determined. The H-02 wetland system functioned well in FY2019. The overall removal efficiency of Cu and Zn are 71.7% and 78.9%. Water collected from effluent after being treated by wetland cells showed lower metal concentrations than the NPDES permit and were much lower than the influent. Meanwhile, the alkaline waters became nearly neutral after running through the wetland cells and ORP decreased significantly. There were not differences of metal concentrations and water quality parameters in the effluent between FY18 and FY19, indicating a relatively stable performance of the H-02 wetland. Metal speciation indicated the reduction of metal toxicity by the treatment cells. Although the major species in total metal were in dissolved phase, DGT-indicated labile metal concentrations were much lower in the stream compared to that in the influent. We did not

observe any increase in the percentage of DGT-indicated metals in total metals after water being treated, also demonstrating the good performance of the H-02 wetland in FY2019.

Metal biogeochemistry in the sediment

Multiple approaches were applied to understand metal biogeochemistry in the sediment in FY19: sediment cores were collected, and DGT and pore water samplers were deployed monthly in the two wetland cells. Sediment quality parameters (DOC, sulfate, and chloride in pore waters), total metal concentrations in bulk sediment, and labile metal concentrations in pore water and indicated by DGT were determined. Most metals were retained in the surface sediment and associated with the organic rich flocculent near the sediment-water interface, suggesting they were transported to the sulfide mineral layer that was deeper in the sediment. Similarly, the highest concentrations peaked in March (when the dominating sulfur reaction shifted from sulfide oxidation to sulfur reduction) and June (when more labile organic ligands were produced). Additionally, labile metal concentrations in deeper sediment (upper 14-24 cm) increased from June to August, demonstrating bioturbation transported labile metals from surface to deeper sediment. DOC and sulfate in pore waters changed seasonally: sulfate concentrations peaked in February which corresponds to the sulfur cycles; DOC concentrations peaked in May and June and they were lower in spring and summer than that in fall and winter. But we did not find out any temporal trend of chloride concentrations.

Controlled studies

We continued using DGT exposures coupled with aquatic organism bioassays to assess the use of DGT as a tool for monitoring metal contamination and estimating metal bioavailability in FY19. Musselaccumulated metals positively correlated to particulate metals in the wet season because feeding instead of filtering dominated the exposure pathway, and to both DGT-accumulated metals and modeled labile metals in the dry season when filtering was the main exposure route. Metal mixtures suggested similar internal regulations in the mussel where interactions between Cu and Zn did not impede their uptake and accumulation. But mussel's active regulation of metals enhanced its ability of elimination at the initial stage of deployment and it took at least >8 days to show a clear bioaccumulation pattern, suggesting a longer exposure duration in future study that parallels DGT and bivalve.

Mercury (Hg) in the wetland cells

Mercury (Hg) was included in the study of FY19. The only sources of Hg to H-02 wetland is the atmospheric deposition. We found out the wetland cells served as a huge trap of deposited Hg, and meanwhile anaerobic bacteria in the sediment transfer inorganic Hg to methylmercury and moves methylmercury from lower trophic position to higher trophic position through the wetland food web. Different Hg methylation was observed between the two wetland cells and between the H-02 and the A-01 wetland, which can be attributed to the different microbial communities. There is a potential risk that the wetland treatment cells become an important source of Hg to the surrounding environment. Further studies need to be conducted to monitor Hg methylation and track the sources of Hg in the wetland cells.

Conclusions

The H-02 constructed wetland effectively reduces total Cu and Zn concentrations in the surface water released by Tritium Facility to achieve NPDES regulatory limits. Meanwhile, metal toxicity was reduced, and water quality was improved in the water after being treated by the wetland cells. Most deposited metals were retained in the surface sediment, which may lead to decreased removal efficiency and performance when the surface sediment is saturated in the future. The metal-removal processes in both wetland cells are related to season and sulfur cycling. The wetland treatment cells are an important source of Hg to the surrounding environment. DGT technique, as a convenient and accurate passive sampler that indicates metal bioavailability, should be included in routine monitoring. But care needs to be taken when it is used in the wet season.

Major Impacts of Research

- 1) This research supports the use of cost-effective constructed wetlands for the treatment of metal contaminated wastewater and supports DOE's goal of employing "green technologies" for waste cleanup and remediation. Constructed wetlands play an important role in the SRS environmental plan to achieve both federal and state regulatory compliance for the discharge of effluent waters.
- 2) This research evaluates the potential transport of contaminants from constructed wetlands to surrounding environments and and supports DOE commitment to good ecological stewardship.
- 3) Results of our studies support the EPA's goal of advancing our understanding of metal biogeochemistry in wetland systems and developing better tools for predicting the fate and effects of metals in aquatic ecosystems.

Other Project Personnel

Erin Peck, Research Professional - SREL Cher Nicolson, Research Technician - SREL Anna Nieman, Research Technician - SREL

John Perry, Research Technician - SREL

External Collaborators

NA

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H-02 Constructed Wetland Studies: Amphibian Ecotoxicology

<u>Funding Entity</u> NNSA – Tritium <u>Start Date and Funding Amount</u> October 2018; \$324,498 <u>SREL PI and co-PI's</u> Dr. Stacey Lance and David Scott

Objectives

Our research at the H-02 constructed wetland complex focuses primarily on several questions related to these treatment wetlands: 1) Over time, what amphibians, reptiles, and plants have become established in the wetlands? 2) Do the elevated trace metal levels (e.g., copper and zinc) in the wetlands affect amphibian reproductive success, disease ecology, and population dynamics? 3) How do the amphibian diversity and numbers compare to other, more natural, wetlands? and 4) As the constructed wetlands age, how will the amphibian community respond?

Summary of Research Activities

This report summarizes our amphibian studies related to the H-02 treatment wetlands from October 2018 to September 2019. Following up on previous research, during this year we continued analyzing data on amphibian communities and disease prevalence from 20 wetlands on the SRS. We expanded our analyses to include boosted regression trees to deal with the very large data set and are currently writing a manuscript for submission to *Ecology*. Similarly, we continued work to examine the landscape genetics of two species, the Southern toad and Southern leopard frog, from across the SRS. We created genomic libraries for next-generation sequencing to capture single nucleotide polymorphism (SNP) loci from the capture probes we designed last year. We are currently analyzing the genetic data for ~ 600 individuals. From these data, we will be able to establish baseline levels of genetic diversity in both species and examine patterns of gene flow across the SRS. These data are critical for establishing whether the contaminated wetlands on the SRS, such as the H-02 wetlands, are acting as ecological sinks or traps. In addition to these site-wide studies, we undertook two new pilot studies to determine whether environmental DNA techniques can be used to assess the relative abundance and genetic diversity of amphibians in wetlands. To accomplish this, we conducted controlled laboratory studies with known abundances of southern leopard frogs and southern toads in aquaria and will compare how quantitative PCR and SNP analysis methods perform in estimating abundance. We also conducted a complementary study with southern leopard frogs in the H-02 wetlands where we used traditional catch per unit efforts to estimate abundance and we will compare how quantitative PCR and SNP analysis methods perform in estimating abundance. We continued to work towards publishing data collected in previous years and had two manuscripts published. Finally, we completed the 41st year of monitoring at RB, and finished analyzing the data in the context of community shifts in response to environmental change and subsequent impacts on nutrient cycling.

Conclusions

- 1) Ranavirus remains present in multiple wetlands on the SRS and poses a risk to cause population dieoffs.
- 2) Ranavirus prevalence is driven more by environmental factors, including canopy cover, air and water temperature than by community level factors, such as species richness.
- 3) Ambystomatid salamanders are highly sensitive to copper contamination under laboratory conditions, however one species, *Ambystoma talpoideum* has now colonized the H-02 wetlands.
- 4) At the A-01 wetland complex amphibian populations may have locally adapted within 5 generations to become tolerant to copper contamination.
- 5) The amphibian community at Rainbow Bay has shifted from long- to short-hydroperiod species over four decades in response to drought and associated shortened wetland hydroperiods.

6) Ignoring adult mortality in models of terrestrial flux of biomass leads to erroneous conclusions and the probability of terrestrial flux of nutrients due to amphibian movements is tightly linked to wetland hydroperiod and amphibian biomass, but not specific species.

Major Impact(s) of Research

- 1) Our continued time series of metal concentrations in the H-02 system (in sediments, water, and biota) will enable informed assessment of how this type of constructed treatment wetland functions, and whether it provides suitable wildlife habitat in addition to enhancing water quality.
- 2) Our *in situ*, mesocosm and laboratory studies demonstrate the importance of looking a) at multiple stressors, b) beyond the larval period, and c) at multiple source populations. We have found significant latent effects that lead to completely different conclusions than the larval study alone—effects on juvenile survival were apparent five months later, largely due to Cu effects on body size at metamorphosis.
- 3) Our disease studies are ongoing, but are demonstrating the complexity of variables involved with disease incidence and prevalence in amphibians. The nature of the wetland—metal-contaminated vs. clean, permanent vs. ephemeral, and constructed treatment wetland vs. natural— impacts disease prevalence and variables are confounded with each other.
- 4) Exposure to a stressful environment may lead to accelerated growth of tadpoles but that may come at a cost of weakened immune responses.
- 5) Our combined research at H-02, A-01 and D-Area are providing insight into how amphibian populations may locally adapt to metal contamination. Results from A-01 and D-Area suggest that within a few more generations amphibians at H-02 may be more tolerant of copper, however this may come with tradeoffs that reduce fitness in non-contaminated environments.
- 6) Our understanding of the factors that drive the population dynamics of amphibians in natural systems, based on the long-term RB study, will allow predictions of the effects of climate change on isolated wetlands and provide insights to land managers who may need to design protective measures for rare species.

Other Project Personnel

Dr. Krista Capps, Faculty – OSE/SREL Cara Love, PhD Student – SREL Dr. Jason O'Bryhim, Research Professional – SREL Adam McFall, Research Technician – SREL <u>External Collaborators</u> Dr. Scott Weir (Queens University)

Julie Ziemba (UGA)

Gabriela Rodriguez (University of South Carolina)

Adam McFall (University of South Carolina Aiken)

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H-02 Constructed Wetland Studies—Ecosystem Health

Funding Entity NNSA - Tritium Start Date and Funding Amount October 2018; \$152,812 SREL PI and co-PI's Dr. Gene Rhodes and Dean Fletcher

Objectives

The H-02 wetland water treatment system was constructed as a green technology initiative to remove metals and buffer pH in discharge waters from the NNSA Tritium Processing facility. Constructed wetlands have been widely employed to reduce nutrients and prevent eutrophication of surface waters. Their application for treatment of metal contaminated wastewater is still being refined. Our overall goal is to assess wetland effectiveness and develop strategies to maintain or improve treatment efficiency of the NNSA constructed wetlands to ensure long-term sustainability. Information is needed to evaluate: (1) the distribution of biologically available contaminants throughout and downstream of the system, (2) effects of disturbance events (e.g., storms) on wetland effectiveness, (3) the potential export of metals to terrestrial or downstream food webs by aquatic biota, (4) effects of daily discharge fluctuations on wetland efficiency, (5) the potential of managing the retention basin as a pre-filter for the treatment cells. Previous work indicated considerably higher levels of biologically available Cu below the wetland than expected. Efforts are focusing on identifying the conditions under which metals leave the wetland system and on developing strategies to improve wetland treatment efficiency.

Summary of Research Activities

Metal Mobilization by Stormwater Flows

Previous work found significant transport of metals during storm events. In FY 19, we further evaluated the consistency of these patterns. Such events have the potential of being a factor contributing to the higher than expected levels of bioavailable metals below the wetland treatment cells. ISCO sampling units equipped with 24 bottle carousels are in operation immediately above and below the treatment cells and were used to collect high resolution time series of water samples along with water quality data from associated multi-parameter probes. Methodology had previously been established by combining data from sampled water with water quality data collected from the multi-parameter probes. Sampling units were triggered by rainfall events when at least 0.3 cm of rain fell in 45 minutes or less. Samples were simultaneously collected from the treatment cell influents and effluents. We collected hourly composite samples composed of subsamples taken every 15 minutes; this evens out any momentary spikes uncharacteristic of overall discharge. Such a spike could result from an unusually large amount of suspended solid being sucked into the intake tube during a single sampling. Concurrently the multiparameter probes collected rainfall, discharge and water quality parameters every 15 minutes. These measures were averaged to coincide with the hourly water sample composite. In early summer, 192 water samples were collected and processed. Total and dissolved metal (Cu and Zn) concentrations were determined for each element, thus a total of 384 samples analyzed. Flux is the mass of an element that is entering or leaving the wetland at a point in time. In our case, it was calculated from metal concentrations in the hourly samples and average discharge during the hourly sampling. Load is the amount of element passing a point over an interval of time. Because of the high resolution of our sampling, we simply added the hourly flux measures to determine the load of element entering or leaving the treatment cells over the 96-hour sampling period. Water samples were also filtered to determine the total suspended solids (TSS) in each sample.

Water was sampled for 4 days during and after a substantial storm event in June over which 7.38 cm (2.90 inches) of rain fell after an extended dry period. We sampled for a longer than usual time period, because of continued rain, high water levels in the retention basin, and the resultant sustained high discharge level. In this event, 1.83 cm (0.72 inches) of rain fell during the first two hours with the 24-hour total only

increasing to 1.86 cm. A total of 0.06, 1.40, and 4.05 cm (0.02, 0.55, and 1.59 inches) fell on days 2, 3, and 4, respectively. A "first flush" resulted from a rapid rise in discharge after the initial rain event. Discharge ranged from 0.024-0.027 m³/sec during the 24-hour period before the first rain event after which discharge raised to 0.035 m³/sec, then discharge peaked at 0.040 m³/sec on day 4. During 96 hours after rain started, treatment cell influent total Cu (tCu) concentrations averaged 28.56 ppb (18.04-72.49), whereas effluents averaged 11.13 ppb (8.93-21.87). The primary first flush of tCu lasted about 2 hours. During the first two hours, influent tCu concentrations were elevated to 72.17 and 72.49, whereas effluent concentrations were only elevated to 21.87 and dropped to 13.00 ppb by the second hour. During the following 94 hours tCu effluent concentrations only exceeded 14 ppb in three hourly samples. Elevation of both discharge and concentrations influenced the hourly fluxes and the 96-hour load. During the first 2-hour flush, 16.7 g of TCu entered the wetland cells with 3.9 g leaving in the effluents. During the first 24 hours, a total of 110.1 g of tCu entered the wetland cells and 39.1 g left. Despite more rain falling between 72 and 96 hours, only 61.3 g entered the wetland cells and 31.3 left. Similar to our previous work, this illustrates the importance of the first flush after an extended dry period. Over the duration of the 96-hour period, a total of 322.9 g of tCu entered the treatment cells and 126.6 g left. FY20 efforts will compare this to base flow fluxes and to other constructed wetlands as available in the published literature. A pulse of suspended solid materials leaving the retention basin during the first flush is evident by an average TSS of 71.6 mg/L during the first two hours, but only an average of 10.2 mg/L during the next 94 hours. TSS was on average reduced in treatment cell effluents, but was still elevated to 54.0 mg/L during the first flush followed by an average of 4.08 mg/L during the following 94 hours. These fluctuations in TSS caused significant changes in the form of Cu entering and leaving the treatment cells during the 96hour sampling event. During the first 2-hour flush, 43% of the Cu entering the treatment cells was dissolved, thus 57% of the Cu was associated with particulate materials. During the next 94 hours, only 19% of the Cu was associated with particulates. Concurrently in the treatment cell effluents, 40% of the Cu was associated with particulate materials in the first flush, but only 11% in the following 94 hours. Treatment cell efficiency was calculated from the tCu load entering the treatment cells versus that leaving. During the first flush, 76% of the tCu was removed from the effluent line by the treatment cells. This subsequently decreased to 60% over the next 94 hours.

Total Zinc (tZn) concentrations of the first two hourly samples were 172 and 183 ppb in the wetland cell influents and 33.4 and 14.8 ppb in the effluents. During the next 94 hours, tZn averaged 63.2 ppb in the wetland cell influents, but only 7.55 ppb in the effluents. Over the 96 hours, tZn in wetland cell effluents averaged 91.8, 64.1, 53.8, and 52.6 ppb in days 1, 2, 3, and 4 respectively, while effluents averaged 10.0, 6.67, 6.55, and 8.29 ppb. During the first 2-hour flush, 41.1 g of tZn entered the treatment cells, and 5.4 g left. During the entire 96-hour period, 744 g entered and 90.1 g left the treatment cells. Similar to Cu, much of the tZn was associated with particulates during the first flush (averaged 70% in influents and 53% in effluents), but during the next 94 hours this reduced to averages of 46% and 24% in the influents and effluents, respectively. Removal efficiencies of tZn based on load was 87% during the first flush and 88% during the next 94 hours, thus a large portion of the tZn was removed by the treatment cells. During the first hour of the first flush, TSS in the wetland cell effluents exceeded that of the influent (influent 95.69 vs 80.62 mg/L). Thus, more suspended solids were leaving the wetland cells than entering indicating a flushing of materials from the wetland cells. However during the next 95 hours, TSS in the effluent only exceeded that in the influent in 5 hourly samples. Despite the wetland cells being a net exporter of suspended solids during acquisition of these individual hourly samples, across the all 96 of the hourly samples, the hourly load of both tCu and tZn was higher in the wetland cell influents than effluents. Thus, even though significant amounts of metals left the wetlands in the effluents, more metals were entering than leaving the wetland cells. Consequently, the wetland system was functioning to lessen impacts on the receiving stream.

On the first, third and fourth 24 hour periods of the June stormwater sampling event, three grab samples were collected from each of the pipes that release water into the retention basin. TSS were below 2 mg/L

in all samples. Thus, this preliminary assessment suggests that TSS entering the wetland cells are solids being remobilized from the retention basin, not from the retention basin influent pipes. Whether metal concentrations of retention basin influents or effluents were highest differed by pipe and day. These patterns will be further evaluated with additional sampling in FY20. Treatment cells appeared to be overloaded over the course of the storm event. The levels of TSS and metal fluxes leaving the retention basin may indicate better management of the retention basin is warranted. Much work remains to be done evaluating effects of factors such as level of rainfall, time since last rainfall, as well as seasonal variability. Since water and sediment chemistry changes seasonally this could greatly influence what leaves the wetlands during storm events. The metal load of the suspended solids also must be better evaluated.

Metal Transport by Emerging Macroinvertebrates

We are continuing our work examining the export of contaminants from the wetlands to terrestrial food webs by emerging insects. In addition to analyzing whole-body concentrations of metals/metalloids in teneral (emerging) dragonflies, we are also analyzing concentrations in exuviae (nymph exoskeleton) that is shed upon emergence. The latter indicates what proportion of the nymph's contaminant load was shed in the exuviae and left behind compared to what actually left the wetlands in the emerging teneral. Our previous work established that whether elements were being exported from the wetland by emerging dragonflies or shed in their exuviae depended upon element and varied by species. This was expanded by the evaluation of emerging damselfies. Trace element concentrations were analyzed in a total of 151 teneral damselflies and 63 exuviae. Analyzed teneral damselflies of the family Coenagrionidae were distributed across 2 genera, and 4 species. Some elements (e.g. Al, Fe, Mn, Pb) were largely shed in the exuviae, whereas others tended to leave the wetland in the teneral (e.g. Cu, Zn, Mg, B). However, the proportion of trace elements accumulating in/on exuviae versus teneral differed among species, but similarities within genera were observed. Both species of Ishnura consistently shed higher concentrations of all elements than the two Enallagma species. This was not dependent upon body, but some aspect of their biology. Using these data, variation among sites and habitats will be evaluated in FY20. Overall, trace element body burden of nymphs are indicative of trophic doses when preved upon directly, but not necessarily to that of predators preying on emerging damselflies. Work by Xu et al. in the H-02 wetland system has established changes in water chemistry during the plant growing season which corresponds to the dragonfly and damselfly flying season. It is possible that these changes in water chemistry could cause corresponding changes in accumulation in the odonates. Our previous work analyzed dragonflies and damselflies largely from a single time-period. Sample preparation has begun to evaluate whether the proportion of contaminants shed with the exuviae changes throughout the season. These analyses will be completed in FY20.

Factors Influencing Metal Accumulation in Macroinvertebrates

Different taxa of dragonfly nymphs stratify vertically in wetlands. Our work conducted in streams and wetlands frequently found taxa associated with sediments, particularly fine sediments rich in organic matter, tend to accumulate higher concentrations of metals/metalloids. For example, the genus *Anax* that typically lives in aquatic vegetation well above bottom sediments accumulated lower Cu concentrations than *Eythemis* that is more closely associated to bottom sediments. We collected 100 *Anax longipes* 100 *Erythemis simplicolis* from uncontaminated reference wetlands and held them in cages on frames in H-02 retention basin. Equal numbers of each species were held at four water depths ranging from on the sediment to 75 cm above sediment which corresponded to just under the water's surface. Nymphs were held for two months and then removed for trace element analyses. Water was sampled at each depth with water diffusion samplers and surface sediments were sampled. Evaluation of these samples will clarify whether previously observed differences are due to exposure to contaminated sediment or biological/physiological differences between taxa. Sample processing is in progress and will be completed in FY20.

Conclusions

- 1) Combined with FY18 information, severity of the storm influenced outfall discharge, metal concentrations, and consequently fluxes of metals that entered and left the treatment cells.
- 2) During more severe storm events, elevation of both metal concentrations and discharge rate caused a more substantial metal load to enter and leave the wetlands. Wetland efficiency is reduced during these more severe events.
- 3) Metal concentrations and fluxes were generally most elevated in a "first flush" that occurred early in the storm event. Higher concentrations of suspended solids leaving the wetland during the first flush resulted in a greater portion of tCu being associated with particulate material. During later hours, a larger portion of the tCu was dissolved. Wetland metal removal efficiency also changed over the course of the storm events.
- 4) Even though substantial amounts of metals left the wetlands, overall greater masses of metals entered the treatment cells than left in the effluents. Thus in the evaluated storm events, the H-02 wetlands functioned to lessen impacts on the receiving stream. Preliminary analyses indicate mobilization of sediments in the retention basin during storm events. Future work will continue evaluating whether the heavy loads entering the treatment cells during storms are originating from the retention basin or water entering it.
- 5) Elements differed in their propensity to accumulate in dragonfly and damselfly exuviae that are shed during emergence and remains in the wetland versus being transferred to terrestrial food webs by the tenerals.
- 6) Concentrations of trace elements accumulating in nymphs are indicative of trophic doses when preyed upon directly, but not necessarily to that of predators preying on emerging dragonflies.

Major Impacts of Research

- This research supports the use of cost effective constructed wetlands for the treatment of metal contaminated waste water and supports DOE's goal of employing "green technologies" for waste cleanup and remediation. Constructed wetlands play an important role in the SRS environmental plan to achieve regulatory compliance for the discharge of effluent waters.
- Our research evaluates the potential transport of contaminants from constructed wetlands to downstream waters or terrestrial environments and supports DOE commitment to good ecological stewardship. Evaluations are aimed at developing management strategies to maximize wetland effectiveness.
- 3) Overall, identification of factors reducing constructed wetland efficiency will allow us to prescribe and ultimately implement management strategies to maximize wetland efficiency.

Other Project Personnel

Christina Fulghum, Research Technician – SREL Paul Stankus, Research Professional – SREL Erin Spivey, Research Technician – SREL Jennifer Dirks, Research Technician – SREL Dr. J Vaun McArthur, Emeritus Senior Ecologist – SREL Dr. Fanny Coutelot, Postdoc – SREL Angela Lindell, Research Professional – SREL External Collaborators

Dr. Brian Bledsoe (UGA)

- Fletcher, D. E., A. H. Lindell, J. C. Seaman, P. T. Stankus, N. D. Fletcher, C. D. Barton, R. A. Biemiller, and J V. McArthur. 2019. Sediment and Biota Trace Element Distribution in Streams Disturbed by Upland Industrial Activity. Environmental Toxicology and Chemistry 38:115-131.
- Fletcher, D. E., A. H. Lindell, P. T. Stankus, N. D. Fletcher, B. E. Lindell, and J V. McArthur. (in press). Metal Accumulation in Dragonfly Nymphs and Crayfish as Indicators of Constructed Wetland Effectiveness. Environmental Pollution.

Restoration of the MOX stream (Tributary U8): Pretreatment Assessment

Funding Entity

NNSA - MOX <u>Start Date and Funding Amount</u> May 2018; \$213,645 <u>SREL PI and co-PI's</u> Dr. Olin E. Rhodes Jr. and Dean E. Fletcher

Objectives

Our overall goal is to provide a comprehensive assessment of the Upper Three Runs tributary (U8) that is located beside the MOX construction site. Contaminants accumulating in stream sediments and biota are being assessed as well as impact on hydrologic, geomorphic, and biologic stream features. The potential of restoration or enhancement strategies that would improve and better protect the integrity of the system are being evaluated.

Summary of Research Activities

Integration of our work on tributary U8 into additional SRS stream studies has expanded the geographic scope as well as incorporated additional critical expertise into our collaborative effort. The NNSA-MOX funded study, while providing a nucleus for the U8 work to be built around, has benefited by contaminant analyses that placed it into a broader spatial perspective with comparisons to other stressed SRS streams. Use of the same study taxa and environmental media along with identical laboratory and field protocols has allowed seamless comparison of data among these efforts. Overall, our contaminant assessments have included five streams in the Upper Three Runs basin along with the main stem of Upper Three Runs. The latter evaluates whether U8 is increasing contaminants in the main stem of Upper Three Runs. Geomorphic and hydrologic analyses of these streams have also been conducted in collaboration with the University of Kentucky and funded by the USDA Forest Service-SR in a project aimed at setting up a mitigation bank for SRS headwater streams. The restoration potential of U8 is being evaluated and a pre-treatment baseline being established.

Evaluating impacts of environmental contamination is an essential component of planning stream and riparian restorative work. Previous work established that contaminants were not accumulating in sediments of the downstream reaches of U8. The stream bottom was severely scoured of organic matter, fine sediments and associated metals/metalloids. Despite this, several contaminants were elevated in aquatic biota that integrate contaminant exposure over time and from multiple sources. Thus, contaminants could be pulsing through the system from upstream. To evaluate potential upstream sources and to thoroughly establish a pre-restoration baseline of contaminant distributions, a total of 165 sediment samples were collected from the perennial, intermittent and ephemeral channels as well as all basins in the U8 system. Cs-137 was analyzed using an auto-gamma counter, total mercury concentrations with a direct mercury analyzer, and 18 trace elements (Be, Mg, Al, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Sr, Cd, Ba, Pb, and U) by inductively coupled plasma mass spectrometry. Organic matter and clay content was determined for each sample and water velocity recorded at each sample location. Cs-137 concentrations were negligible in all samples. Results showed that trace elements were still largely scoured from sediments in the perennial stream downstream of the riprap weir below the west-side outfalls at the MOX construction site. However, concentrations tended to slightly increase in the depositional segment on the UTR floodplain. Even though the riprap weir does not have sufficient water retention time to prevent structural downstream damage to the stream channel, most elements accumulated immediately above the weir, with an area of scouring upstream of the west-side outfalls. Highest concentrations of several elements including As, Hg, Zn, Pb, and Ni tended to accumulate upstream of the MOX construction site, particularly in areas of the F Area coal ash basin. It appears coal combustion waste had been distributed from the vicinity of the basin. Many elements were also elevated in the older (compared to most active basins in the U8 drainage) North SWDF Sedimentation basin that has had more time to accumulate elements. Other basins have successfully accumulated elements with

basins differing in accumulated elements. Overall, industrial activities and engineering of the valley have resulted in broad distribution of several metals/metalloids. Restorations efforts will need to be designed to prevent further distribution of contaminants from these sources.

Many aquatic insects have complex life cycles that involve an immature aquatic nymph or larval stage that emerges from the water as a flying adult. This emergence connects aquatic and terrestrial food webs and results in aquatic habitats subsidizing food resources to terrestrial organisms. Consequently, stream disturbances that impact stream macroinvertebrate communities have the potential of also impacting the surrounding terrestrial food webs. Our previous work established impacts to the aquatic biota. We are now assessing whether these impacts are influencing the number and taxonomic composition of aquatic insects emerging from tributary U8. Traps to capture insects emerging from streams were first designed, and then their effectiveness tested. Ten traps were deployed in each tributary U8 and Tinker Creek reference tributary TC5. Traps were deployed over a total of 8 weeks with 28 trapping days. All insects were identified and counted. Over 6 times more emerging aquatic insects were captured in the TC5 traps than those in U8. Additionally individuals from TC5 were distributed across 18 families versus 9 in U8. Ephemeropterans (mayflies), plecopterans (stoneflies), and trichopterans (caddisflies) are collectively referred to as EPT and are known to be sensitive to stream disturbance. Consequently, the proportion of the community composed of EPT taxa is a sensitive indicator of stream disturbance. EPT taxa made up 49% of the individuals emerging from the reference stream TC5, but only 26% of those in U8. These analyses indicate an adverse effect of stream disturbance on insect emergence.

Locations were identified to place stream gaging stations in tributaries U8 and U6 (below outfall F-02) in 2020. A detailed evaluation of stormwater flows from these stations will provide the necessary assessment to allow calculation of the size of sedimentation basins required to sufficiently protect these streams from further structural damage. New collaborations were established to bring in expertise from the Institute for Resilient Infrastructure Systems in the University of Georgia's College of Engineering to make these calculations and model how different water retention scenarios will protect the integrity of the downstream stream channel. Future efforts will provide a design for the needed structures. **Conclusions**

U8 is severely degraded with impaired hydrology, channel form, substrate composition, and biological communities; impacts on stream macroinvertebrate communities and life histories have been observed.

- 2) Scouring by excessive flows has reduced organic matter content in the streams, influencing contaminant dynamics and potentially stream energetics.
- 3) Stream disturbance is reducing aquatic insect emergence, and thus aquatic to terrestrial food subsidies.
- 4) Evaluation of sediments throughout the U8 drainage indicated no concern of Cs-137 contamination.
- 5) Trace elements were scoured from the perennial stream, but upstream sources exist.
- 6) Highest concentrations of many trace elements were generally found upstream of the MOX construction site and associated with the area around the coal combustion waste basin or and an older sedimentation basin.
- 7) Restoration efforts will be designed to reduce further redistribution of contaminants from this area.

Major Impact(s) of Research

- We are assessing potential physical and biological impacts of the MOX-site construction and other activities in the U8 drainage and providing a comprehensive baseline of its present condition including contaminant distributions.
- 2) Extensive contaminant analyses combined with the physical characterization are helping distinguish whether contaminants are involved in the observed biological impairments or the impacts are primarily the result of excessive runoff.
- 3) A group of onsite and offsite collaborators are completing necessary evaluations to develop a successful restoration plan for tributary U8.

4) Overall, identification of specific stream impairments and establishment of baseline conditions will provide the foundation to restore the U8 system and subsequently monitor restoration effectiveness.

Other Project Personnel

Paul Stankus, Research Professional – SREL Christina Fulghum, Research Technician – SREL Erin Spivey, Research Technician – SREL Jennifer Dirks, Research Technician – SREL John Seaman, Senior Scientist – SREL **External Collaborators** Christopher Barton (University of Kentucky) Brian Bledsoe (UGA) Roderick Lammers (UGA) Daniel Buhr (UGA) James Fudge (SRNS) Andy Horcher (USDA Forest Service)

- Fletcher, D. E., A. H. Lindell, J. C. Seaman, P. T. Stankus, N. D. Fletcher, C. D. Barton, R. A. Biemiller, and J V. McArthur. 2019. Sediment and Biota Trace Element Distribution in Streams Disturbed by Upland Industrial Activity. Environmental Toxicology and Chemistry 38:115-131.
- Fletcher, D. E., B. E. Lindell, A. H. Lindell, P. T. Stankus, N. D. Fletcher, J V. McArthur, and J. C. Seaman. 2019. Basins, beaver ponds, and the storage and redistribution of trace elements in an industrially impacted coastal plain stream on the Savannah River Site, SC. Environment International 133: 105174. 11 pp.

Support for SREL Environmental Missions on the SRS

Funding Entity NNSA-MOX Start Date and Funding Amount March 2018; \$285,355 SREL PI and Co-PI's Dr. Olin E. Rhodes, Jr. and P.J. Perea

Objectives

SREL will assist the SRS NNSA mission and MOX Project by educating the public through community outreach activities that include organizing tours and exhibits featuring the local ecology and associated research; conducting environmental education workshops for teachers, students, and the general public, as well as for site personnel; development of a variety of environmental education materials for diverse audiences; increasing internet accessibility of information; distributing ecological information; presentation of data and reports on the local and regional environment; and assisting in educational efforts about the importance of environmental stewardship and National Environmental Research Park (NERP) programs at the SRS. Accomplishments relative to these tasks will be summarized in an annual report. **Summary of Program Activities**

The SREL Environmental Outreach Program utilizes information from SREL's ongoing research and long-term research efforts to provide training and services to MOX and other SRS employees. The program also educates the public locally, regionally, and nationally about ecological research findings associated with on-site activities.

NNSA has continued to provide critical funding that has facilitated SREL's ability to achieve the goal of maintaining informative outreach programs for SRS personnel and stakeholders, as well as programs that educate the public through outreach. These programs enhance an individual's understanding of environmental issues affecting the SRS and increase general ecological awareness. In fiscal year 2018, SREL conducted engaging educational presentations to K-12 schools and adult audiences. These outreach efforts achieved DOE and NNSA goals of enhancing the public's knowledge and understanding of the ecological health of the SRS, and the importance of environmental stewardship and the National NNSA has continued to provide critical funding that has facilitated SREL's ability to achieve the goal of maintaining informative outreach programs for SRS personnel and stakeholders, as well as programs that educate the public through outreach. These programs enhance an individual's understanding of environmental issues affecting the SRS and increase general ecological awareness. In fiscal year 2018, SREL conducted engaging educational presentations to K-12 schools and adult audiences. These outreach efforts achieved DOE and NNSA goals of enhancing the public's knowledge and understanding of the ecological health of the SRS, and the importance of environmental stewardship and the National Environmental Research Park (NERP) programs on the SRS. SREL also distributed educational resource materials to facilitate these goals.

The Outreach Program was readily available to conduct Lunch and Learn presentations to site personnel at the MOX Facility, provided tours for DOE site interns, new MOX employees, and children of SRS personnel. SREL developed and distributed literature and developed displays on animals and plants native to the SRS and the surrounding communities, produced materials on specific research programs, and maintained the MOX Conservation Garden at the facility.

The SREL Outreach Program is designed to enhance SREL's overall mission of acquiring and communicating environmental knowledge and to highlight NNSA's and DOE's focus on environmental issues on the SRS. To accomplish these goals, education initiatives are used that include 1) Environmental Safety on the SRS, 2) On-site outreach to the general public and site personnel, and 3) Off-site outreach activities at schools, civic organizations, and community events. Issues as diverse as

wildlife safety in the field, wildlife population declines, potential responses of organisms to contamination, distribution of abundance of sensitive species, impacts of invasive species, wetland dynamics and remediation, water quality, and dispersal of organisms from radioactively or chemically contaminated sites are all important to on-site personnel and the general public.

Major Impact(s) of Program

- 1) SREL has been heavily involved in assisting the MOX project by facilitating the environmental component of the LEED (Leadership in Energy and Environmental Design) certification requirements.
- 2) SREL's Outreach program has consistently communicated information about the environmental activities on the SRS based on ecological research conducted on the site by SREL scientists. Regardless of format presentations, tours, or exhibits, the SREL Outreach program's numerous efforts continue to succeed in raising awareness through education of the rich ecological diversity of the SRS MOX area and the region. The program continues to succeed in developing the general public's knowledge of and appreciation for the fauna and flora native to the area and in so doing develops an active interest in environmental stewardship, and an interest in protecting the integrity of the environment on the SRS and the communities that surround it.

Other Project Personnel

Vicky Sutton-Jackson, Public Relations Coordinator - SREL

Sean Poppy, Outreach Coordinator - SREL

Amanda Hurst, Animal Caretaker – SREL

Judy Greene-McLeod, Research Professional – SREL

Megan Winzeler, Outreach Coordinator – SREL

Products

- 1) Conducted 27 scheduled tours; number of attendees 534 (includes 18 SRS Public Tours, attendees 408; 9 tours for on-site employees/visitors, attendees 126)
- 2) Provided 10 Wildlife Safety talks; number of attendees 2,817 (includes 7 talks to SRS employees, number of attendees 2,757; 3 talks to professional groups, number of attendees 60)
- 3) Presented 299 STEM classroom education programs for elementary and secondary students; number of attendees 17,009
- 4) Presented 48 environmental programs for college, professional and adult audiences; number of attendees 1,793
- 5) Provided 34 STEM exhibits at local and regional events; number of attendees 31,693
- 6) Conducted 36 Ecologist for a Day programs (school field trips to SREL's Conference Center); number of attendees 1,172
- 7) Conducted 1 Touch an Animal Day event (August 24, 2019 at H. Odell Weeks Activity Center, Aiken, SC); number of attendees 1,583
- 8) Provided 59 REMOP programs for the public: number of attendees 4,499
- 9) Provided 26 presentations at regional library summer reading programs –number of attendees 2,145

*Total Outreach events: 541; total estimated attendance: 63,245

Assessing Effects of Copper Exposure in Model Fish and Insect Species

<u>Funding Entity</u> NNSA <u>Start Date and Funding Amount</u> October 2018; \$60,000

SREL PI and Co-PI's

Dr. Ben Parrott and Dr. Guha Dharmarajan

Objectives

Metal contamination resulting from industrial use has the potential to impair water systems throughout the United States. Impacts to biota in these systems are often predicted using toxicological exposure models that do not fully capture complex interactions between environmental factors (e.g. temperature variability) and/or are limited to predicting bioavailability and uptake. The H-02 treatment wetland system on the Savannah River Site is designed to remove metals from waters discharged from the NNSA Tritium Processing Facility. Copper and zinc levels remain elevated in this system and understanding their sublethal effects on organismal health and fitness remain a concern. Goals designated for the project involve two lines of research: (1) aquatic vertebrates and (2) aquatic invertebrates as biomonitors and models of environmental health

Summary of Research Activities

Aquatic vertebrates

The experiments described in the proposal have been completed and resulting data are currently being analyzed. Briefly, two independent cohorts of medaka embryos were exposed to copper sulfate (10 ppb and 100 ppb) across a range of ambient temperatures (27°C, 30°C, and 33°C). Hatch rates, time to hatch, and absorbed Cu levels were tested and recorded. Further, subsets of exposed fish were reared to 4 months post fertilization at which point growth rates were recorded and fish underwent fin regeneration challenges. Fish were subsequently necropsied and gonadal, hepatic, and CNS tissues were collected for telomere measurements, gene expression assays, and gonadal histology. Data is currently being analyzed.

Aquatic invertebrates

Experiments have been completed to test the fitness effects of copper exposure on Aedes aegypti, the model invertebrate chosen for this study. To test for effects of copper on mosquito development, mosquito eggs were synchronously hatched in three replicated containers with water containing copper (as CuSO₄) at 0, 300, 600, 1200, 2400, 4800 ppb. Preliminary analyses of the data revealed that copper had surprisingly negligible effects on mosquito development, with significant negative effects on pupation rates only evident at > 1200 ppb. To test for copper accumulation dynamics we carried out a separate experiment in which mosquitoes were reared in replicated containers at three sublethal concentrations of copper (0, 300, 600 ppb). We sampled live mosquito larvae, pupae, pupal husks and adults, and stored all samples at -20 °C. The tissue samples have been freeze dried and will be tested for copper concentrations using the ICP-MS. Finally, to test for sublethal effects of copper on adult mosquitoes, we raised mosquito larvae at three sublethal concentrations of copper (0, 300, 600 ppb). Adult mosquitoes were blood fed using dog blood that was infected and uninfected with filarial parasites. We quantified the effects of copper exposure on daily mortality hazard of adult female mosquitoes, as well as fecundity and vectorial capacity (i.e., the proportion of larval filaria developing to infectious stages in the mosquito). Data for all experiments have been collected and currently are being analyzed. Conclusions

Data for both projects are currently being analyzed and conclusions are not yet available.

Major Impact(s) of Research

1) The work seeks to address the impacts of copper exposure under ecologically relevant temperature regimens on endpoints associated with health and reproduction in aquatic vertebrates.

2) This work also will improve our understanding of the impacts of heavy metals on invertebrate populations, and can further refine our understanding of the parameters that need to be assessed when using invertebrates as bioindicators of ecosystem health.

Other Project Personnel

Emily Bertucci, PhD Student – SREL Faith Leri, Research Technician – SREL Marilyn Mason, Research Technician – SREL Erik Neff, MS Student – SREL Shannon Gregory, Research Technician – SREL <u>External Collaborators</u> NA

Products

Neff, E., M. Tanelus, and G. Dharmarajan. 2019. Effects of Copper Contamination on Mosquito Development, Fitness, and Vector Competence. SREL Graduate Student Symposium. July 19, 2019. Aiken, SC.

Tritium Distribution and Cycling on the Savannah River Site

<u>Funding Entity</u> NNSA <u>Start Date and Funding Amount</u> October 2018; \$78,000 <u>SREL PI and co-PI's</u> Dr. John C. Seaman

Objectives

The objective of the current project is to develop and refine monitoring protocols for evaluating organically bound tritium (OBT) levels in various plant and animal receptor species. The SRS provides an excellent setting for evaluating low-level tritium cycling dynamics in the environment, including the transformation of tritiated hydrogen gas (HT) and tritiated water (HTO) to OBT.

Summary of Research Activities

Continued analysis of biological samples from the SRS, including samples from the SRS Mixed Waste Management Facility (MWMF) Tritium Irrigation Site (i.e., irrigation source pond, soil profile, pine needles, limbs, leaf litter, etc.) and various control sites. In FY19, research focused on comparing OBT recovery using the muffle furnace method compared to the bomb calorimeter. Although the results from both methods were well correlated, discrepancies between further confirmed the need for certified reference materials (CRMs) that can verify the quality of the data. Aspects of the current research will continue through a collaboration with Drs. Lee (Albany State University) and Brigmon (SRNL) outlined in *Microbiome-Based Reporting of Tritiated Water Application at a Mixed Waste Disposal Site* will be funded by the DOE Minority Serving Institutions Partnership Program (MSIPP). The soil microbiome will be monitored in coordination with required biannual sampling at the site to assess remediation efficacy. SREL will then analyze free tritiated water and organically bound tritium (OBT), the tritium fraction with the highest biological residence time, in the same soil microbiome samples. In addition, SREL is under consideration to host the International OBT Workshop for 2021.

Conclusions

- 1) HTO levels in soils and plants tissues were lower than tritium source (dilution from precipitation)
- 2) $OBT \neq HTO$ (OBT reflects long-term exposure)
- 3) OBT levels in samples analyzed by the muffle furnace method compared to the bomb calorimeter were generally well correlated.

Major Impact(s) of Research

- 1) Provide data for estimating tritium dose calculations in risk assessment scenarios, particularly under specific management strategies (i.e., prescribed burns) where OBT may be released to the environment in a more available form (e.g., water vapor).
- 2) Enable site management to estimate potential human and ecological tritium exposure levels associated with new SRS mission activities, ongoing site cleanup efforts, accidental release, and routine site custodial activities.

Other Project Personnel

Matt Baker, Research Professional – SREL

Christina Logan, Research Professional – SREL

Morgan Shapiro, Lab Technician – SREL

Jeffery Lott, Lab Technician – SREL

Liyun Zhang, Graduate Assistant - SREL

External Collaborators

Dr. Robin Brigmon (SRNL)

Dr. Yon Jin Lee (Albany State University)

Products

Baker, M.R, and J.C. Seaman. 2019. Estimating Evapo-Transpiration Losses for Tritium at the MWMF: 2018 End of Year Summary Report. Submitted to SRNS-ACP March 27, 2019.

- Seaman, J.C., F.M. Coutelot, and M.R. Baker. 2019. Tritium Distribution and Cycling on the Savannah River Site: Evaluating Organically Bound Tritium. Update Submitted to NNSA.
- Lee, Y.J. (Albany State University), R. Brigmon, and J.C. Seaman. 2019. Microbiome-Based Reporting of Tritiated Water Application at a Mixed Waste Disposal Site. Submitted to DOE Office of Environmental Management's (DOE-EM) Minority Serving Institutions Partnership Program (MSIPP), 5-31-19.

TASK 8. <u>SREL will continue to serve as a regional resource for scientific expertise and environmental</u> research. <u>SREL staff scientists will continue to provide special technical assistance to other</u> <u>site contractors, area stakeholders, other researchers, and the public.</u> <u>SREL will also continue</u> <u>to collaborate with scientist from other institutions</u>

In FY19, SREL faculty, staff, and students conducted a diversity of environmental research projects both on and off of the SRS in support of their mission to pursue collaborations and funding to serve as a regional source of scientific expertise and to provide technical assistance to other site contractors, stakeholders, other researchers, and the public. Due to both the specific technical expertise represented by research faculty and staff at SREL and the unique opportunities for scientific research represented on the Savannah River Site, SREL scientists are often sought out as potential collaborators by researchers across the globe. SREL staff served as collaborators on both funded and non-funded research involving environmental remediation, ecotoxicology and environmental stewardship and, as Principal Investigators or co-Investigators on funded research all over the United States and internationally. In addition, SREL faculty, staff and students served as hosts for over 198 researchers from other universities, federal and state agencies, and non-governmental organizations to discuss and conduct collaborative research and funding. Research, external funding, and requests for technical assistance conducted in support of this task are outlined below:

Collaborations and Externally Funded Research on the SRS

Costs of Incubation: Linking Incubation-Induced Alterations in Phenotype to Changes in Fitness

<u>Funding Entity</u> National Science Foundation <u>Start Date and Funding Amount</u> October 2018; NFP <u>SREL Collaborator</u> Robert Kennamer

Objectives

Our overall goals have been to examine the importance of incubation temperature during early development, and to provide a better understanding of how reproductive tradeoffs made by females influence their fitness. In one recent part of the project, we collected eggs from SRS wood duck nests and artificially incubated the eggs at different temperatures known to influence phenotype. Following hatching, the ducklings were subjected to a battery of challenges that are typical for wood duck ducklings, including for example, the ability to successfully exit a nest cavity and make vocalizations to attract the attentions of an attending adult female.

Summary of Research Activities

In FY19, results were presented at multiple meetings and manuscripts were published, submitted, and in preparation.

Conclusions

- 1) Our studies have determined how the early thermal and social environments interact to influence offspring behavior and suggested that these factors may play an important role in shaping offspring behaviors and performance metrics that are critical for survival.
- 2) Our studies have documented that significant variability in egg temperatures exists within incubated wood duck clutches, enough so that duckling phenotypes, including important survival-related traits, can be impacted.

Major Impacts of Research

1) Illustrates the potential importance of incubation as related to offspring quality/performance.

Other Project Personnel

John Hallagan, Research Technician - Virginia Tech

Sydney Hope, PhD student - Virginia Tech **External Collaborators**

Dr. Bill Hopkins - Virginia Tech University Dr. Sarah DuRant – University of Arkansas

- Hepp, G.R. and R.A. Kennamer. Managing nest boxes for wood ducks: what have we learned? The 72nd Annual Conference of the Southeastern Association of Fish and Wildlife Agencies, Mobile, AL. October 22, 2018 (Oral presentation).
- Hepp, G.R. and R.A. Kennamer. Using nest boxes to learn about and help manage cavity-nesting ducks. The 72nd Annual Conference of the Southeastern Association of Fish and Wildlife Agencies, Mobile, AL. October 22, 2018 (Oral presentation).
- Hope, S.F., S.E. DuRant, F. Angelier, J.J. Hallagan, I.T. Moore, R.A. Kennamer, and W.A. Hopkins. Incubation behavior is related to prolactin and egg temperature in a wild bird. The 2019 Society for Integrative and Comparative Biology Annual Meetings, Tampa, FL. January 6, 2019 (Oral presentation).
- Hepp, G.R., R.A. Gitzen, and R.A. Kennamer. Relative importance of vital rates to populations dynamics of wood ducks. The 8th North American Duck Symposium, Winnipeg, MB. August 26-30, 2019 (Oral presentation).
- Hope, S.F., S.E. DuRant, J.J. Hallagan, M.L. Beck, R.A. Kennamer, and W.A. Hopkins. Within-clutch variation in egg incubation temperature: A constraint on the evolution of clutch size. Conference on Advances in Avian Reproduction, University of Lincoln, UK. September 3-6, 2019 (Oral presentation).
- Hope, S.F., S.E. DuRant, J.J. Hallagan, M.L. Beck, R.A. Kennamer, and W.A. Hopkins. *In prep.* Incubation temperature as a constraint on clutch size and a cost of brood parasitism. Journal: *American Naturalist.*
- Grimaudo, A., S.F. Hope, S.E. DuRant, R.A. Kennamer, J.J. Halagan, and W.A. Hopkins. *In review*. Environmental temperature and body condition influence inter-individual variation in night incubation behavior in wood ducks (*Aix sponsa*). Submitted to: *Journal of Avian Biology*.
- Hope, S.F., R.A. Kennamer, A. Grimaudo, J.J. Halagan, and W.A. Hopkins. *In review*. Incubation temperature affects duckling body size and food acquisition despite no effect on behavior. Submitted to: *Integrative Organismal Biology*.
- Hope, S.F., S.E. DuRant, F. Angelier, J.J. Halagan, I.T. Moore, C. Parenteau, R.A. Kennamer, and W.A. Hopkins. *In review*. Prolactin is related to incubation constancy and egg temperature following a disturbance in a precocial bird. Submitted to: *Hormones and Behavior*.
- Hepp, G.R., R.A. Gitzen, and R.A. Kennamer. *In press*. Relative importance of vital rates to population dynamics of Wood Ducks. *Journal of Wildlife Management*.
- Hope, S.F., C. Beunaventura, Z. Husain, S.E. DuRant, R.A. Kennamer, W.A. Hopkins, and C.K. Thompson. 2019. Limited support for thyroid hormone or corticosterone related gene expression as a proximate mechanism of incubation temperature-dependent phenotypes in birds. *Frontiers in Physiology* 10:1-9. doi: 10.3389/fphys.2019.00857.
- Hope, S.F., R.A. Kennamer, S.G. van Montfrans, and W.A. Hopkins. 2019. Incubation temperature and social context affect the exodus of precocial ducklings. *Behavioral Ecology* 30:518-527. doi: 10.1093/beheco/ary192.

<u>Total water use and source partitioning in woody bioenergy crops determined by coupled mass flux and</u> <u>stable isotope signatures</u>

Funding Entity

USDA National Institute of Food and Agriculture <u>Start Date and Funding Amount</u> September 2015; \$915,593 <u>SREL Collaborators</u> D. Aubrey <u>Objectives</u> The overall goal of this proposed research is to cor

The overall goal of this proposed research is to compare the water relations and the impact on local hydrology of intensively managed pine and eucalypt woody bioenergy crops over stand development. Specifically, we will quantify total water use and determine water sources for two preeminent woody bioenergy crops. Loblolly pine is the cornerstone of southern US forest production, whereas eucalypts represent even greater productivity potential. Our results will be used to parameterize a process based model to predict watershed-scale changes that might occur to the hydrologic cycle based on increasing the intensity of management and planting a woody crop species with even higher production potential than loblolly pine.

Summary of Research Activities

Experimental plots were prepped and planted in year 1. Silvicultural treatments were applied in years 1 through 5. Experimental instrumentation to measure hydrologic inputs and outputs was deployed at the beginning of the third growing season in year 3 of the project. During the fourth year of our project, we continued water balance measurements that are central to this project. This includes the continued monitoring of precipitation, throughfall, and soil moisture, as well as continued monitoring of six trees of each species (loblolly pine and *Eucalyptus benthamii*) per plot (6 plots per species) to measure whole tree water use. It also includes the routine collection of water from plant tissues, soils, and groundwater for isotope analysis. We have been measuring leaf area index of the experimental plots monthly since May 2016. A new cryogenic vacuum extraction method has been implemented for the purposes of extracting water from plants and soils for the comparison of stable isotopes of ²H and ¹⁸O. These stable isotopes are conservative tracers that can be used to tease out where trees are sourcing water from within the soil column. This method can then be used to model tree water usage and inform sustainable land management practices with respect to water resources and woody crops. We are continuing to work on evaluating issues of bias in the sampling and extraction of xylem water. We have completed our measurements on experimental plots.

Conclusions

In addition to the work related to water budgets, we continue to learn a great deal about *Eucalyptus* and its potential as a production plantation species in the southeast US. We have found (and reported through a journal article) that *Eucalyptus* is susceptible to a fungus that causes a leaf disease that severely impacts growth. We have also found that non-native *Eucalyptus* leaf beetle populations find and exploit *Eucalyptus* stands in the southeast. These issues, along with the issues of frost tolerance, make *Eucalyptus* a rather risky choice for landowners and, despite the larger impact on the hydrologic budget compared to a native species like loblolly pine, we do not expect this species to become a large part of the forest production in the southeast until these issues are overcome. Finally, we have shown that it is not just the non-native shown that native species, like sweetgum (*Liquidambar styraciflua* L.) may exhibit much lower water use efficiencies and use a much larger amount of soil water to produce similar amounts of biomass as loblolly pine.

Major Impact(s) of Research

1) Provides quantitative information on how the hydrologic cycle would be influenced by land-use shifts away from standard, low-input, forestry management for pulpwood and timber toward intensively managed forest stands for bioenergy production

2) Provides general information about production potential of Eucalyptus in southeast US.

Other Project Personnel

Mackenzie Dix, Research Assistant - SREL

Scott Oswald, PhD Student - UGA

Seth Younger, PhD Student - UGA

External Collaborators

P. Caldwell (USDA Forest Service)

R. Jackson (UGA)

J. McDonnell (University of Saskatchewan)

C. Miniat (USDA Forest Service)

Products

- Dix, M.J. and D.P. Aubrey. 2019. Calibration approach influences sapflow estimates. Ecological Society of America. Louisville, KY. (Oral presentation)
- Dix, M.J. and D.P. Aubrey. 2019. Calibration approach influences sapflow estimates. Ecological Society of America. Louisville, KY. (Oral presentation)
- Griffiths, N.A., B.M. Rau, K.B. Vaché, G. Starr, M.M. Bitew, D.P. Aubrey, J.A. Martin, E. Benton, and C.R. Jackson. 2018. Environmental effects of short-rotation woody crops for bioenergy: What is and isn't known. Global Change Biology Bioenergy, 11:554-572.
- Caldwell, P.V., C.R. Jackson, C.F. Miniat, S.E. Younger, J.A. Vining, J.J. McDonnell, and D.P. Aubrey. 2018. Woody bioenergy crop selection can have large effects on water yield: A southeastern United States case study. Biomass and Bioenergy, 117: 180-189.

Oswald, S.W. and D.P. Aubrey. 2018. Increasing biomass production on limited land area through an optimal planting arrangement. BioEnergy Research, 11(1): 13-21.

- Younger, S., D.P. Aubrey, and C.R. Jackson. 2018. Water budget comparison of early rotation Loblolly Pine and Eucalyptus benthamii in the Upper Coastal Plain of South Carolina. Warnell Graduate Student Symposium. Athens, GA. (Poster presentation)
- Brockman, L.E., S.E. Younger, C.R. Jackson, D.P. Aubrey, K.F. Janzen, and J.J. McDonnell. 2018. Stable isotope and mass balance applications in describing soil water contributions to loblolly pine and sweetgum xylem water. Warnell Graduate Student Symposium. Athens, GA. (Poster presentation)
- Brockman, L., S. Younger, C.R. Jackson, K. Janzen, D.P. Aubrey, and J. McDonnell. 2017. Systematic errors associated with scaling soil CO2 efflux in forest ecosystems and their impact on the global carbon cycle. American Geophysical Union, New Orleans, LA. (Poster presentation)
- Aubrey, D.P. 2017. Forest Ecophysiology at the Savannah River Ecology Laboratory. Joseph W. Jones Ecological Research Center. (Invited seminar)
- Aubrey, D. P., C.R. Jackson, C. R. Miniat, J.J. McDonnell, and P.V. Caldwell. 2016. Hydrologic Budgets for Short Rotation Loblolly Pine and Eucalyptus. USDA-NIFA AFRI Sustainable Bioenergy Annual Project Director Meeting. Tampa, FL. (Invited oral presentation)

Differences in the physiological response of Liquidambar styraciflua caused by season of burn.

Funding Entity

USDA Forest Service-Southern Research Station

Start Date and Funding Amount

August 2015; \$60,000

SREL Collaborators

D. Aubrey

Objectives

The goal of this project was to determine how the season of burning influenced mortality, resprouting, and subsequent growth of American sweetgum.

Summary of Research Activities

Planted seedlings in 20 L pots in spring 2016 and maintained them for an entire growing season. Subjected a subset of seedlings to experimental burns prior to leaf-out in early March 2017. Subjected another subset of seedlings to experimental burns after leaf-out and leaf expansion in June 2017. Whole plant carbon budgets were collected using gas exchange. Whole-plant harvests were conducted routinely throughout the growing season following experimental burns.

Conclusions

- 1) Contrary to the hypothesis—and previous reports—the dormant season burn resulted in higher mortality than the growing season burn and fire intensity did not explain differences in mortality.
- 2) Root mass of burned trees increased after burning, but was lower than controls at the end of the year and season of burn did not impact root mass at the end of the season.
- 3) Root carbohydrate reserves decreased during leaf-out, but quickly returned to pre-burn levels and, regardless of season, root carbohydrate reserves were similar to controls at end of season.

Major Impact(s) of Research

1) Results from this study will provide scientists and land managers empirical data to support or reject current thoughts on timing of prescribed fire and its efficacy in controlling sweetgum in southeastern forests.

Other Project Personnel

S. Ruswick, MS Student - UGA

External Collaborators

R.O. Teskey (UGA)

J.J. O'Brien (USDA Forest Service)

- Aubrey, D.P., S. Ruswick, and J. O'Brien. 2018. Does season of burn influence the efficacy of prescribed fire in controlling sweetgum? Ecological Society of America, New Orleans, LA. (Poster presentation)
- Ruswick, S., J. O'Brien, D.P. Aubrey. 2018. Sweetgum saplings burned in the growing season have lower mortality and similar biomass to those burned in dormant season. Warnell Graduate Student Symposium. Athens, GA. (Oral presentation)
- Aubrey, D.P, D.R. Coyle, J.J. O'Brien, B.S. Hornsby, D.J. Moorhead, and J.T. Nowak. Fire and the ecophysiology of southern pines. 2019. Southern Forest Insect Work Conference. Savannah, GA (Invited oral presentation)
- Aubrey, D.P. 2019. Understanding Ecosystem Process & Resilience through the lens of Longleaf Pine Physiology. Jones Center. (Invited seminar)

Canopy complexity, physiological function, and ecosystem resilience of longleaf pine.

Funding Entity

Tall Timbers Research Inc. <u>Start Date and Funding Amount</u> August 2018; \$23,188 <u>SREL Collaborators</u>

D. Aubrey

Objectives

The goal of this project is to better understand how canopy architectural complexity of old-growth longleaf pine trees may influence physiological function and, ultimately, ecosystem resilience. Briefly, we hypothesize that individual with complex canopies may maintain positive water relations during periods of moderate drought because of lower branches with foliage that maintain higher water potentials.

Summary of Research Activities

We instrumented a series of old-growth longleaf pine trees representing complex and simple canopy architecture in spring 2018 and measured physiological processes throughout the 2018 growing season. Measurements are ongoing.

Conclusions

- 1) Lower branches exhibited higher water potential than upper branches within the same continuous canopies; however, water potential was higher in discrete canopies compared to continuous canopies measured at the same height.
- 2) Photosynthetic assimilation in discrete canopies was higher than continuous canopies at the same height, and there were no differences between upper and lower continuous canopy positions.
- 3) Whole-tree water use was higher in continuous than discrete canopies, with the majority of water use occurring in upper portions of continuous canopies.

Major Impact(s) of Research

1) Our data suggest lower strata of complex canopies are less water stressed than upper portions of continuous and discrete canopies; however, upper portions of both canopy types maintain similar or higher photosynthetic rates than lower portions of continuous canopies.

Other Project Personnel

Laura Fowler, MS Student - UGA

External Collaborators

NA

- Fowler, E., K. Hiers, J. O'Brien, D. Johnson, and D.P. Aubrey. 2019. Does canopy complexity influence physiological function and contribute to ecosystem resilience of old-growth longleaf pine? Warnell Graduate Student Symposium. Athens, GA.
- Fowler, E., J.K. Hiers, J.J. O'Brien, E.L. Loudermilk, S. Pokswinski, E. Rowell, and D.P. Aubrey. 2019. Does canopy complexity promote resilience in old-growth longleaf pine forests? Ecological Society of America. Louisville, KY.

Comparative hydrologic budgets and water use efficiencies of developing bioenergy plantations.

Funding Entity

USDA National Institute of Food and Agriculture Start Date and Funding Amount September 2019; \$498,663 **SREL Collaborators** D. Aubrey **Objectives**

Our overarching goal is to improve our understanding of how the major outputs of the forest hydrologic cycle change through stand development and manipulate those dynamics through silvicultural approaches to increase water use efficiency (WUE).

Summary of Research Activities

We will construct hydrologic budgets by quantifying the hydrologic inputs (precipitation and interception) and outputs (E_t and E_s) in experimental plots of the most promising alternative woody bioenergy crop species for production in the southeastern US (i.e., coppiced sweetgum and *Eucalyptus*) and compare them with the current management system for this region (i.e., loblolly pine). In doing so, we will gain the most comprehensive species comparisons of water use and WUE to date that consider dynamic changes in E_s and E_t that occur as forest stands transition through stand development from the point of establishment when leaf area is low and the soil surface is bare to the point of canopy closure when leaf area has maximized and soil surfaces contain years of litter inputs that impede E_s . In addition to comparing species, we will compare silvicultural approaches that impose drastically different densities (i.e., coppice vs. standard row plantings of sweetgum; 6,364 seedlings ha⁻¹ and 1,336 seedlings ha⁻¹, respectively). Different densities will result in different rates of canopy leaf area development and, therefore, different dynamics in hydrologic budgets. We will also perform a series of manipulative experiments to gain a mechanistic understanding of how litter type, litter depth, shading, and boundary layer mixing influence E_s . We will also measure fine- and coarse-root dynamics among species and silvicultural approaches to calculate WUE, not only with respect to aboveground production and bioenergy production, but also with respect to belowground carbon sequestration. Our proposed research will provide rigorous data accounting for dynamic shifts in key hydrologic processes that occur through stand development and will demonstrate silvicultural approaches to increase WUE, thereby informing decisions on how to manage bioenergy production systems.

Conclusions

This research has just begun, thus there are no conclusions at this time.

Major Impact(s) of Research

1) By taking a systems approach to quantifying evapotranspiration and its components (soil evaporation and transpiration) in woody feedstock (loblolly pine, American sweetgum, and Eucalyptus) across entire harvest rotations, we will capture dynamic shifts occurring through stand development and provide the most comprehensive assessment of WUE to date. We hypothesize that inherent differences in leaf habit and area among species, coupled with silvicultural options related to density and resource availability, provide opportunity to reduce soil evaporation during early stand development, thereby increasing rotation-length WUE and reducing the overall ecological footprint of woody bioenergy production systems. Our empirical approach, assessing rotation-length hydrologic budgets and generating integrated measures of WUE using a systems framework, will provide data that can be used directly by landowners in the Southeast US as a decision support tool for selecting woody bioenergy crop species and silvicultural approaches. Our conceptual framework, which relies broadly on functional traits of potential woody bioenergy crop species, formulates a hypothesis that will be tested with our empirical data and thus, generalized patterns, processes, and controls can be transferrable to other forestry and agriculture production systems. This project addresses the urgent need for quantitative information regarding trade-offs between maximizing forest production and maintaining sustainable water yields.

Other Project Personnel C.R. Jackson, Faculty - UGA External Collaborators NA

Products

No publications, presentations, or reports have been prepared yet.

Short rotation woody crop biomass and nitrogen.

Funding Entity

USDA Forest Service, Southern Research Station

Start Date and Funding Amount

July 2018; \$100,000

SREL Collaborators

D. Aubrey

<u>Objectives</u>

The goal of this project is to: (1) quantify key components for soil-vegetation N budget in developing plantation systems; and (2) to determine productivity, biomass accumulations rates, and plant nutrient uptake requirements for loblolly pine plantations as a function of changing resource availability and changing crop tree sink strengths through alterations in plantation densities.

Summary of Research Activities

Over the past six years, data has been collected to: (a) quantify nitrogen mineralization, (b) measure nitrogen leachate, (c) quantify forest floor development, (d) assess the soil nutrient supply relative to demand by the trees, (d) quantify tree productivity, and nutrient and biomass accumulation, (e) measure above- and below-ground biomass accumulation.

Conclusions

- 1) Herbicide application during the first year of forest stand development was the single most important silvicultural treatment to facilitate stand productivity.
- 2) There was no difference in productivity between operational and one-half operational levels of fertilization.
- 3) Higher planting densities did not impact individual tree growth or form during early stand development.

Major Impact(s) of Research

1) Our results suggest that herbicide application may be the most important silvicultural treatment to promote growth during the first year following planting and that current operational fertilization levels far exceed the uptake capacity of developing forest plantations.

Other Project Personnel

Gabriel Ferreira, Postdoc - SREL

External Collaborators

NA

Products

Ferreira, G.W.D., B. Rau, and D.P. Aubrey. 2019. Growth and biomass allocation during early loblolly pine development as influenced by fertilization and herbicide. Ecological Society of America. Louisville, KY.

Managing forests for ecological services and environmental sustainability.

<u>Funding Entity</u> USDA Forest Service. <u>Start Date and Funding Amount</u> May 2019; \$479,480 <u>SREL Collaborators</u> D. Aubrey

Objectives

Our overarching goal is to improve our understanding of how carbon inputs to soil influence soil organic carbon (SOC) accrual and storage dynamics through stand development and across multiple harvest rotations, thereby demonstrating how manipulating inputs through silvicultural approaches can influence the magnitude and timing of SOC equilibrium.

Summary of Research Activities

We developed a conceptual model, based on carbon inputs to soil and their decomposition rates, and parameterized it with empirical data to test the hypothesis that changing the silvicultural approach to increase carbon inputs to soil can result in increased equilibria of SOC inputs, and ultimately, increased SOC accrual and storage. To challenge/validate our model, we will establish plots of intensively managed loblolly pine and sweetgum and construct belowground carbon budgets that account for all biomass inputs to soil, decay rates of those inputs, soil CO₂ efflux, and subsequent changes in SOC across multiple rotations.

Conclusions

This research has just begun, thus there are no conclusions at this time.

Major Impact(s) of Research

1) Our empirical approach, assessing biomass production and carbon inputs to soil through early stand development and across multiple harvest rotations, will provide data that can be used directly by landowners in the Southeast US as a decision support tool for selecting production forest species and silvicultural approaches that simultaneously maximize productivity, increase atmospheric CO₂ sequestration, improve soil health, and enhance ecosystem resilience. Our conceptual framework, which relies broadly on silvicultural decisions and the functional trait of resprouting, formulates a hypothesis that will be tested with our empirical data and thus, generalized patterns, processes, and controls can be broadly transferrable to other production forestry systems nationally and globally.

Other Project Personnel

Laura Fowler, MS Student - UGA

External Collaborators

NA

Products

No publications, presentations, or reports have been prepared yet.

Effects of methylmercury on mosquito development and oviposition behavior

Funding Entity

SREL and NSF-REU

Start Date and Funding Amount

June 2017; NFP

SREL Collaborators

G. Dharmarajan and X. Xu

Objectives

To quantify the effects of methylmercury on early-life mortality and oviposition behavior of yellow fever mosquitoes (*Aedes aegypti*)

Summary of Research Activities

In this study we use the yellow fever mosquito, *Aedes aegypti*, to test how methylmercury (MeHg) affects oviposition site selection. We found that mosquito larval development rate and survival were negatively affected at MeHg concentrations ≥ 100 ppb. Adult females not exposed to MeHg as larvae avoided oviposition sites with high MeHg concentrations (> 50 ppb), but MeHg exposure at the larval stage significantly affected this oviposition site selection. Specifically, females raised from larvae exposed to non-toxic MeHg levels (i.e., 5-50 ppb) showed a significant increase in preference for oviposition sites contaminated with toxic MeHg concentrations (≥ 500 ppb), compared to unexposed controls. Importantly, however, this maladaptive behavioral response is abolished in female mosquitoes raised from larvae exposed to toxic MeHg concentrations (i.e. 100 ppb), and these mosquitoes showed a significant increase in preference for MeHg uncontaminated oviposition sites, compared to unexposed controls. Thus, in mosquitoes,

Conclusions

- 1) Methylmercury (MeHg) negatively impacts development and survival of mosquitoes
- 2) Adult female mosquitoes unexposed to MeHg avoid oviposition sites with high MeHg
- 3) Females exposed to non-toxic MeHg levels prefer oviposition sites with high MeHg, but females exposed to toxic MeHg levels prefer MeHg uncontaminated oviposition sites

Major Impact(s) of Research

- 1) The magnitude of MeHg exposure in one generation can impact MeHg exposure in subsequent generations by altering oviposition site selection behavior.
- 2) To our knowledge, this is the first study to demonstrate transgenerational effects of behavior on contaminant exposure
- 3) Our results have broad implications for our understanding of how contaminant-mediated behavioral modifications can feedback on contaminant exposure risk across multiple generations, and how behavior can affect the evolution of organisms inhabiting a heterogeneous environments.

Other Project Personnel

E. Neff, MS student - SREL

External Collaborators

M. Tanelus (University of South Carolina Upstate)

R. Maness (Presbyterian College)

- Neff E., A. Coleman, R. Maness, M. Tanelus, X. Xu and G. Dharmarajan (2019) Effects of methylmercury on mosquito oviposition behavior: Maladaptive response to non-toxic exposure. Science of The Total Environment 667: 248-254
- Neff, E., A. L. Coleman, R. W. Maness, M. Tanelus, X. Xu and G. Dharmarajan (2019) Mother does not always know best: Effects of methylmercury on mosquito oviposition behavior. 29th Annual Molecular Parasitology & Vector Biology Symposium. May 1, 2019. Athens, GA

<u>Restoring Headwater Streams and Riparian Corridors at the Savannah River Site, SC: Part A-</u> <u>Mitigation Plan Proposal Supporting Documentation</u>

Funding Entity

USDA Forest Service-Savannah River <u>Start Date and Funding Amount</u> June 2017; \$27,042 <u>PI and co-PI's</u> Dean E. Fletcher

Objectives

Our overall goal has been to provide assessments of legacy and current stream disturbances to enable Savannah River Site management organizations and regulatory oversight agencies to move forward with specific restoration/enhancement treatments and a monitoring plan for a stream restoration-mitigation project.

Summary of Research Activities

Stream restoration and enhancement provides opportunity to correct or improve previous alterations that have destroyed, diminished, or impaired the character and function of stream systems. The Savannah River Site (SRS) provides an ideal research opportunity for restoration of coastal plain streams. SRS stream disturbances span a temporal range from pre-SRS legacy impacts, through the early infrastructure development in the early 1950s, to more recent and current industrial activities. In a collaborative effort, a multiphase program was conducted to characterize SRS streams, identify risks of legacy and recent disturbances, and identify disturbed stream reaches with potential for restoration. Three levels of assessments were completed with each level providing an increased level of detail and scientific rigor. A Level I assessment involved a broad scale survey of potential stream disturbances and stream basin characterization. Level II assessments assessed the effects of stream alterations on physical stream condition in a subset of Phase I identified streams. Level III assessments further evaluated a selected subset of stream reaches by measuring additional hydrology, physicochemistry, biology, and geomorphology features. This comprehensive stream evaluation identified management options and is guiding prescriptions for potential restorative actions.

Our work has identified enhancement/restoration opportunities that span a broad range of complexity, cost, and risk. Current plans are being developed for some of the more cost effective opportunities with lower levels of risk of environmental disturbance by the management activities. Experience gained by these restoration and enhancement efforts in less complex situations, may be applied to more costly systems in the future. The headwaters of Tinker Creek offer several suitable areas/reaches for restoration and enhancement that could be performed cost effectively, in a timely fashion and with overarching goals that could improve stream and riparian condition. Restoration opportunities in the Tinker Creek watersheds include ditch filling, in-stream habitat enhancement, and dam removal. Riparian enhancement would involve opening of the closed-canopy forest to release native cane in the understory. Use of fire to manage forest understory including cane stands will be evaluated. Prescriptions for these specific stream enhancements as well as required permit materials are being assembled.

Conclusions

- 1) Amount of industrial area in a drainage has a strong influence on instream geomorphology, chemistry, and macroinvertebrate communities; streams receiving excessive stormwater runoff are generally the most disturbed streams within our study systems.
- 2) Basin and valley characteristics interact with landscape disturbance to influence level of stream disturbance.
- 3) Enhancement of tributaries of Tinker Creek represent cost-effective options for establishment of a mitigation bank.

Major Impact(s) of Research

- 1) We have identified effects of legacy and current disturbances on stream chemistry, hydrology, geomorphology, and biology on select SRS streams.
- 2) Through an effort with onsite and offsite collaborators, we are proposing restoration plans including post-treatment monitoring for 4 headwater streams while using a fifth as a reference system. These efforts may establish better management strategies for the riparian zones of sandhills streams.
- 3) A framework upon which a headwater stream mitigation bank can be built is being developed and restoration Prospectus written.

Other Project Personnel

Paul Stankus, Research Professional – SREL

Christina Fulghum, Research Technician – SREL

J Vaun McArthur, Senior Research Scientist – SREL

External Collaborators

Christopher Barton (University of Kentucky)

Richard Biemiller (University of Kentucky)

Andy Horcher (USDA Forest Service)

John Blake (USDA Forest Service)

- Fletcher, D. E., A. H. Lindell, J. C. Seaman, P. T. Stankus, N. D. Fletcher, C. D. Barton, R. A. Biemiller, and J V. McArthur. 2019. Sediment and Biota Trace Element Distribution in Streams Disturbed by Upland Industrial Activity. Environmental Toxicology and Chemistry 38:115-131.
- Fletcher, D. E., B. E. Lindell, A. H. Lindell, P. T. Stankus, N. D. Fletcher, J V. McArthur, and J. C. Seaman. 2019. Basins, beaver ponds, and the storage and redistribution of trace elements in an industrially impacted coastal plain stream on the Savannah River Site, SC. Environment International 133: 105174. 11 pp.

<u>Development of Innovative Biostimulation and Bioaugmentation Strategies for In-Situ Uranium</u> <u>Bioremediation of DOE Contaminated Ecosystems</u>

Funding Entity

DOE - Minority Serving Institutions Partnership Program (MSIPP) <u>Start Date and Funding Amount</u> October 2019; \$25,000 <u>SREL Collaborators</u> Dr. John C. Seaman <u>Objectives</u>

This project focused on the SRS Steeds Pond/Tims Branch ecosystem in an effort to reduce solubility, bioavailability and toxicity of U and Ni co-contaminants. The current research addresses the following objectives: 1a) evaluate the efficacy of P sources (phytate, glycerol phosphate, phosphate rock) as biostimulants, in aerobic soil slurry microcosms; 1b) identify and isolate the microbial "first-responders" to U+phytate and Ni individually and in combination; 2) evaluate U/ Ni bioremediation via biostimulation, bioaugmentation, or a combination thereof using the "first-responder" bacterial and fungal isolates; 3) conduct genomics and proteomics studies on isolated strains; and 4) train a steady stream of under-represented students in the field of environmental biotechnology, genomics, proteomics, chemistry and environmental restoration.

Summary of Research Activities

This newly funded project is a continuation of ongoing collaboration between FAMU and SREL. Largescale legacy of environmental contamination with U resulted from years of nuclear materials production in the U.S. Towards this end, we continue to obtain innovative strategies for remediation and restoration of uraniferous soils and sediments of the Savannah River Site (SRS). Remediation of such environments using conventional excavation-disposal or pump-and-treat approaches has been deemed unfeasible. Less expensive in-situ remediation, by applying chemical and/or microbial additives, can alter contaminant speciation, thus enhancing solid-phase partitioning to reduce migration, bioavailability and associated toxicity. Towards this end, we have successfully demonstrated the effectiveness of PO₄ compounds, such as phytate, glycerol phosphate and phosphate minerals (e.g., apatite), as biostimulants to stabilize heavy metals and radionuclides through sorption and/or the formation of secondary phosphate precipitates. However, detrimental impacts of P amendments in soils has also been demonstrated, in that nickel (Ni) became more labile relative to U. Therefore, in sites where both U and Ni occur as mixed contaminants, such as the SRS Steed Pond soils, further research is required. A supplemental technique, called bioaugmentation, which entails isolation of native U-bioremediative microbiota and evaluation of their efficacy as bioremediative amendments with or without additional P biostimulants, is being developed. P

Conclusion

- 1) Demonstrated the effectiveness of PO₄ based amendments in immobilizing U in SRS soils.
- 2) Identified potential problems associated with the use of soluble PO₄ sources for U immobilization.
- 3) Identified additional SRS microbial isolates that are insensitive to U and Ni contamination, and developed a U biosensor.

Major Impact of Research

The current research has the potential to impact the long-term disposition of U contaminated areas within Steeds Pond/Tims Branch watershed on the SRS, and other U contaminated sites associated with DOE Facilities.

Other Project Personnel (technical staff, graduate students, undergraduates, postdocs, etc.)

M.B. Baker, Research Professional - SREL

C. Logan, Research Professional - SREL

External Collaborators (and Affiliations)

Dr. Victor Ibeanusi (Florida A&M University)

Dr. Ashvini Chauhan (Florida A&M University)

Dr. Charles Jagoe (Florida A&M University)

Dr. Dan Kaplan (SRNL)

- Baker, M.R., FM. Coutelot, and J.C. Seaman. 2019. Phosphate amendments for chemical immobilization of uranium in contaminated soil. Environment International. 129; 565-572. doi.org/10.1016/j.envint.2019.03.017.
- Jaswal, R. A. Pathak, B. Edwards, R. Lewis, J.C. Seaman, P. Stothard, K. Krivushin, J. Blom, O. Rupp, A. Chauhan. 2019. Metagenomics-guided Analysis, Isolation and Characterization of Uranium Resistant Microbiota Using a High-throughput Approach. Genes. 10:325.
- Li, R., V. Ibeanusi, J. Hoyle Gardner, C. Crandall, C. Jagoe, J.C. Seaman, A. Anandhi, G. Chen. 2019. Bacterial-facilitated uranium transport in the presence of phytate at Savannah River Site. Chemosphere 233, 351-357.
- Agarwal, M., R.S. Rathore, C.H. Jagoe, J.C. Seaman, X. Xu, A. Chauhan. 2019. Isolation and characterization of a mercury resistant strain, Stenotrophomonas sp. WM2019 Conference. March 3-7, 2019, Phoenix, AZ.
- Jaswal, R., A. Pathak, J. Hunt, J.C Seaman, A. Chauhan. 2019. Innovative Techniques to Isolate Uranium Resistant Soil-Borne Biodegradative Microorganisms. WM2019 Conference. March 3-7, 2019, Phoenix, AZ.

Capture-Mark-Recapture Studies of Large Mouth Bass on Par Pond Reservoir

Funding Entity

DOE-EM Support to SREL

Start Date and Funding Amount

October 2017; \$30,000

SREL Collaborators

Dr. Olin E. Rhodes, Jr. and Matt Hamilton

Objective(s)

To estimate the population size and characterize movement patterns of largemouth bass, Micropterus salmoides, in relation to the distribution of contaminants on Par Pond reservoir.

Summary of Research Activities

We have individually marked >1400 largemouth bass from Par Pond with a RFID (Radio-frequency identification) PIT (Passive Integrated Transponder) tag. With each capture, morphometric data (e.g., length, weight, sex), GPS coordinates, and physical condition (i.e., health status) are recorded before each individual is released at the point of capture. This initial capture effort is ongoing and will provide baseline data for characterizing movement patterns and estimating largemouth bass abundance on Par Pond.

Conclusions

1) This research is in the initial stages of data collection.

Major Impact(s) of Research

1) This research will provide insight into the seasonal distribution and the potential for contaminant exposure and transport of largemouth bass on a man-made reservoir.

Other Project Personnel

Chris McBride, Assistant Director – SREL Megan Winzeler, Project Coordinator II – SREL Brian Morton, Property Management Coordinator – SREL Austin Coleman, MS Student – UGA

External Collaborators

NA

Products

No publications, presentations, or reports have yet been prepared.

Impact of long-term chronic exposure to low dose ionizing radiation on organismal health

<u>Funding Entity</u> SREL <u>Start Date and Funding Amount</u> NFP

SREL Collaborators

Dr. Ben Parrott and Dr. Olin E. Rhodes

Objectives

The overall goal of this research is to assess the effects of chronic low dose ionizing radiation over long periods of time on and the associated epigenome.

Summary of Research Activities

Environmental stressors influence developmental and aging trajectories leading to either health or disease. Ionizing radiation (IR) represents perhaps the most universal ecological stressor. With background levels present since the dawn of evolution, highly conserved pathways act to repair DNA damage caused by UV and other sources of IR. This project aims to assess how long-term chronic exposures to low doses of IR impact organismal health through altering epigenetic and genomic processes in a model fish (*Oryzais latipes*).

Epigenetic mechanisms mediate genome-by-environment interactions, and due to recent advances in sequencing technologies, the molecular modifications underlying these interactions can be assessed at genomic scales. DNA methylation is the best characterized epigenetic modification with roles in regulating gene expression and promoting chromosomal stability, and alterations to DNA methylation patterning characterize specific malignancies and in some cases, predict cancer prognosis. We will resolve epigenetic responses and genome-wide changes to gene regulation across IR doses and across different dose lengths. Further, we will assess the influence of IR exposure on the transcriptome. In addition to identifying biomarkers of IR exposure, these findings are expected to provide fundamental insights into how IR affects genomic processes associated with disease in environmentally, occupationally, and ecologically relevant exposure contexts.

Conclusions

Fish exposed to IR in the LoDIF system (ranging from 3-6 months, and 3 months recovery) have been collected and necropsied. Effects of IR exposure are observed on reproductive function and overall body condition. Global shifts in DNA methylation in response to ionizing radiation are not observed. RNA-seq analysis on hepatic tissue reveals an influence of IR exposure on gene expression patterns. Ongoing analyses are focused on identifying molecular pathways and markers that underlie organismal responses to IR.

Major Impact(s) of Research

- 1. Establishing a IR model to assess impacts of chronic low-dose exposure. Once established, the model can be extended to examine impacts of combinatorial exposures.
- 2. The project will produce findings that are relevant to both wildlife as well as human health.

Other Project Personnel

Emily Bertucci, PhD Student - SREL

Marilyn Mason, Research Technician - SREL

External Collaborators

NA

Products

Bertucci, E.M., Mason, M.W., Camus, A.C., Rhodes, O.E., Parrott, B.B. Chronic low dose irradiation alters the hepatic transcriptome, but not the global DNA methylome in medaka (*Oryzias latipes*). *In Review*.

- Bertucci, E.M., Mason, M.W., Camus, A.C., Rhodes, O.E., Parrott, B.B. Effects of low dose irradiation on the global DNA methylome in medaka (*Oryzias latipes*). 2019. Annual meeting of the Society for Integrative and Comparative Biology, Tampa, FL
- Topolski, C.R., M.W. Mason, E.M. Bertucci, O.E. Rhodes, Jr., and B.B. Parrott. Impacts of chronic exposure to gamma radiation on the DNA methylome using the medaka fish model. July 25, 2017. Savannah River Ecology Laboratory NSF REU symposium. Savannah River Ecology Laboratory, Aiken, SC, USA
- Topolski, C.R., M.W. Mason, E.M. Bertucci, O.E. Rhodes, Jr., and B.B. Parrott. Impacts of chronic exposure to gamma radiation on the DNA methylome using the medaka fish model. July 27, 2017. University of South Carolina Summer Research Symposium; University of South Carolina, Columbia, SC, USA
- Bertucci, E.M. and B.B. Parrott. Using small fish to understand environmental impacts of ionizing radiation. Poster presented at SREL's Touch An Animal Day. 2017.

Adaptive and disruptive epigenome-by-environment dynamics: molecular mechanisms to ecological <u>impacts</u>

 Funding Entity

 National Science Foundation

 Start Date and Funding Amount

 August 2018; \$571,839

 SREL Collaborators

 Dr. Ben Parrott

 Objectives

 The overall goal of this research is to determine the origins of natural variation in the epigenome.

Summary of Research Activities

The developmental environment can have profound impacts on an individual. For example, in many nonmammalian vertebrates including some fish, most turtles, and all crocodilians, environmental temperatures occurring during a specific period of incubation will determine if individuals develop as either a male or female. Previous findings have revealed a critical role for the endocrine system and epigenetics, heritable changes in gene function without changes in DNA sequence, in determining how the environment impacts traits. This project will determine the mechanisms by which environments and hormone signaling act on epigenetic processes to shape developmental trajectories and produce phenotypic diversity. The project focuses on species displaying temperature-dependent sex determination, to address how temperature induces and coordinates radically different phenotypic trajectories. The study will support the training and mentoring of two graduate students and two undergraduate researchers, with recruiting efforts aimed at under-represented groups in the sciences. The project will develop a primer focused on communicating common misperceptions about epigenetics. The findings will be presented to students at area schools as part of an established outreach program. Together, this work will advance the understanding of the mechanisms that determine how organisms interact with the environments and will provide training opportunities for the next generation of scientists.

The work will employ state of the art sequencing approaches to advance a developmental- and endocrinebased understanding of how adaptive epigenetic responses occur in nature, and how epigenetic responses are disrupted by environmental stressors for which a shared evolutionary history is absent. The Parrott Lab has previously observed widespread sexually dimorphic DNA methylation patterning across the gonadal genome. However, the developmental and molecular processes by which a bipotential genome acquires a sexually dimorphic epigenome is unclear. Developmental windows of environmental sensitivity will first be identified by resolving the temporal dynamics of the DNA methylome during temperature-dependent sex determination and reproductive development. Combinatorial treatments of temperature and hormones will then reveal the degree to which temperature and endocrine signals act on overlapping or distinct regions of the epigenome. Finally, the project will investigate the influence of environmental quality on the sexually dimorphic methylome. Given the inherent environmental sensitivity of temperature-dependent sex determination, it is hypothesized that environmental variables (other than temperature) also influence this process to affect the development and subsequent function of the reproductive system. Together, experiments will reveal how environmental and biological signals are integrated into developmental processes that result in phenotypic diversity. Findings from this work will be disseminated in peer-reviewed publications, presentations at national and international scientific, and more broadly via outreach activities in partnership with area schools.

Conclusions

Work so far has modeled the influence of ecological and climatic factors on the thermal environment experienced by developing alligator embryos. When future climatic predications are integrated into the constructed model, sex ratios of alligator populations are predicted to be dramatically affected in the next 20 years. In the absence of significant compensatory behaviors in maternal nest site choice, skews in

population sex ratios will likely negatively influence the ability of these populations to persist in the long-term.

Major Impact(s) of Research

- 1) Establish fundamental insights into how environmental cues are sensed and integrated into developmental trajectories
- 2) Provide a basic understanding of how natural environmental conditions interact with contaminant exposures in biological systems

Other Project Personnel

Samantha Bock, PhD Student - SREL

Jameel Moore, Undergraduate Researcher/Technician - Benedict College

Junsoo Baie, Undergraduate Researcher/Technician - Augusta University

External Collaborators

NA

- Parrott, B.B. Environmental sex determination in the Anthropocene. 2019. Invited Seminar. Interdisciplinary Conservation Seminar Series. Athens, GA, USA
- Bock, S.L., Lowers, R.H., Rainwater, T.R., Stolen, E., Drake, J.M., Wilkinson, P.M., Weiss, S., Back, B., Guillette, L.J., Parrott, B.B. Ecological drivers of nest temperature variation in the American alligator: implications of environmental change for a species with temperature-dependent sex determination. *In review*.
- Bock, S.L., Hale, M.D., Leri, F.M., Rainwater, T.R., Parrott, B.B. Post-transcriptional mechanisms respond rapidly to ecologically-relevant thermal fluctuations during temperature dependent sex determination. *In prep.*
- Bock, S.L., Lowers, R.H., Rainwater, T.R., Stolen, E., Drake, J.M., Wilkinson, P.M., Weiss, S. Back, B., Guillette, L.J., Parrott, B.B. Ecological drivers of nest temperature variation in the American alligator: predicting the impact of future climatic scenarios. 2019. Oral Presentation, Palmetto Alligator Research & Management Symposium, Georgetown, SC, USA
- Bock, S.L., Lowers R.H., Rainwater, T.R., Hale, M.D., Parrott, B.B. Linking proximate mechanisms of developmental plasticity to environmental variation in nature. 2019. Oral Presentation, Odum School of Ecology Graduate Student Symposium, Athens, GA, USA
- Bock, S.L., Lowers, R.H., Rainwater, T.R., Hale, M.D., Parrott, B.B. Insights from the field: using a multi-year dataset of nest thermal profiles to investigate temperature-dependent sex determination in the American alligator. 2019. Oral Presentation, Annual meeting of the Society for Integrative and Comparative Biology.
- Bock, S.L., Lowers, R.H., Rainwater, T.R., Hale, M.D., Parrott, B.B. Proximate and persistent effects of ecologically-relevant thermal fluctuations during temperature-dependent sex determination in the American alligator. 2019. Poster Presentation, North American Society for Comparative Endocrinology Biennial Meeting, Gainesville, FL, USA
- Bock, S.L and Parrott, B.B. *Characterizing variation in nesting thermal dynamics of the American alligator and consequences for reproductive development.* 2-3 February, 2018, Oral Presentation, Odum School of Ecology Graduate Student Symposium, Athens, GA.

Addressing reproductive dysfunction in an environmental model of endocrine disruption

Funding Entity SREL Start Date and Funding Amount NFP SREL Collaborators Dr. Ben Parrott

Objectives

The overall goal of this research is to determine how ecologically relevant thermal dynamics impact developmental responses to endocrine disrupting contaminant exposures.

Summary of Research Activities

The etiology of many reproductive disorders is complex and likely involves interactions between an individual's genetics and external environmental factors, including exposures to anthropogenic stressors. Environmental contaminants that interfere with the native functioning of the endocrine system have been linked to reproductive abnormalities and population declines in wildlife and humans globally. Alligators from environments contaminated by endocrine disrupting compounds (EDCs) display disorders of the reproductive system including alterations in circulating sex hormone levels, a decreased robustness of sexually dimorphic gene expression, and morphological abnormalities of ovarian follicles. Investigations into a population of alligators inhabiting a contaminated system in Florida, Lake Apopka, have uncovered the roots for a subset of these abnormalities in altered estrogen signaling during embryonic development, including shifts in ovarian function and transcription that persist into later life stages.

Using a model in which juvenile alligators, collected as eggs from Lake Apopka and a reference site, were raised under identical laboratory conditions and challenged with either a vehicle control or a gonadotropin hormone (FSH) that stimulates ovarian function, we employed targeted gene expression analyses and a non-biased RNAseq-based method to uncover the depth and possible etiology of population-level differences in ovarian function associated with contaminant exposure. After identifying core transcriptional networks shared between both populations, including FSH-responsive genes involved in steroid hormone production, cell proliferation, and oocyte development, we uncovered a cohort of responsive genes unique to each site that are putatively linked to developmental contaminant exposure. Furthermore, in the non-challenged ovary, we identified a large proportion (~40%) of transcripts that differ by population. Collectively, these findings indicate a substantial role for developmental contaminant exposure in shaping future ovarian function. Given prior evidence in the alligator linking similar functional shifts to precocious estrogen signaling, next steps entail identifying cohorts of dysregulated genes at Apopka that are recapitulated in estrogen-exposed reference animals and functional pathways enriched in population-specific responses.

Conclusions

To date, findings show widespread transcriptomic divergence between ovaries from a contaminated site and a nearby reference site. Further, we show that treating embryos from the reference site with estrogen recapitulates alterations observed in those animals from a contaminated site, suggesting that exposure to estrogenic contaminants during developmentally sensitive windows underlies reproductive abnormalities observed in alligators from Lake Apopka.

Major Impact(s) of Research

- 1. Establish fundamental insights into how EDC exposure during development affects reproductive development and reproductive health
- 2. Provides a basic understanding of how variable environmental conditions interact with contaminant exposures in biological systems

Other Project Personnel

Matthew Hale, PhD Student - SREL Samantha Bock, PhD Student - SREL

External Collaborators

NA

- Parrott, B.B., Hale, M.D. Precocious estrogen signaling during embryonic development underlies persistent alteration of ovarian transcriptional networks in an environmental model of endocrine disruption. 2019. Invited Talk. North American Society for Comparative Endocrinology Conference. Gainesville, FL, USA
- Parrott, B.B. Wildlife as models for understanding mechanisms of reproductive disorders. 2019. Invited Seminar. University of Georgia, Department of Cellular Biology Seminar Series, Athens, GA, USA
- Parrott, B.B. Adaptive and disruptive mechanisms underlying epigenome-by-environment interactions. 2019. Invited Seminar, University of South Carolina- Aiken, Department of Biology Seminar Series, Aiken, SC, USA
- Hale, M.D., Parrott, B.B. Precocious estrogen signals underlie altered ovarian function in a model of environmental health. *In review*
- Galligan, T.M., Hale, M.D., Bermudez, D., Cloy-McCloy, J.A., Guillette L.G., Parrott, B.B. 2019. Assessing impacts of precocious steroid exposure on thyroid physiology and gene expression patterns in the American alligator. *General and Comparative Endocrinology* 271(15):61-72
- Hale, M.D., Cloy-McCoy, J.A., Doheny, B.M., Galligan, T.M., Guillette, L.G., Parrott, B.B. 2019.
 Embryonic estrogen exposure recapitulates persistent ovarian transcriptional programs in a model of environmental endocrine disruption. *Biology of Reproduction* 100(1): 149-161
- Hale, M.D., McCoy, J., Galligan, T., Bangma, J., Nilsen, F., Doheny, B., Guillette, L.J., Parrott, B.B.
 2018. Precocious estrogen signaling during sex determination leads to persistent alterations in ovarian function in an environmental model of endocrine disruption, the American Alligator.
 Poster Presentation. 8th International Symposium on Vertebrate Sex Determination (VSD); Kona, HI.
- Hale, M.D., Galligan, T., Guillette, L.J., Parrott, B.B. 2018. Linking historical exposures to modern-day signaling: dioxin and the American alligator. Oral presentation. 22nd Odum School of Ecology Graduate Student Symposium; Athens, GA.
- Hale, M.D., Cloy-McCoy, J.A., Doheny, B.M., Parrott, B.B. 2018. Reproductive Biology of Crocodilians, published in *Encyclopedia of Reproduction* (2nd edition)

Forest Health Metrics: DNA methylation age predictors in Loblolly pine

Funding Entity

USDA USFS-SRS

Start Date and Funding Amount

September 2019; \$19,098 SREL Collaborators

Dr. Ben Parrott and Dr. Doug Aubrey

Objectives

The overall goal of this research is to develop an DNA methylation based epigenetic clock for Loblolly pine.

Summary of Research Activities

Aging is amongst the most universal biological processes, as organisms generally experience declines in molecular and physiological function over time, ultimately resulting in increased risk of death. DNA methylation is perhaps the best-studied epigenetic modification and when genomic patterning of the DNA methylome is resolved at the nucleotide level, the stereotypical nature of age-associated changes can be modeled to generate "epigenetic clocks". Epigenetic clocks summarize the readout of age-associated hyper- and hypo-methylation from a selection of loci across the genome which are collectively capable of predicting chronological age with high accuracy. Interestingly, the degree to which an individual's "epigenetic age" exceeds their true chronological age (also called epigenetic-to-chronological age discordance) is associated disease including cardiovascular disease, cancer, and even all-cause mortality (e.g., risk of dying). Thus, there is exciting potential for measures of epigenetic-to-chronological age discordance to advance our understanding of trait development in other species.

DNA methylation has well-studied roles in plants including the repression of transposable elements, as well as regulating gene expression, organismal development, and adaptive responses to environmental cues. However, the dynamics of epigenetic aging have not been fully described nor have attendant epigenetic clocks been developed in the plant kingdom. The overarching goal of the proposed work is to discover broadly conserved epigenetic signatures of aging in economically important tree species and to develop epigenetic clocks capable of predicting chronological age that can be used in ecological, conservation, and management contexts. Findings will provide insights into the basic biology underlying fundamental aging processes and will also provide a framework for exploring the consequences of accelerated epigenetic aging on traits associated with stand productivity, wood quality, and ecosystem processes and attributes, including resilience.

Conclusions

Conclusions are not yet available.

Major Impact(s) of Research

1. Establishing a epigenetic aging clock for trees will open the door for applications relevant to assessing commercial traits, forest productivity, plant disease, as well as the basic biology and evolution of life history traits.

Other Project Personnel

Emily Bertucci, PhD Student - SREL

External Collaborators

Dr. Andy Horcher (USDA Forest Service) <u>Products</u> Products are not yet available.

Environmental and Ecological drivers of biological aging trajectories

Funding Entity SREL **Start Date and Funding Amount** NFP **SREL Collaborators** Dr. Ben Parrott

Objectives

The overall goal of this research is to assess both the influence of environmental factors on biological aging pathways and the attendant consequences in the context of health and disease

Summary of Research Activities

Many diseases are age-related, with disease risk increasing with age. However, instead of understanding the commonalities that tie their etiology to biological aging pathways, biomedical and wildlife disease studies are typically aimed at preventing and treating specific diseases in isolation. Further, although it is clear that both ecological and environmental factors influence life history strategies and biological aging, the mechanism linking the outside environment to molecular pathways responsible for organismal phenotypes are not known. The work is focused on elucidating the mechanisms by which environmental stressors influence aging trajectories in a small, experimentally tractable medaka fish (Oryzias latipes) model.

The first objective of the study is to develop aging biomarkers for the medaka model that can be used as endpoints in dose-response studies. The work will develop and validate DNA methylation epigenetic aging clocks, telomere length assays, fin regeneration assays, and respirometry assays. These endpoints will be assessed in a known-aged cohort of fish to validate their relationship to age. The second objective will to test the influence of exposure to environmental stressors to aging biomarkers. For example, fish will be exposed to low doses of gamma radiation, methyl mercury, elevated temperatures, crowding, and copper. Fish will be exposed during different life stages in order to identify ontogenetic windows of environmental hypersensitivity. The third objective is aimed at identifying the endocrine pathways that mediate the influence of environmental factors on aging rates and trajectories. Glucocorticoid hormones are the primary stress hormone in vertebrates and the levels of glucocorticoids will be experimentally manipulated (increased via treatment with exogenous hormones or blocked using pharmacological approaches) to assess the influence of biological aging. Further, sex differences are observed in vertebrates and the work will investigate the role of sex steroid signaling (e.g., androgens, estrogens) in the sex-biased divergence of aging trajectories.

Collectively, this work will produce novel aging biomarkers that can be used to investigate environmental influences on aging trajectories with ultrafine resolution. The work will advance our understanding of the how environments impact health and disease through a unified lens of aging biology, and will also link evolutionary theories underlying life history variation to molecular pathways involved in biological aging. Conclusions

DNA methylation clocks have been developed and they are highly accurate. Validation is currently underway for additional aging biomarkers.

Major Impact(s) of Research

1) Findings will advance our understanding of age-associated disease and the influence of the environment.

Other Project Personnel Emily Bertucci, PhD Student - SREL **External Collaborators** NA

- Parrott, B.B., Bertucci, E.M. Epigenetic aging clocks in ecology and evolution. 2019. *Trends in Ecology* and Evolution 34(9): 767-770
- Bertucci, E.M., Parrott, B.B. Characterization of the age-related DNA methylome and development of an epigenetic age predictor in medaka (*Oryzias latipes*). 2019. University of Alabama Birmingham Integrative Aging Symposium, Birmingham, AL, USA
- Bertucci, E.M., Parrott, B.B. Characterization of the age-related DNA methylome in medaka (*Oryzias latipes*). 2019. Savannah River Ecology Laboratory Graduate Student Symposium, Aiken, SC, USA
- Bertucci, E.M., Parrott, B.B. Effects of environmental stressors on aging trajectories in medaka. 2019. Odum School of Ecology Graduate Student Symposium, UGA, Athens, GA, USA

<u>Ecological Factors Affecting the Success of Rabies Elimination in the Southeastern US</u> Funding Entity

USDA-APHIS

Start Date and Funding Amount

December 2018; \$132,822

SREL Collaborators

O. E. Rhodes, Jr., G. Dharmarajan, and J. C. Beasley

Objectives

- 1. Quantify variance in raccoon and opossum densities among three major habitat types found commonly in the Southeastern US
- 2. Evaluate the role of isolated wetlands surrounded by presumably inhospitable upland pine habitats in supporting raccoon and opossum populations
- 3. Quantify realized levels of rabies vaccine bait uptake by raccoons in the presence of bait competition from opossums and other mesopredators and mammals in the Southeastern US
- 4. Evaluate the relative densities of raccoons measured by camera and live traps in the Southeast US.

Summary of Research Activities

A total of 175 unique raccoons and 54 unique opossums were captured during the spring 2019 across the two rounds of trapping. On average recaptures from Year 1 or Year 2 (i.e., 2017 or 2018) were 27% and 3.7% for raccoons and opossums, respectively. Sex ratios continue to be male-biased as in Year 1 for both raccoons and opossums. Initial analyses of the opossum data reveals that habitat plays an important role in determining opossum densities in the Southeastern US, with high opossum densities being associated with bottomland swamp and riparian hardwood habitats compared to pine and isolated wetland.

As part of this study we also undertook raccoon translocation to evaluate how n=anthropogenic movement of raccoons can affect space use and impact disease transmission dynamics. Preliminary analyses of the GPS telemetry data from translocated animals indicates that translocation led to a ~20-fold increase in home range (Transformed estimate = 26.656; B ± SE = 3.283 ± 0.575 ; DF = 6.000; t = 5.710; P = 0.001). We also have fitted 71 unique opossums with GPS transmitters. Preliminary analyses reveal that home ranges (minimum convex polygon 90%) are smaller in the non-breeding season (A=69.65 ha, SD=23.39) than breeding season home ranges (A= 137.33 ha, SD= 130.12). Interestingly, there also appears to be evidence of landscape partitioning by males within close proximity during the breeding season. In Spring 2019, we also deployed cameras in a total of 12 grids in pine and bottomland hardwood sites and carried out a total of 360 trap-nights of camera surveys in these two habitats. We found photographic evidence of raccoons in all sites, as well as evidence for a number of other potential ORV bait competitors including: bobcats, coyotes, feral swine and opossums.

Conclusions

This study is ongoing and no conclusions are available at this time.

Major Impact(s) of Research

The major impacts of this research will be to: (1) Obtain baseline ecological information for making disease management decisions regarding important rabies reservoir (raccoon) and nontarget (opossum) species in the Southeastern US; (2) Quantify levels of oral rabies vaccine (ORV) bait uptake by raccoons and levels of ORV bait competition due to other non-target species (e.g., opossums); (3) Additionally, this project will investigate and validate an index of raccoon density using passive methods of detection in an effort to reduce field personnel requirements during future program monitoring and planning efforts.

Other Project Personnel

Matt Hamilton, Research Professional II – SREL David Bernasconi, MS student – SREL Lonnie Helton, MS student – SREL Wesley Dixon, MS student – SREL Tori Locke, Research Technician – SREL Kylie Bosch, Research Technician – SREL Patrick Ryan, Research Technician – SREL Marie McGee, Research Technician – SREL Bradley Carter, Research Technician – SREL Jessica Mehta, Research Technician – SREL Cody Wertman, Research Technician – SREL Erik Neff, Graduate student – SREL Amelia Russell, Research Technician – SREL Austin Coleman, Research Technician – SREL Chris Leaphart, Graduate student – SREL <u>External Collaborators</u> Richard B. Chipman (USDA-APHIS) Amy T. Gilbert (USDA-APHIS)

Products

Bernasconi, D. A., M. T. Hamilton, R. B. Chipman, A. T. Gilbert, J. C. Beasley, G. Dharmarajan, O. E. Rhodes, Jr. (2019) A multiyear analysis of the influence of landscape attributes on Virginia Opossum abundance. Joint Conference of the American Fisheries Society and The Wildlife Society. Sept. 29-Oct. 3, 2019. Reno, Nevada

Evaluation of contact structure and disease dynamics in free-ranging wild pigs

Funding Entity

USDA – Wildlife Services – National Wildlife Research Center

Start Date and Funding Amount

February 2017; \$58,883

SREL Collaborators

Dr. James C. Beasley

Objectives

The objectives of this study are to quantify the contact structure of invasive wild pig populations, and the influence of supplemental feeding and experimental population reduction on contact rates among groups. These data also will be used to evaluate the degree of territoriality among adjacent wild pig social groups (i.e. sounders).

Summary of Research Activities

During spring 2017 we captured 7 adjacent sounders as well as several adult male wild pigs on the SRS and fitted 1-3 members of each group with a GPS transmitter and contact logger. Samples were collected from all trapped pigs for DNA and disease analyses. After several months of establishing baseline data, we created bait stations to assess the influence of baiting on movements. Beginning in September 2017 we began culling groups at the core of the distribution of collared animals to assess the impact on animal movements. We have now retrieved the collars from these animals are currently analyzing the data collected.

Conclusions

Analyses for this research have just begun; there are no conclusions at this time.

Major Impact(s) of Research

- 1. This research will provide some of the first data to date on the contact structure of wild pig social groups.
- 2. Data collected during this study will provide information on how wild pig movements are impacted by the removal of adjacent social groups, data essential to the development of robust management plans for this species.
- 3. Data derived from this study will be integrated into national-level research by the USDA to implement management of wild pigs.

Other Project Personnel

Sarah Chinn, Ph.D. Student - SREL

Peter Schlichting, Postdoc – SREL

External Collaborators

Dr. Toni Piaggio (USDA-APHIS-WS-NWRC)

Dr. Kim Pepin (USDA-APHIS-WS-NWRC)

Dr. Amy Davis (USDA-APHIS-WS-NWRC)

Dr. Samantha Wisely (University of Florida)

- Dr. Raoul Boughton (University of Florida)
- Dr. Kurt VerCauteren (USDA-APHIS-WS-NWRC)

- Eckert, K.D., D.A. Keiter, and J.C. Beasley. 2019. Animal visitation to wild pig (*Sus scrofa*) wallows and implications for disease transmission. Journal of Wildlife Diseases 55:488-493
- Beasley, JC. 2018. Wild Pig Research on the Savannah River Site. South Carolina Wild Pig Task Force Meeting, Columbia, SC (oral presentation)
- Wilber, M.Q., S.M. Chinn, J.C. Beasley, C. Webb, and K. Pepin. 2019. Functional responses predict resource use of a rapidly expanding invasive mammal. Society for integrative and comparative biology. Tampa, FL (oral presentation)

Evaluation of crop damage by wild pigs

Funding Entity

USDA-APHIS – Wildlife Services – National Wildlife Research Center

Start Date and Funding Amount

February 2017; \$36,000

SREL Collaborators

Dr. James C. Beasley

Objectives

The objectives of this study are 1) to quantify timing and spatial distribution of wild pig damage to corn and peanut fields in SC, as well as 2) assess the utility of UAV's for quantifying wildlife damage to agricultural crops.

Summary of Research Activities

We selected crop fields and conducted walking surveys for fields adjacent to the Congaree National Forest, the Savannah River Site in SC, and in South-Central South Carolina during the planting seasons of 2017-2019. During 2018 and 2019 we also conducted aerial surveys of fields using UAV's to evaluate the efficacy of UAV's for quantifying wildlife damage to crops. All surveys have been completed and we are currently analyzing the data and preparing publications from this research.

Conclusions

This research is still in progress. Preliminary conclusions suggest the majority of wild pig damage to corn fields occurs shortly after planting, followed by during development and maturation of ears. Damage to peanuts occurred almost exclusively following planting. Damage to both crops increased in areas with greater area of forest and wetlands adjacent to crop fields, while fields surrounded by agriculture and the presence of paved roads were associated with lower damage estimates. Although analyses of aerial survey data are ongoing, drone surveys were effective at distinguishing wildlife damage to both corn and peanut fields.

Major Impact(s) of Research

- 1) This research will provide important data on the timing and spatial distribution of wild pig damage to corn and peanut fields, two important agricultural crops in the southeast
- 2) Data derived from this study will be integrated into national-level management of wild pigs by the USDA to mitigate damages caused by invasive wild pigs.

Other Project Personnel

Chris Boyce, M.S. Student - SREL

External Collaborators

Dr. Kurt VerCauteren (USDA-APHIS-WS-NWRC)

Steve Smith (USDA-APHIS)

- Beasley, JC. Wild Pig Research on the Savannah River Site. 2018. South Carolina Wild Pig Task Force Meeting, Columbia, SC (oral presentation)
- Boyce, C., and J.C. Beasley. 2018. Agricultural damage timing and extent by wild pigs along the Congaree River. Warnell Graduate Student Symposium. Athens, Georgia (oral presentation)
- Boyce, C. 2019. Timing and extent of crop damage by wild pigs (*Sus scrofa*) to corn (*Zea mays*) and peanuts (*Arachis hypogaea*). Thesis. University of Georgia, Athens GA. 85pp.
- Boyce, C., K. VerCauteren, and J.C. Beasley. 2019. Agricultural damage timing and extent by wild pigs along the Congaree River. Savannah River Ecology Laboratory Graduate Student Symposium. Aiken, South Carolina (poster presentation)

To initiate data collection from ongoing and additional feral swine field studies

Funding Entity

USDA-APHIS – Wildlife Services – National Wildlife Research Center

Start Date and Funding Amount

September 2018; \$338,960.00

SREL Collaborators

Dr. James C. Beasley, Lindsay Clontz, and Allison Rakowski

Objectives

The objectives of this study are 1) to assess the efficacy of Kaput[®] feral hog bait for controlling wild pigs and the behavioral and physiological impacts to wild pigs following consumption, 2) quantify the accessibility of feeders designed for use with Kaput[®] feral hog bait by pigs and non-target species, 3) determine the impacts of Kaput[®] feral hog bait on non-target species, and 4) determine warfarin levels in tissues of wild pigs following consumption of Kaput[®] feral hog bait.

Summary of Research Activities

During fall 2018 through spring 2019 we conducted pen trials where we quantified the response of pigs fed Kaput[®] feral hog bait. Following the pen trials, we captured, tagged, and attached radio collars to wild pigs, raccoons, opossums, and mice within select areas of the SRS. During summer 2019 we deployed pig-specific feeders containing corn, followed by Kaput[®] feral hog bait, to determine the accessibility of the bait to both wild pigs and non-target species. These trials are currently ongoing through fall 2019. **Conclusions**

This research is still in progress; there are no conclusions at this time.

Major Impact(s) of Research

- 1) This research will provide the first independent evaluation of Kaput[®] feral hog bait for use in controlling invasive wild pigs.
- 2) Data derived from this study will be integrated into national-level management of wild pigs by the USDA to mitigate damages caused by this invasive species.
- 3) Results from this study also will be used by individual states to further develop management plans for controlling invasive wild pigs.

Other Project Personnel

Ms. Amy Hilger, Research Technician - SREL

- Ms. Sara Cheatham, Research Technician SREL
- Mr. Lexington Belyeu, Research Technician SREL
- Mr. Chad Argabright, Research Technician SREL

External Collaborators

Dr. Kim Pepin (USDA-APHIS-WS-NWRC)

Dr. Nathan Snow (USDA-APHIS-WS-NWRC)

Dr. Kurt VerCauteren (USDA-APHIS-WS-NWRC)

Products

Beasley, JC. Evaluation of Kaput for use in wild pigs. 2019. National Wild Pig Task Force Meeting, Starkville, MS (oral presentation)

Beasley, JC. Evaluation of Kaput for use in wild pigs. 2019. Georgia Wildlife Services Annual Meeting, Athens, GA (oral presentation)

Collaborations and Externally Funded Research Non - SRS

The Ecological Study of Birds in the Vicinity of Augusta Regional Airport at Bush Field

Funding Entity

City of Augusta, GA <u>Start Date and Funding Amount</u> October 2018; \$92,421

SREL Collaborator Robert Kennamer

Objectives

Conduct bird hazard research associated with the placement of a wastewater treatment wetland system adjacent to a commercial airport and provide wildlife hazard consultation to airport and wastewater treatment plant personnel.

Summary of Research Activities

Since December 2001, we have been monitoring temporal and spatial activities of birds around Augusta Regional Airport. Within 2 years of the 2001 completion, the 360-acre wastewater treatment wetlands served as a nighttime roost for millions of migratory blackbirds that crossed the airfield daily at sunrise and sunset during fall/winter periods. We have been investigating the use of habitat alteration techniques to displace blackbirds, including use of airboats since 2008 to mechanically crush wetland vegetation in treatment wetlands each fall. Results have been highly significant, with long-term post-crush bird monitoring indicating that blackbird roosting within the wetlands became almost non-existent and blackbird activity around the airport was reduced. The ability of the wetland vegetation to process wastewater effluent was not negatively affected by vegetation alteration.

Conclusions

- 1) Long-term monitoring showed a reduction in blackbirds around the airport by 2 orders of magnitude and this reduction has been maintained for seven years now.
- 2) Fall crushing of wastewater treatment wetlands vegetation has not harmed the vegetation and regrowth has been experienced each subsequent spring.

Major Impacts of Research

- 1) The work demonstrated that with thoughtful wildlife hazard management, including the use of novel techniques, one can mitigate undesirable wildlife attraction associated with certain land-use activities.
- 2) Successful reduction of the bird-aircraft strike hazard was accomplished through non-lethal means.
- 3) Wastewater effluent concentrations of TSS, NH₃-N, and BOD₅ were reduced (improved) as a result of the vegetation crushing (i.e., vegetation crushing contributed an added benefit).

Other Project Personnel

Matthew Strassburg, Research Technician III - SREL

External Collaborators

D. Allen Saxon, Jr. (Augusta, GA Utilities Department)

Tim Weegar (Augusta Regional Airport)

- Kennamer, R. A., M.D. Strassburg, and I. L. Brisbin, Jr. 2018. Abundance, Distribution, and Movement Patterns of Avifauna in the Vicinity of Bush Field Airport: 2017–2018 Report. Final report submitted to Augusta Utilities Department, 169pp.
- Kennamer, R. A. A review of the SREL annual report on bird activity around AGS. Wildlife Hazard Management Group Meeting, Augusta Regional Airport, GA. November 8, 2018. (Oral Presentation).
- Kennamer, R. A. An update of SREL's recorded bird activity around Augusta Regional Airport. Wildlife Hazard Management Group Meeting, Augusta Regional Airport, GA. June 18, 2019. (Oral Presentation).

<u>Use of fecal genotyping and spatial capture-recapture modeling to investigate coyote abundance in</u> <u>South Carolina</u>

Funding Entity

SCDNR Start Date and Funding Amount

January 2017; \$50,000

SREL Collaborators

Dr. Stacey L. Lance

Objectives

The overall goals of this research are to estimate coyote densities among regions in South Carolina and to evaluate densities relative to landscape composition and other variables.

Summary of Research Activities

Coyote scats from a previous study on the SRS were collected over four seasons for two years on the SRS. To date we have extracted DNA from all scats have completed an initial screen of all samples across 9-10 microsatellite loci. We have confirmed recaptures of individuals across seasons and years. The new project is an extension of that work and will include those samples. In the summer of 2019 a PhD student, Jordan Youngmann, led a team to collect scat along transects in 8 different locations representing three ecoregions within the state. In total 220 scats have been collected and we have extracted DNA from all of the samples. In addition, we have developed single nucleotide polymorphism loci for coyotes.

Conclusions

The data are still being collected.

Major Impact(s) of Research

1) Data will be acquired to estimate coyote abundance in different ecoregions

2) Data will be used to inform deer management policies

Other Project Personnel

Dr. Jason O'Bryhim, Research Professional - SREL

Adam McFall, Research Technician - SREL

External Collaborators

Dr. Gino D'Angelo (UGA)

Dr. Karl Miller (UGA)

Dr. John Kilgo (USDA Forest Service)

Jordan Youngman (UGA)

Products

Youngmann, J. L., G. J. D'Angelo, J. C. Kilgo, S. L. Lance, K. V. Miller, and C. Ruth. 2019.

Use of non-invasive genetic sampling to estimate population density, resource selection, and genetic structure of coyotes in South Carolina. Georgia Department of Natural Resources Annual Meeting. (Poster presentation).

<u>Multi-year mating dynamics and population structure in a coastal population of Alligator</u> <u>mississippiensis at the Tom Yawkey Wildlife Center</u>

Funding Entity

MUSC, SC-DNR, The Yawkey Foundation

Start Date and Funding Amount

April 2017; \$74,020

SREL Collaborators

Dr. Stacey L. Lance, Dr. Ben Parrott, and Joshua Zajdel

Objectives

The overall objectives in this project are 1) identify parentage for clutches of alligators from the Yawkey Wildlife Center (YWC) collected from 2011-2015, 2) characterize the breeding population with respect to animal size, range, and multiple paternity and 3) quantify the genetic diversity of alligators at YWC and compare to populations from other parts of South Carolina and Florida.

Summary of Research Activities

We have completed the objectives of identifying parentage and characterizing the breeding population at YWC. In total we examined 151 nests and were able to assign a mother to 78 nests and at least one father to 38. The majority of maternity assignments matched the female that was caught at the nest (80%). We have completed an initial analysis of single nucleotide polymorphisms in 24 alligators and designed \sim 2000 baits to use for screening animals from all localities sampled. We are analyzing sequence data for \sim 200 samples.

Conclusions

- 1. A few large males are achieving most of the male reproductive success.
- 2. Females appear to pay a fitness cost by mating multiply.
- 3. Data are still being collected for the population genetics objective.

Major Impact(s) of Research

- 1. Mating systems of alligators appear to be driven by sexual conflict between males and females.
- 2. Multiple mating may overall reduce alligator fertility and decrease genetic diversity.
- 3. Our population genetics data will impact management practices moving forward as pressure increases to expand the hunting of alligators.

Other Project Personnel

NA

External Collaborators

Dr. Thomas Rainwater (Clemson University) Phil Wilkinson (SCDNR)

Jamie Dozier (SCDNR)

- Zajdel, J., S. L. Lance, T. R. Rainwater, P. M. Wilkinson, M. D. Hale, and B. B. Parrott. Mating dynamics and multiple paternity in a long-lived vertebrate. *Ecology and Evolution* 9:10109-10121.
- Zajdel, J., S.Lance, T.Rainwater, P.Wilkinson and B.Parrott. Multiple paternity within American alligator nests at the Tom Yawkey Wildlife Center. 2019. Palmetto Alligator Research and Management Symposium. Georgetown, SC. (Platform presentation).
- Zajdel, J., S.Lance, T.Rainwater, P.Wilkinson and B.Parrott. Multiple paternity within American alligator nests at the Tom Yawkey Wildlife Center. 2018 Annual meeting of the Southeastern Partners in Amphibian and Reptile Conservation in Helen, GA. (Platform presentation).

Panmixia, promiscuity, and nest parasitism among Wood Storks (Mycteria americana)

Funding Entity

University of South Carolina

Start Date and Funding Amount

May 2016; \$14,988

SREL Collaborators

A. Lawrence Bryan Jr. and Dr. Stacey Lance

Objectives

To sample US Wood Storks (nestlings) from selected colonies throughout their range and use a subset of storks to develop a panel of SNP (single nucleotide polymorphism) capture probes. These probes will then be used for capturing loci from additional stork DNA samples for genotyping via next generation sequencing to provide a robust examination of genetic structure and mating system in US WOST colonies as well as in other parts of their range.

Summary of Research Activities

Blood samples were obtained from > 130 total stork nestlings from 4 southeastern U.S. colonies in 2016 and will be compared to archived samples from 2 additional colonies. Additionally, samples from Brazil have been acquired. A subset of 24 stork samples were used to create genomic libraries for sequencing and identification of SNP loci. A total of 12,228 capture probes were designed to target 6114 loci. Currently, library preps have been made and sequenced and we are analyzing the data.

Conclusions

None at present, pending completion of genetic analyses.

Major Impact(s) of Research

When completed, will have a thorough analyses of the genetic structure of the US population of Wood Storks and should know the prevalence, if any, of non-standard mating strategies.

Other Project Personnel

NA

External Collaborators

Dr. Kristina Ramstad (USC-Aiken)

Dr. Natalia Bayona (UGA)

Products

No publications, presentations, or reports have yet been prepared.

Conservation and management of gopher frogs in South Carolina

Funding Entity

Longleaf Alliance; Greenville Zoo; United States Fish and Wildlife Service **Start Date and Funding Amount** April 2019; \$1000; September 2019; \$19,500 **SREL Collaborators** Dr. Stacey Lance

Objectives

Gopher frogs (*Lithobates capito*) are an uncommon species historically distributed throughout the southeastern coastal plain of the United States. Gopher frog populations have been declining due to loss of both their terrestrial uplands and their breeding sites. They are now listed as endangered at the state level and being considered for listing at the federal level. Within SC, gopher frog conservation is a high priority of the Department of Natural Resources and the population strongholds are the Savannah River Site and Francis Marion National Forest. It is unclear whether populations exist between these two widely separated areas. Our objectives are to 1) identify private lands with suitable gopher frog wetland and upland habitat, 2) survey for gopher frogs on these lands and 3) make management recommendations.

Summary of Research Activities

To date we have compiled a list of potential landowners to work with based on habitat suitability models for the gopher frogs. We have met with land managers at seven plantations and selected six wetlands to monitor for the Longleaf Alliance National Fish and Wildlife Federation funds, three for the USFWS Coastal Program funds, and one for the Greenville Zoo funds. We have also coordinated with the USFWS and Orangeburg National Fish Hatchery to begin a headstarting program in Spring 2020.

Conclusions

None at present.

Major Impact(s) of Research

None at present

Other Project Personnel

Dr. Jason O'Bryhim, Research Professional - SREL Adam McFall, Research Technician - SREL

External Collaborators

Lisa Lord (Longleaf Alliance) Melanie Olds (USFWS) Jason Ayers (USFWS) Jarrett Hill (Orangeburg National Fish Hatchery) Products

No publications, presentations, or reports have yet been prepared.

Molecular diet analysis of coyote scat through implementation of DNA metabarcoding

Funding Entity

SCDNR Start Date and Funding Amount July 2019; \$80,059

SREL Collaborators

Dr. Stacey L. Lance

Objectives

The overall goal of this research is to use DNA metabarcoding to determine the diet of coyotes in three ecoregions of South Carolina. An emphasis is placed on assessing the percentage of coyotes that are feeding on turkeys and deer.

Summary of Research Activities

Coyote scats from a previous study on the SRS were collected over four seasons for two years on the SRS. To date we have extracted DNA from all scats have completed an initial screen of all samples across 9-10 microsatellite loci. We have confirmed recaptures of individuals across seasons and years. The new project is an extension of that work and will include those samples. In the summer of 2019 a PhD student, Jordan Youngmann, led a team to collect scat along transects in 8 different locations representing three ecoregions within the state. In total 220 scats have been collected and we have extracted DNA from all of the samples. In addition, we have developed single nucleotide polymorphism loci for coyotes.

Conclusions

The data are still being collected.

Major Impact(s) of Research

1. Data will be acquired to estimate coyote diet in different ecoregions

2. Data will be used to inform deer and turkey management policies

Other Project Personnel

Dr. Jason O'Bryhim, Research Professional - SREL

Adam McFall, Research Technician - SREL

External Collaborators

Dr. Gino D'Angelo (UGA) Dr. Karl Miller (UGA)

Dr. John Kilgo (USDA Forest Service)

Jordan Youngman (UGA)

Products

No publications, presentations, or reports have been prepared yet.

Levels of Alpha-gal in ticks: Implications for red meat allergy

<u>Funding Entity</u> SREL <u>Start Date and Funding Amount</u> October 2016; NFP <u>SREL Collaborators</u>

G. Dharmarajan

Objectives

The purpose of this study was to characterize α -gal in *Amblyomma americanum* and *A. maculatum* ticks reared in the lab and those collected naturally from hunter killed deer on the SRS and adjoining areas

Summary of Research Activities

Delayed anaphylaxis linked red meat is a newly recognized allergic disease. Individuals bitten by the lone star tick (Amblyomma americanum) may develop IgE antibodies to the carbohydrate galactose- α -1,3-galactose (α -Gal; a carbohydrate moiety). Upon exposure of sensitized subjects to mammalian meat containing α -Gal on glycoproteins or glycolipids, delayed anaphylaxis may ensue, often three to six hours after ingestion. Tick saliva and salivary gland samples were processed to recover N-glycans. The N-linked glycans were permethylated for structural characterization by mass spectrometry. Briefly, the permethylated glycans were reconstituted in 100% MeOH and introduced to the mass spectrometer (Thermo Fusion Tribrid Orbitrap) with offline emission. These data were used to search for a Hex-Hex-HexNAc signature, and when glycoforms matching a Hex-Hex-HexNAc signature were found samples were manually fragmented at masses corresponding to each of the possible N-linked glycoforms. **Conclusions**

A large number of N-glycans was found in each of the tick samples, and these varied greatly from extended high mannose structures to highly complex and hybrid structures. No indications of the α -Gal glycoforms were found in any of the samples from the Amblyomma maculatum (Gulf Coast Tick) samples, either by not finding a mass corresponding to the glycoform, or if the mass was found, MS/MS fragmentation indicated that the glycoform did not contain the Hex-Hex-HexNAc signature fragment. No indications of the α -Gal glycoforms were found in the salivary glands from unfed *Amblyomma americanum*. However, multiple alpha-Gal glycoforms were found in salivary glands and saliva from partially or fully blood fed *Amblyomma americanum*.

Major Impact(s) of Research

- 1) Our preliminary data reveal α -Gal glycoforms are only associated with saliva and salivary glands of partially or fully fed *A. americanum*.
- The presence of α –Gal in A. americanum has significant implications for red meat allergy in South Carolina, especially amongst individuals that have high risk of tick bite (e.g., hunters)

Other Project Personnel

Amanda Hurst, Technician – SREL

External Collaborators

- P. Azadi (UGA)
- S. Karim (University of Southern Mississippi)

- Crispell G., S. Commins, S. A. Archer-Hartman, S. Choudhary, G. Dharmarajan, P. Azadi and S. Karim (2019) Identification of Alpha-Gal containing antigens in North American ticks and potential link with red meat allergy. *Frontiers in immunology* 10, 1056
- Archer-Hartmann, S. A., G. Crispell, S. Karim, G. Dharmarajan, P. Azadi (2018) Tick Bites and Hamburgers: N-Glycosylation analysis of saliva and salivary glands from the ticks responsible for Alpha-Gal Syndrome. Society for Glycobiology. November 5-8, 2018. New Orleans, LA
- One NSF Ecology and Evolution of Infectious Diseases grant has been submitted on the basis of these data

Evolutionary moving envelopes to predict range expansion (EMERGE)

Funding Entity

SREL, Department of Environmental Health Sciences UGA **Start Date and Funding Amount** August 2017; NFP **SREL Collaborators** G. Dharmarajan

Objectives

Emerging infectious diseases impact the lives of millions of people globally. Close to a third of emerging infectious diseases are those caused by pathogens transmitted by vectors, such as mosquitoes and ticks, and the aim of this study is to develop a new approach – Evolutionary Moving Envelopes to predict Range Expansion (EMERGE) to better predict range expansion of disease vectors.

Summary of Research Activities

We have carried out a thorough analysis of the patterns of range expansion in three tick species : the lone star tick (Amblyomma americanum), black-legged tick (Ixodes scapularis) and the cattle fever tick (Rhipicephalus microplus). Our analyses revealed that climate change alone is not sufficient to explain the observed patterns of range expansion in these species over the past several decades. We have also completed the development of the preliminary EMERGE model. The model uses an explicit, and novel, evolutionary genetic framework to integrate two powerful established methods: environmental niche modeling and landscape resistance. We have also used the 3RAD workflow to obtain RADseq data from seven A. americanum individuals from two populations. We obtained data from 128,899 loci in total, with 19,843 polymorphic loci genotyped in \geq 75% of individuals. The loci contained a total of 69,815 SNPs.

Conclusions

Climate change alone is insufficient to explain observed range expansion in ticks, and that dispersal and adaptation play important roles. Patterns of range expansion are not gradual but take the form of discrete jumps, an indication of gene swamping (i.e., the negative effects of migrant load on local adaptation). Large allele frequency differences between source and edge populations at selectively neutral loci, an indication of allele surfing (i.e., the combined effects of low density and strong founder effects)

Major Impact(s) of Research

- 1) Initial tests with EMERGE reveal that range expansion of ticks is likely driven by intricate interactions between climactic factors, dispersal and local adaptation.
- 2) EMERGE's novel framework in conjunction with its ability to explicitly integrate genomic information, provides us with a unique opportunity of obtaining a more mechanistic understanding of the factors driving range expansion in vectors.
- 3) By integrating niche modelling and genomic approaches our study will culminate in a robust new approach to modeling range expansion that can readily be applied to other species of epidemiological (e.g., other vector species) and ecological (e.g., invasive species) importance.

Other Project Personnel

NA

External Collaborators

T. Glenn (UGA)

- M. Yabsley (UGA)
- J. Busch (Northern Arizona University)
- P. Olafson (USDA)
- D. Thomas (USDA)

Products

Dharmarajan, G., T. C. Glenn, T. Kieran, N. Bayona-Vásquez and M. Yabsley (2019) The roles of climate, ecology and evolution in range expansion of disease vectors. 93rd Annual Meeting of the Southeastern Branch of the Entomological Society of America. March 3-6, 2019. Mobile, Alabama

Dharmarajan, G., Travis Glenn, Troy Kieran, Natalia Bayona-Vásquez and Michael Yabsley (2019) Integrating spatial models and genomic data to elucidate range expansion dynamics of disease vectors. Evolution 2020, June 21-25, 2019. Providence, RI

Three proposals have been submitted based on the EMERGE model to USDA-AFRI and NIH.

Pathogen-mediated behavioral modification in ticks: implications for tick-borne disease dynamics

Funding Entity

SREL, Department of Environmental Health Sciences UGA, University of Southern Mississippi Start Date and Funding Amount

March 2017; NFP

SREL Collaborators

G. Dharmarajan

Objectives

Lyme disease is a bacterial infection transmitted by ticks to more than 300,000 Americans annually. In the eastern US, the pathogen causing Lyme disease (*Borrelia burgdorferi*) is transmitted by the black-legged tick (*Ixodes scapularis*). This study proposes that behavioral manipulation of ticks will impact the spread and persistence of Lyme disease. Thus, the main aims of this study are to test if *I. scapularis* ticks colonized from natural populations in the North vs. the South: (1) differ in pathogen load when infected, and fitness (i.e., ability to survive desiccation and freezing) when infected or uninfected by *B. burgdorferi*, and how these factors are likely to impact vector competence; and (2) differ in tissue-specific gene expression profiles when infected or uninfected by *B. burgdorferi*, and if there is evidence of positive or negative selection at differentially expressed genes using genomic approaches

Summary of Research Activities

Initial mathematical modeling of the system has been completed, and these results have been analyzed within a spatial epidemiological framework. We have also collected preliminary data of tissue-specific transcriptomics from the lone star tick (*Amblyomma americanum*), which reveals that environmental stress and infection with pathogens *interact strongly to affect tick gene expression profiles*. We assembled 344 million Illumina reads, totaling over 34 billion nucleotides derived from 4 cDNA libraries constructed from adult female *A. americanum* that were unfed, or blood-fed for 12-48 hrs, 72-144 hrs, and 6-11 days. The libraries were assembled, and 5,792 coding sequences (CDS) that mapped 143 million reads were extracted. Digital expression of GRPs in four libraries revealed 9 differentially regulated *grp* genes, these differentially regulated *grp* genes were validated using qRT-PCR. We have also used the 3RAD workflow to obtain RADseq data from 16 Ixodidae individuals from four species (*I. scapularis, Amblyomma americanum, A. maculatum,* and *Dermacentor variabilis*). We obtained data from 332,057 loci in total, with 4,484 polymorphic loci genotyped in \geq 75% of individuals (i.e., polymorphic loci shared among three genera of ticks). These loci contained a total of 13,136 SNPs.

Conclusions

The project is at a very preliminary stage, and no conclusions can be drawn at this time

Major Impact(s) of Research

This project will help improve our understanding of Lyme disease mechanisms and dynamics, and is especially timely given rapid northward range expansion by *I. scapularis*

Other Project Personnel

J. Fredrick, PhD student – UGA

T. Kiernan, PhD student – UGA

N. Vasquez, Postdoc – UGA

External Collaborators (and Affiliations)

T. Glenn (UGA)

S. Karim (University of Southern Mississippi)

Products

Frederick, J. J., A. Thompson, G. Dharmarajan, M. Yabsley and T. C. Glenn (2019) Using Next Generation Sequencing and Behavior Biology to Understand Ixodes scapularis & Lyme Disease. Environmental Health Sciences Symposium. University of Georgia. February 28, 2019. Athens, GA

The preliminary data has used to submit a R01 grant to National Institutes of Health

Ecological and evolutionary dynamics of Avian malaria, an emerging infectious disease in wild birds

Funding Entity SREL Start Date and Funding Amount October 2016; NFP SREL Collaborators

G. Dharmarajan

Objectives

Emerging infectious diseases are considered to be one of the greatest challenges of our times, and their recent proliferation has been associated with anthropogenic factors such as global climate change. Avian malaria (AM) – a vector-borne disease caused by protozoan parasites *Plasmodium* spp. (Plasmodiidae) and *Haemoproteus* spp. (Haemoproteidae) is an important emerging disease in bird populations where the pathogen has been recently introduced (e.g., Hawaii). However, avian malaria is relatively benign in its native range, and previous research has shown that introduced *Plasmodium* parasites associated with large-scale mortalities originated from India, This study will characterize patterns of avian malaria in a natural bird community in India to improve our understanding of the disease. Birds were sampled using mist netting in the sky islands located in the southern 600 km mountain range of Western Ghats. The objective of this study is to elucidate the ecological and evolutionary dynamics of avian malaria in bird communities in the tropical sky-islands of the Western Ghats, India

Summary of Research Activities

Blood, sampled (50-100 µl) from the ulnar vein of birds was used to genomic DNA using Qiagen Blood and tissue extraction kit (Qiagen, Hilden, Germany) following manufacturer's protocol and screened for avian malaria infection. A nested Polymerase chain reaction (PCR) approach was employed to amplify the partial mitochondrial cytochrome b gene (478bp) of avian haemosporidian parasitesPositive samples were sequenced in both forward and reverse direction using BigDye v. 3.1 on an ABI 3130 automated DNA sequencer. Paired DNA sequences were aligned, trimmed and assembled in Geneious 9.1.5 for analyses.

Conclusions

A total of 1177 birds from 28 bird species were sampled, and our data revealed that 24 out of 28 species were infected with avian malaria parasites (*Plasmodium* spp. and *Haemoproteus* spp.) with an overall prevalence of 41.6% (490 individuals). We identified a total of 47 different mitochondrial cytb lineages, with 18 *Plasmodium* and 29 *Haemoproteus* parasites, but only six *Plasmodium* and four *Haemoproteus* lineages showed identity with sequences available in the Genbank/Malavi database. *Haemoproteus* lineages showed greater levels of species specificity as compared to *Plasmodium*

Major Impact(s) of Research

- 1) Our data reveal that relatively few lineages of avian malaria are shared between India and areas where the parasite has been newly introduced, thus indicating that disease emergence is associates with rare *Plasmodium* lineages
- 2) Our data also reveal that *Plasmodium* is generally less species specific compared to *Haemoproteus*, which could explain why EIDs are generally associated with the former rather than the latter pathogen.

Other Project Personnel

P. Gupta, PhD student - SREL

External Collaborators

R. V. Vijayan (Indian Institute of Science Education and Research)

Products

Gupta P, C. K. Vishnudas, U. Ramakrishnan, VV Robin and G. Dharmarajan (2019) Geographical and host species barriers differentially affect generalist and specialist parasite community structure in a tropical sky-island archipelago. Proceedings of the Royal Society B 286: 20190439

- Gupta, P., VV Robin and G. Dharmarajan (In Review) Towards a more healthy conservation paradigm: Integrating disease and molecular ecology to aid biological conservation. Journal of Genetics
- Gupta, P., C. K. Vishnudas, U. Ramakrishnan, V. V. Robin and G. Dharmarajan (2019)
 Disentangling the effects of host and environmental factors on parasite assemblages and infection dynamics in a montane biodiversity hostspot. 29th Annual Molecular Parasitology & Vector Biology Symposium. May 1, 2019. Athens, GA and Evolution 2020, June 21-25, 2019. Providence, RI
- Gupta, P., C. K. Vishnudas, U. Ramakrishnan, V. V. Robin and G. Dharmarajan (2019)
 Biogeography of haemosporidian parasites infecting diverse bird communities inhabiting the
 Shola sky islands of the Western Ghats, India. 9th Biennial Conference of the International
 Biogeography Society. January 8-12th, 2019. Málaga, Spain and Evolution 2020, June 21-25,
 2019. Providence, RI

One grant has been submitted to Department of Biotechnology, Govt. of India

Factors affecting parasite resistance and tolerance in mosquitoes: implications for vectorial capacity

Funding Entity

SREL, Filariasis Reagent and Resource Center UGA, Department of Infectious Diseases UGA Start Date and Funding Amount

August 2017; NFP

SREL Collaborators

G. Dharmarajan

<u>Objectives</u>

Recent decades has seen the reemergence of many mosquito-borne diseases in human and animal populations (e.g., filariasis and malaria). The evolution of resistance against anti-parasitic drugs is one of the most important drivers of reemergence The focus of this study is to test if mosquitoes act to enhance or reduce the risk of a drug resistant parasite genotype spreading in the vertebrate host population. The objectives of this study are to: (1) Test if resistant and susceptible strains of DI differ in terms of their pathogenicity and virulence in mosquitoes, and test how these differences impact mosquito resistance and tolerance to infection and thus mosquito vectorial capacity; (2) Test if there are competitive interactions between the strains within an individual mosquito, and determine if these competitive interactions impact vector competence; (3) Test if natural mosquito populations reveal signatures of local adaptation to sympatric vs. allopatric parasite populations, and test if local adaptation tends to increase or decrease vector competence

Summary of Research Activities

Studies of natural variation in resistance and tolerance to parasites have been completed. The experimental study design for the effects of resistance and tolerance on the spread of drug resistance has been finalized, and the drug resistant and susceptible strains of DI have been identified. These strains are currently being maintained in infected dogs at the Filariasis Reagent and Resource Center, UGA, and infection experiments are underway.

Conclusions

We find significant inter-population variation in tolerance and elevated tolerance where transmission intensity is high. Additionally, as expected, we find that increased tolerance is associated with higher vectorial capacity. Consequently, our results indicate that high transmission intensity can lead to the evolution of more competent disease vectors, which can feed back to impact disease risk. The effects of resistance and tolerance on the spread of drug resistant parasite genotypes is being investigated now.

Major Impact(s) of Research

- 1) The study has important implications for our understanding of the factors affecting the evolution of parasite resistance and tolerance in mosquitoes, and hence has implications for our understanding of the evolution of vector competence
- 2) This study will elucidate the role of the mosquito vector in the rise and spread of drug resistant genotypes of mosquito-borne parasites in natural populations, and will thus improve the control of these diseases.

Other Project Personnel

Erik Neff, MS student – SREL

External Collaborators (and Affiliations)

A. Moorehead (UGA)

R. Kaplan (UGA)

Products

Dharmarajan G., K. D. Walker, T. Lehmann (2019) Variation in tolerance to parasites in natural Asian tiger mosquito populations and its effect on vectorial capacity. Current Biology 29: 3946-3952

Dharmarajan, G., K. D. Walker, E. Neff and T. Lehmann (2019) Variation in tolerance to parasites affects vectorial capacity of natural Asian tiger mosquito populations. Evolution 2020, June 21-25, 2019. Providence, RI

Wood Stork (Mycteria americana) Species Status Assessment (SSA)

Funding Entity

U.S. Fish and Wildlife Service

Start Date and Funding Amount

June 2019; \$20,000

PI and Co-PI's

A. L. Bryan and O. E. Rhodes

Objectives

We were asked to assist in the drafting of a Species Status Assessment (SSA) for the wood stork, a species listed as federally "threatened" under the Endangered Species Act. An SSA utilizes historical and current data (habitat use, population distribution and trends, recovery efforts, etc.) to examine threats to the species as well as project best- and worst-case future scenarios for the species (e.g., different projected global climate change scenarios).

Summary of Research Activities

We initiated the drafting of the SSA in June of 2019, which involved acquisition of literature and data from regional sources. Chapters associated with the SSA are currently being drafted.

Conclusions

Drafting the SSA is on-going, so there are no conclusions at this time.

Major Impact(s) of Research

The Species Status Assessment summarizes known information regarding habitat use, population trends and potential treats to the recovery of the species. It is a compliance step for federally threatened and endangered species.

Other Project Personnel

NA

External Collaborators

NA

Products

No publications, presentations or technical repolts at this time.

Kings Bay Rare, Threatened and Endangered Wildlife Surveys: Wood Storks and Wading Birds

Funding Entity

DoD-Navy/LG2 Environmental Solutions Start Date and Funding Amount

September 2018; \$30,160

PI and Co-Pl's

Larry Bryan

Objectives

SREL has monitored Wood Stork use of selected aquatic habitats of the Kings Bay Submarine Base (coastal Georgia) periodically for> 15 years. In this installment, we monitored along the same survey route as the previous two monitoring years for comparison, as well as (1) monitoring breeding attempts at a historical colony site and (2) monitoring roost use at two known roosts. Other species of wading birds also will be monitored during all activities.

Summary of Research Activities

We initiated these surveys in October 2018 and completed them in September 2019. We have documented locations (habitat use) of Wood Storks, Roseate Spoonbills, White Ibis as well as herons (Great Blue, Little Blue, & Tricolor) and egrets (Great, Snowy, & Cattle).

Conclusions

Wood storks nested and foraged on the base during this period. We are currently summarizing these findings.

Major Impact(s) of Research

Field aspects of the project has just been completed, thus no impacts at present.

Other Project Personnel

NA

External Collaborators

NA

Products

No publications, presentations or technical repolts at this time.

Addressing Modern Day Health Risks Stemming from Historical Dioxin Contamination

 Funding Entity

 South Carolina Department of Health and Environmental Control

 Start Date and Funding Amount

 February 2017; \$12,194

 SREL Collaborators

 Dr. Ben Parrott

 Objectives

 The overall goal of this research is to determine if dioxin levels are correlated to biomarkers of exposure in developing alligator embryos.

Summary of Research Activities

2,3,7,8-tetrachlorodibenzo-*para*-dioxin (TCDD, or "dioxin") is the most toxic congener of the class of chemical chemicals known as dioxins, and is one of the most toxic anthropogenic contaminants known. With an array of deleterious effects associated with exposure, including mortality, multi-organ toxicity, carcinogenicity, teratogenicity, and tumor-promotion, US Federal and state regulatory agencies initiated a series of surveys in the late 20th century to characterize the scope of dioxin contamination in the environment and its sources. This investigation uncovered the role of Kraft pulp mills as sources of high levels of dioxin contamination in receiving waters and downstream aquatic systems (US EPA 1989). Of the 104 total surveyed, the International Paper Company's Georgetown facility (Georgetown County, SC) had the highest levels of dioxin (TCDD) in its effluent (US EPA 1989, 1990a, 1990b) at 0.640ppt.

We have recently detected significantly elevated gene expression of an established biomarker of dioxin exposure, *CYP1A2*, in liver tissue from embryonic American alligators at a site directly adjacent to this historically-dioxin contaminated system. This project directly measures TCDD and dioxin-like congeners and seeks to connect them to previously established biomarkers of exposure and developmental processes.

Questions: (1) At what levels are 2,3,7,8-TCDD and other dioxin-like congeners associated with a historical point source detectable as maternally-deposited contaminants in alligator egg yolk from the Tom Yawkey Wildlife Center and Heritage Preserve (TYWCHP)? (2) Do these levels in yolk correlate to hepatic expression of *CYP1A2* in the matching embryo?

Conclusions

We have found significantly elevated levels of dioxins at TYWCHP relative to a reference site. While yolk contaminant levels were not correlated to hepatic *AHR* or *CYP1A* expression, we found an unexpected association between the relative amount of yolk utilized by an embryo and expression of both the *AH* receptor isoform *AHR1B* and *CYP1A2*. This latter point is suggestive of possible novel mechanisms modulating the effects of dioxin exposure in the alligator.

Major Impact(s) of Research

- 1) Connecting specific dioxin congener levels in alligator yolks to alterations in embryonic development
- 2) Estimate the presence of historical contaminants in biological systems
- 3) Provide data and rationale for studies assessing human exposure and potential health impacts.

Other Project Personnel

Matthew Hale, PhD Student - SREL

External Collaborators

Dr. Thomas Rainwater (Clemson University)

Products

Hale MD, Bertucci EM, Rainwater TR, Wilkinson PM, Parrott BB. The impact of maternally derived dioxins on embryonic development and hepatic AhR signaling in a long-lived apex predator. 2019. Chemosphere 229:489-499

- Hale MD, Galligan T, Guillette, Jr. LJ, Parrott BB. 2018. Linking historical exposures to modern day signaling: dioxin and the American alligator. 22nd Odum School of Ecology Graduate Student Symposium; Athens, GA. February 2-3, 2018 (oral presentation)
- Hale MD, Galligan TM, Rainwater TR, Moore BC, Wilkinson PM, Guillette LJ, Parrott BB. 2017. AHR and CYP1A expression link historical contamination events to modern day developmental effects in the American alligator. *Environmental Pollution* 230: 1050-1061

<u>Gray wolf movement behavior in relation to kill sites, raccoon dog spatial ecology, scavenging</u> <u>dynamics, and demography of Przwalski's horses in Chernobyl</u>

Funding Entity

University of Georgia <u>Start Date and Funding Amount</u> October 2016; NFP <u>SREL Collaborators</u> Dr. James C. Beasley <u>Objectives</u>

The objectives of this research are to 1) quantify kill rates of gray wolves inhabiting the Chernobyl Exclusion Zone and elucidate the movement behavior gray wolves relative to kill site locations, 2) quantify home ranges, resource selection patterns, and movement behavior of raccoon dogs and relative radiation exposure, 3) assess demographic rates of endangered Przwalski's horses in Chernobyl and use of abandoned buildings, and 4) quantify the efficiency and community composition of vertebrate scavengers.

Summary of Research Activities

During fall 2016 we travelled to the CEZ and deployed GPS collars on wolves and raccoon dogs throughout the CEZ. We collected samples from captured wolves and other wildlife and also deployed remote cameras to elucidate the efficiency and composition of the vertebrate scavenging community. We also deployed remote cameras to assess use of abandoned barns by Przwalski's horses as well as develop preliminary assessments of population demography. During fall 2017 we returned to Belarus to collect GPS collars. We are currently analyzing data collected on these research trips and have begun preparing various publications from this work.

Conclusions

This research is still ongoing, but some analyses are now complete. We recorded >50 kill site locations from GPS-collared wolves, and collected tens of thousands of GPS locations on collared animals that will be incorporated into analyses. Additionally, we completed a scavenging study using remote cameras as well as a study assessing use of abandoned buildings by Przwalski's horses in the CEZ. Results of our scavenging study suggest a highly efficient and diverse scavenging community exists within the CEZ, and that fish carrion is an important resource used by terrestrial vertebrates, thus transporting aquatic nutrients into terrestrial ecosystems. Further, we documented extensive use of abandoned buildings by Przwalski's horses and other large mammals in Chernobyl, suggesting these structures are important to wildlife in the CEZ, particularly during periods of inclement weather.

Major Impact(s) of Research

- 1) This research will provide critical data on the demography of one of the world's only free-ranging populations of Przwalski's wild horses.
- 2) Wolf and raccoon dog GPS data will provide novel information on the movement behavior and diet of carnivores inhabiting the CEZ, data that will be valuable for future management of populations in the CEZ.
- 3) Conducted the first study of vertebrate scavenging ecology in the CEZ, revealing an intact and highly efficient scavenging community.

Other Project Personnel

Sarah Webster, M.S. Student - SREL

Cara Love, Ph.D. Student – SREL

Peter Schlichting, Postdoc – SREL

External Collaborators

Dr. Thomas Hinton (Fukushima University)

Dr. Dima Shamovich (Researcher and Wildlife Tour Guide, Belarus)

Dr. Valery Dombrovski (National Academy of Science, Belarus)

Products

- Schlichting, P.E., C.N. Love, S.C. Webster, and J.C. Beasley. 2019. Efficiency and composition of vertebrate scavengers at the land-water interface in the Chernobyl Exclusion Zone. Food Webs 18:e00107
- Beasley, J.C., M. Byrne, S. Webster, T. Hinton, D. Shamovich, P. Schlichting, S. Lance, and C. Love.
 2017. Home Range Size and Movements of Wolves in the Chernobyl Exclusion Zone. Workshop to develop a management plan for wolves in Belarus. Minsk, Belarus (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. SREL Research Experiences for Undergraduates. Aiken, SC (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Colorado State University. Fort Collins, Colorado (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Warnell Seminar. Athens, GA (oral presentation)
- Webster, S., and Beasley, J. C. (2017). Ecological effects of the Chernobyl accident. Grovetown Middle School (oral presentation)
- Beasley, J.C. 2018. 137Cs Accumulation in Wildlife. International Atomic Energy Agency Mission to Fukushima Prefecture, Japan. Fukushima City, Japan (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. NASA Goddard Space Flight Center. Greenbelt, Maryland (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Citizens for Nuclear Technology Awareness Up and Atom. Aiken, South Carolina (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Reactor Tech Reunion. North Augusta, South Carolina (oral presentation)
- Schlichting, P.E., V. Dombrovski, and J.C. Beasley. In Press. Use of abandoned buildings by Przewalski's wild horses and other wildlife in the Chernobyl Exclusion Zone. Mammal Research
- Beasley, J.C. 2019. Radioactive Wildlife: Impacts of the Chernobyl and Fukushima nuclear accidents on wildlife. Auburn University. Auburn, Alabama
- Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Mississippi State University. Starkville, Mississippi (oral presentation)
- Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Cedar Shoals High School. Athens, GA (oral presentation)
- Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. University of South Carolina – Aiken. Aiken, SC (oral presentation)
- Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. University of South Carolina – Aiken. Aiken, SC (oral presentation)
- Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Midland Valley High School. Aiken, SC (oral presentation)

Distribution and relative abundance of wildlife in Fukushima along a gradient of contamination and human land-use intensity, and spatial ecology of invasive civets

Funding Entity

University of Georgia Office of the Vice President for Research, Office of International Partnerships **Start Date and Funding Amount**

June 2016; \$15,350

SREL Collaborators

Dr. James C. Beasley

Objectives

The overall objectives of this project are to quantify the distribution and relative abundance of mammals and other scavengers in Fukushima, Japan across a gradient of radiation contamination and level of human disturbance. In addition, we will deploy radio collars on civets and potentially wild boar to assess their movement behavior and radiation exposure, which will be used to assess potential sub-lethal effects of chronic radiation exposure.

Summary of Research Activities

During spring 2016 we travelled to the Fukushima exclusion zone and conducted 120 2-month remote camera trials. Data from these cameras have now been analyzed, and we are preparing manuscripts for submission. During fall 2016 and summer 2017 we returned to Fukushima to conduct additional camera trials and attached GPS transmitters to civets and wild boar in the exclusion zone. Data from these studies have been analyzed and we are currently preparing publications stemming from this research.

Conclusions

Preliminary results suggest numerous species of mammals are abundant within the exclusion zone, and that the density of some species, especially wild boar, differs considerably between the exclusion zone and areas that remain occupied by humans. Further, resource selection patterns by civets and raccoons show selection of abandoned croplands and urban areas. Radiation was not found to be a factor driving occupancy, abundance, or movement patterns among the wildlife species we have studied.

Major Impact(s) of Research

This research represents the first assessment of the occupancy and abundance patterns of wildlife communities inhabiting the Fukushima Exclusion Zone. Further, this research is one of the first studies of the spatial ecology of civets and raccoons in Japan. Thus, data generated from this research will provide important insights into the management and conservation of wildlife in Fukushima.

Other Project Personnel

Phillip Lyons, M.S. Student – SREL Matthew Hamilton, Research Professional - UGA External Collaborators

Dr. Thomas Hinton (Fukushima University)

Dr. Kei Okuda (Fukushima University)

Products

- Lyons, P., T.G. Hinton, K. Okuda, M. Hamilton, and J.C. Beasley. 2018. Fukushima's Wildlife: Camera Analysis of Species in and Around the Exclusion Zone. Georgia Chapter of the Wildlife Society. Tifton, GA (oral presentation)
- Lyons, P., T.G. Hinton, K. Okuda, M. Hamilton, and J.C. Beasley. 2018. Fukushima's Wildlife: Mammalian Species in and Around the Exclusion Zone. Warnell Graduate Student Symposium. Athens, Georgia (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. SREL Research Experiences for Undergraduates. Aiken, SC (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Colorado State University. Fort Collins, Colorado (oral presentation)
- Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Warnell Seminar. Athens, GA (oral presentation)

Webster, S., and Beasley, J. C. (2017). Ecological effects of the Chernobyl accident. Grovetown Middle School (oral presentation)

Beasley, J.C. 2018. 137Cs Accumulation in Wildlife. International Atomic Energy Agency Mission to Fukushima Prefecture, Japan. Fukushima City, Japan (oral presentation)

Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. NASA Goddard Space Flight Center. Greenbelt, Maryland (oral presentation)

Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Citizens for Nuclear Technology Awareness Up and Atom. Aiken, South Carolina (oral presentation)

Beasley, J.C. 2018. Radioactive Wildlife: The Secret Life of Wildlife Inhabiting Chernobyl and Fukushima. Reactor Tech Reunion. North Augusta, South Carolina (oral presentation)

Beasley, J.C. 2019. Radioactive Wildlife: Impacts of the Chernobyl and Fukushima nuclear accidents on wildlife. Auburn University. Auburn, Alabama

Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Mississippi State University. Starkville, Mississippi (oral presentation)

Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Cedar Shoals High School. Athens, GA (oral presentation)

Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. University of South Carolina – Aiken. Aiken, SC (oral presentation)

Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. University of South Carolina – Aiken. Aiken, SC (oral presentation)

Beasley, J.C. 2019. Radioactive Wildlife: The Secret Life of Animals in Chernobyl and Fukushima. Midland Valley High School. Aiken, SC (oral presentation)

Beasley, J.C., T. Hinton, K. Okuda, P. Lyons, H. Gerke, and S. Chinn. Ecology, health, and radiation exposure of wildlife inhabiting the Fukushima Evacuation Zone. US DOE-HQ Annual Site Environmental Report Workshop. Aiken, SC (poster presentation)

¹³⁷Cs Activity Levels, Movement Behavior, and Efficiency of Vertebrate Scavengers in the Fukushima <u>Exclusion Zone</u>

Funding Entity

University of Georgia, Office of International Education Start Date and Funding Amount May 2018; \$5,000 SREL Collaborators Dr. James C. Beasley Objectives

The objectives of this project are to 1) quantify the composition and efficiency of vertebrate scavengers within the Fukushima Exclusion Zone, 2) deploy GPS transmitters on rat snakes within the Exclusion Zone to assess their movement behavior, and 3) quantify activity levels of ¹³⁷Cs in snakes throughout Fukushima Prefecture, Japan.

Summary of Research Activities

During summer 2018 we travelled to the Fukushima exclusion zone and conducted remote camera trials, as well as attached GPS transmitters to snakes. We also collected ¹³⁷Cs activity data from both live and road-kill collected snakes. Data from these efforts are currently being analyzed.

Conclusions

We are still analyzing the data from this research; thus, there are no conclusions at this time.

Major Impact(s) of Research

This research represents the first assessment of the efficiency of vertebrate scavengers within the Fukushima Exclusion Zone, and thus will produce novel data on the diversity and efficiency of vertebrate scavengers. These data will inform whether the introduction of radionuclides into the environment has disrupted basic ecological functions such as nutrient cycling. This research also will produce some of the first data to date on the movement behavior of snakes in Japan with GPS telemetry.

Other Project Personnel

Hannah Gerke, M.S. Student – SREL

Wes Dixon, Research Technician - SREL

External Collaborators

Dr. Thomas Hinton (Fukushima University)

Dr. Kei Okuda (Fukushima University)

Products

- Gerke, H., and J.C. Beasley. 2018. Radiocesium Accumulation and Spatial Ecology of Elaphe spp. in Fukushima, Japan. 2018 Annual Southeastern Partners in Amphibian and Reptile Conservation. Helen, Georgia (poster presentation)
- Gerke, H., and J.C. Beasley. 2018. Effects of Anthropogenic Activity on the Efficiency and Composition of Vertebrate Scavenging Communities in Fukushima, Japan. Warnell Graduate Student Symposium. Athens, Georgia (poster presentation)
- Gerke, H.C., T.G. Hinton, T. Takase, and J.C. Beasley. 2019. Radiocesium accumulation and spatial ecology of rat snakes (*Elaphe spp.*) in the Exclusion Zone in Fukushima, Japan. Savannah River Ecology Laboratory Graduate Student Symposium. Athens, Georgia (poster presentation)

Effects of Chronic Radiation Exposure on the Health of Wild Boar in Fukushima Prefecture, Japan

Funding Entity

Fukushima University, Institute of Environmental Radioactivity; University of Georgia, Office of International Education; University of Georgia, Graduate School; Environmental Radioactivity Research Network Center

Start Date and Funding Amount

May 2018; \$17,936

SREL Collaborators

Dr. James C. Beasley and Sarah Chinn

Objectives

The overall goal of this project is to assess the health impacts of chronic radiation exposure in wild boar inhabiting the area surrounding the Fukushima Dai-ichi nuclear accident. This work is being conducted in collaboration with researchers at Fukushima University, Colorado State University, Northern Michigan University, Fukushima Prefecture, and the University of Illinois, where we are testing captured wild boar for a broad suite of potential health impairments at the molecular through individual level.

Summary of Research Activities

During summer 2018 we travelled to the Fukushima exclusion zone to begin capturing boar and collecting samples to be used for this research. Additional research expeditions were conducted in 2019 to collect additional samples to be used for this research.

Conclusions

This research has just begun; there are no conclusions at this time.

Major Impact(s) of Research

This research will produce novel data on the impact of chronic radiation exposure on a large mammal. Thus, data generated from this research will provide important insights into the management and

conservation of wildlife in Fukushima, as well as other sites impacted by radiological contamination.

Other Project Personnel

NA

External Collaborators

Dr. Thomas Hinton (Fukushima University)

Dr. Kei Okuda (Fukushima University)

Ms. Aryn Bordman (Colorado State University)

Dr. Thomas Johnson (Colorado State University)

Dr. Sami Pederson (Colorado State University)

Ms. Maggie Roberts (Colorado State University)

Dr. Rao Veeramachaneni (Colorado State University)

Dr. Jodi Flaws (University of Illinois)

Dr. Yui Nemoto (Fukushima Prefecture)

Dr. Diana Lafferty (Northern Michigan University)

Products

- S. Pederson, M. Li Puma, J. Hayes, T. Hinton, K. Okuda, J.C. Beasley, T. Johnson, L. Li Puma, and K. Freeman. 2018. Cataracts in Boar following the Fukushima Dai-ichi Nuclear Disaster. Radiation Research Society Conference. Chicago, Illinois (Poster presentation)
- Chinn, S.M., T.G. Hinton, K. Okuda, H. Ishiniwa, Y. Nemoto, and J.C. Beasley. 2019. Reproductive health of wild boar in response to chronic radiation exposure. Savannah River Ecology Laboratory Graduate Student Symposium. Athens, Georgia (poster presentation)

Wild pig management in Sumter and Francis Marion National Forest

<u>Funding Entity</u> USDA-USFS

Start Date and Funding Amount

May 2018; \$30,259.00

SREL Collaborators

Dr. James C. Beasley

Objectives

The objective of this study is to assist USDA-USFS personnel working on the Long Cane Ranger District in South Carolina in the management of wild pigs through collection of biological data needed to inform management activities.

Summary of Research Activities

During summer 2018 and summer 2019 we collected samples from wild pigs captured by USFS personnel on the Long Cane Ranger District in SC. Samples primarily included tissue and blood, which will be incorporated into ongoing wild pig studies in South Carolina to evaluate the prevalence of pathogens and elucidate the genetic structure of wild pig populations. In addition, we deployed remote cameras throughout the study area to determine the relative abundance and distribution of wild pigs in the region.

Conclusions

This research is still in progress; there are no conclusions at this time.

Major Impact(s) of Research

- 1) This research will provide data needed to inform management activities targeting invasive wild pigs on the Long Cane Ranger District.
- Data derived from this study will be integrated into state and national-level databases, and ultimately will be used to inform the underlying genetic and disease dynamics of wild pigs in the southeastern U.S.

Other Project Personnel

Jakob Kemp, Research Technician – SREL Amy Hilger, Research Technician – SREL Kyle Cockrell, Research Technician – SREL External Collaborators

Mr. Donny Ray (USDA Forest Service)

Products

This research has just begun; there are no products at this time.

Evaluation of head-starting as a recovery tool for the Mojave desert tortoise

Funding Entity

National Park Service, California Energy Commission

Start Date and Funding Amount

Nov 2010; \$450,000 (NPS), \$313,000 (CEC, Phase I); \$493,089 (CEC, Phase II)

SREL Collaborators

Dr. Tracey Tuberville and Dr. Kurt Buhlmann

Objectives

- 1) Determine behavior, survivorship, and habitat use of head-started juvenile desert tortoises compared to direct-release hatchings (i.e., juveniles released shortly after hatching).
- 2) Develop habitat suitability models for juvenile desert tortoises to identify optimal desert tortoise habitat.
- 3) Evaluate the efficacy of indoor rearing as a head-starting technique.

Summary of Research Activities

Our research activities included monitoring of hatchling and juvenile desert tortoises in outdoor rearing pens, indoor rearing facilities, and radio-tracking of juveniles released into the wild.

Conclusions

Based on preliminary data analysis, most movement occurred within 30 days of release, and those with the greatest movements during that time were less likely to survive through the winter dormancy period. Animals head-started for 1 year in outdoor pens exhibited greater post-release survivorship than animals released immediately after hatching, but rearing animals for longer than 1 year in the enclosures may significantly reduce native forage inside pens. Indoor head-starting was effective in increasing growth, but did not result in increased survival compared to smaller outdoor head-starts when released at 1 yr of age. Hybrid head-starting (involving 1 year of indoor head-starting to increase growth followed by 1 yr outdoor head-starting to harden shells and provide natural cues) may prove a more effective head-starting method.

Major Impact(s) of Research

- 1. Important life history data for a poorly understood life stage of a federally listed species
- 2. Improve management for species by factoring juvenile requirements into management and policy decisions, such as identifying habitats likely to serve as important areas of juvenile recruitment. These data will be helpful when selecting potential solar development sites by identifying areas to avoid.

Other Project Personnel

Pearson McGovern, M.S. Student - SREL

Carmen Candal, M.S. Student - SREL

Collin Richter, Research Technician - University of California-Davis

External Collaborators

Dr. Brian Todd (University of California, Davis)

- Dr. Mark Peaden (University of California, Davis)
- Dr. Kristin Navarra (UGA)

Dr. Nicole Stacy (University of Florida)

Products

- McGovern, P.A. 2019. Changing the survival formula for the Mojave desert tortoise (*Gopherus agassizii*) through head-starting. M.S. Thesis, University of Georgia.
- Tuberville, T.D., K.A. Buhlmann, R. Sollmann, M.G. Nafus, J.M. Peaden, J.A. Daly, and B.D. Todd. 2019. Effects of short-term outdoor head-starting on growth and survival in the Mojave Desert Tortoise (*Gopherus agassizii*). Herpetological Conservation and Biology 14:171-184.
- Daly, J.A., K.A. Buhlmann, B.D. Todd, C.T. Moore, J.M. Peaden, and T.D. Tuberville. 2018. Comparing growth and body condition of indoor-reared, outdoor-reared, and free-ranging juvenile Mojave desert tortoises. Herpetological Conservation and Biology 13:622-633.

- Daly, J.A., K.A. Buhlmann, B.D. Todd, C.T. Moore, J.M. Peaden, and T.D. Tuberville. *In press*. Survival and movements of head-started Mojave desert tortoises. Journal of Wildlife Management.
- Nowakowski, A.J., J.M. Peaden, T.D. Tuberville, K.A. Buhlmann, and B.D. Todd. *In review*. Thermal performance curves based on field movements reveal context-dependence of thermal traits in a desert ectotherm. Submitted to: Landscape Ecology.
- Tuberville, T.D. Developing head-starting strategies for tortoises that work: an iterative process. University of South Carolina Beaufort, Bluffton, SC. 30 Nov 2018. Invited seminar.
- McGovern, P., K. Buhlmann, B. Todd, C. Moore, J.M. Peaden, and T. Tuberville. Post-release movement and survival of differentially head-started Mojave desert tortoises (*Mojave agassizii*): preliminary results. Southwest Partners in Amphibian and Reptile Conservation, Rodeo, NM. July 2019. Platform presentation.
- McGovern, P., K. Buhlmann, B. Todd, C. Moore, J.M. Peaden, and T. Tuberville. Post-release movement and survival of differentially head-started Mojave desert tortoises (*Mojave agassizii*): preliminary results. Turtle Survival Alliance, Tucson, AZ. August 2019. Platform presentation.
- Peaden, J.M., T.D. Tuberville, K.A. Buhlmann, and B.D. Todd. Modeling the impacts of roads and mitigation efforts on the viability of desert tortoise (*Gopherus agassizii*) populations. Desert Tortoise Council Annual Meeting, Tucson, AZ. February 2019. Platform presentation.
- McGovern, P., K. Buhlmann, B. Todd, C. Moore, and T.D. Tuberville. Post-release movement and survival until dormancy of hybrid head-started Mojave desert tortoises (*Gopherus agassizii*). Desert Tortoise Council Annual Meeting, Tucson, AZ. February 2019. Platform presentation.

Head-starting to augment gopher tortoise populations on protected areas in Georgia

Funding Entity

Georgia Department of Natural Resources

Start Date and Funding Amount

October 2013; \$141,158

SREL Collaborators

Dr. Tracey Tuberville and Dr. Kurt Buhlmann

Objectives

Evaluate the effectiveness of head-starting as a means of establishing viable populations of gopher tortoises on protected lands in Georgia.

Summary of Research Activities

We protected and hatched nests from three donor sites in Georgia. We have released head-starts from 2013 and 2014 cohorts as well as hatchlings from the 2015 cohort, and are monitoring them using radio-telemetry post-release. As part of phase II, we head-started and released animals from the 2015 and 2016 cohorts.

Conclusions

Head-starting appears to be an effective way of increasing juvenile survivorship, although site-specific predator pressures from raccoons and fire ants can exert strong influence on outcome.

Major Impact(s) of Research

This work is among the first to evaluate post-release site fidelity and survival of released head-started gopher tortoises. Our results will help determine whether head-starting can be used as a potential recovery tool for the species. Several agencies have now asked for technical assistance and advice regarding initiating head-starting programs for tortoises.

Other Project Personnel

Amelia Russell, M.S. Student - SREL

Louise McCallie, Research Technician - SREL

External Collaborators

John Jensen (Georgia Department of Natural Resources)

- Dr. Terry Norton (Georgia Sea Turtle Center)
- Dr. Kristen Navarra (UGA)

Dr. Nicole Stacy (University of Florida)

Products

Tuberville, T.D. Developing head-starting strategies for tortoises that work: an iterative process. University of South Carolina Beaufort, Bluffton, SC. 30 Nov 2018. Invited seminar.

- Russell, A.L., K.A. Buhlmann, and T.D. Tuberville. Refining head-starting techniques to better augment gopher tortoise (*Gopherus polyphemus*) populations. Warnell Graduate Student Symposium, UGA, Athens, GA. February 2019. Poster.
- Russell, A.L., K.A. Buhlmann, and T.D. Tuberville. Refining head-starting techniques to better augment gopher tortoise (*Gopherus polyphemus*) populations. Southeastern Partners in Amphibian and Reptile Conservation, Black Mountain, NC. February 2019. Poster.

Can waifs be used to restore viability of gopher tortoise populations?

Funding Entity

South Carolina Department of Natural Resources / US Fish and Wildlife Service / Riverbanks Zoo Conservation Support Fund / Animal Welfare Institute

Start Date and Funding Amount

May 2017; \$90,424.

SREL Collaborators

T.D. Tuberville and K.A. Buhlmann,

Objectives

Wild gopher tortoises are frequently translocated when they are displaced by development and these animals have been successfully used to augment depleted wild populations elsewhere. However, waif tortoises – formerly captive tortoises, rehabilitated, or those of unknown origin – are rarely considered for population augmentation due to heightened disease and genetic concerns. However, for peripheral populations that do not have available wild donor populations, waif tortoises may represent the only option for stabilizing populations. We have been releasing waif gopher tortoises at the Aiken Gopher Tortoise Heritage Preserve since 2006. The purpose of this project is 7yEvaluate the survivorship, site fidelity, and health metrics of translocated waif gopher tortoises used to build a viable population of state-protected land in South Carolina.

Summary of Research Activities

We completed comprehensive burrow surveys of the entire preserve, and marked and mapped them. We completed a second field season of trapping gopher tortoises to evaluate survival and site fidelity. We collected several biological samples and submitted oral and cloacal swabs for testing for a panel of common chelonian and other reptile diseases.

Conclusions

Waifs exhibited remarkably high survivorship on par with that documented for wild, unmanipulated populations as well as wild-to-wild translocated populations. In addition, only two pathogens were detected – both *Mycoplasma* species known from wild populations. We suggest that waifs can be used to build isolated viable populations of gopher tortoises when other options are not available. However, we still recommend use of waifs only in select circumstances, when released waifs are unlikely to interact with individuals from wild, stable populations.

Major Impact(s) of Research

Waif adult tortoises represent valuable animals from a species recovery perspective, but methods need to be explored that use them for species recovery, even when these individuals cannot be returned to their original populations of origin. Our research also helps address what to do with the growing waif population in captivity.

Other Project Personnel

Rebecca McKee, Masters Student – SREL Amelia Russell, Research Technician - SREL

External Collaborators

Mr. Will Dillman (SCDNR)

Mr. Barry Kesler (SCDNR)

Mr. Andrew Grosse (SCDNR)

Dr. Matt Allender (University of Illinois)

Dr. Nicole Stacy (University of Florida)

Products

McKee, R.K. 2019. An island of misfit tortoises: evaluating the use of waif animals to recover populations on the brink. M.S. Thesis, University of Georgia.

McKee, R., K. Buhlmann, W. Dillman, B. Kesler, C. Moore, and T. Tuberville. Aiken Gopher Tortoise Heritage Preserve: A project history and overview of success to date. Presentation to the South Carolina Heritage Trust Program Board, Columbia, SC. October 2018. Platform presentation.

- McKee, R., K. Buhlmann, W. Dillman, B. Kesler, N. Stacy, M. Allender, C. Moore, and T. Tuberville. An island of misfit tortoises: assessing the survival and health of translocated waif gopher tortoises. Turtle Survival Alliance, Tucson, AZ. August 2019. Platform presentation.
- McKee, R.K., K.A. Buhlmann, C. Moore, W. Dillman, B. Kesler, and T.D. Tuberville. Staying alive: waif tortoise survival and population dynamics following translocation (speed talk). Warnell Graduate Student Symposium, UGA, Athens, GA. February 2019. Platform presentation.
- McKee, R.K., K.A. Buhlmann, C. Moore, W. Dillman, B. Kesler, and T.D. Tuberville. Staying alive: waif tortoise survival and population dynamics following translocation (speed talk). Southeastern Partners in Amphibian and Reptile Conservation, Black Mountain, NC. February 2019. Platform presentation.
- McKee, R., K.A. Buhlmann, W. Dillman, B. Kesler, C.T. Moore, and T.D. Tuberville. An island of misfit tortoises: estimating survival of translocated waif tortoises. Gopher Tortoise Council, Archbold Biological Station, FL. October 2018. Platform presentation.
- McKee, R., K.A. Buhlmann, W. Dillman, B. Kesler, C.T. Moore, N. Stacy, and T.D. Tuberville. An island of misfit tortoises: health and survival of waif gopher tortoises following translocation. The Wildlife Society Meeting, Cleveland, OH. October 2018. Poster.

External (non-SRS) Funding Received in FY19

Conservation and management of gopher frogs

<u>in South Carolina</u> <u>Funding Entity</u> Longleaf Alliance via National Fish and Wildlife Federation <u>Start Date and Funding Amount</u> August, 2018; \$20,500. <u>SREL Investigators and Roles</u> S. Lance (PI) <u>External Collaborators and Affiliations</u> Lisa Lord, Longleaf Alliance

Multi-year mating dynamics and population structure in a coastal population of Alligator mississippiensis at the Tom Yawkey Wildlife <u>Center</u> <u>Funding Entity</u> South Carolina Department of Natural Resources and the Yawkey Foundation <u>Start Date and Funding Amount</u> April, 2017-19, \$74,020 <u>SREL Investigators and Roles</u> S. Lance (Co-PI) B. Parrott (Co-PI) J. Zajdel (Graduate Student) <u>External Collaborators and Affiliations</u> Dr. Thomas Rainwater, Clemson University

<u>Use of fecal genotyping and spatial capture-</u> <u>recapture modeling to investigate coyote</u> <u>abundance in South Carolina</u> Funding Entity

South Carolina Department of Natural Resources and Warnell School of Forestry and Natural Resources.

Start Date and Funding Amount July, 2019; \$165,760 (\$73,600 to SREL). **SREL Investigators and Roles**

S. Lance (Co-PI)

External Collaborators and Affiliations Dr. G. D'Angelo, Warnell School of Forestry and Natural Resources, UGA. Dr. J. Kilgo, United States Forest Service, Savannah River Site. Dr. K. Miller, Warnell School of Forestry and Natural Resources, UGA. Jordan Youngman, Warnell School of Forestry and Natural Resources, UGA **<u>Restoring and enhancing longleaf pine and</u>** <u>associated wetland habitat for at-risk species</u> <u>Funding Entity</u> Longleaf Alliance via USFWS Coastal Program <u>Start Date and Funding Amount</u> September, 2019; \$19,500. <u>SREL Investigators and Roles</u> S. Lance (PI) <u>External Collaborators and Affiliations</u> Lisa Lord, Longleaf Alliance

Molecular diet analysis of covote scat through *implementation of DNA metabarcoding* South Carolina Department of Natural Resources **Start Date and Funding Amount** July, 2019; \$80,059 **SREL Investigators and Roles** S. Lance (Co-PI) External Collaborators and Affiliations Dr. G. D'Angelo, Warnell School of Forestry and Natural Resources, UGA. Dr. J. Kilgo, United States Forest Service, Savannah River Site. Dr. K. Miller, Warnell School of Forestry and Natural Resources, UGA. Jordan Youngman, Warnell School of Forestry and Natural Resources, UGA

Total water use and source partitioning in woody bioenergy crops determined by coupled mass flux and stable isotope signatures. Funding Entity USDA National Institute of Food and Agriculture Start Date and Funding Amount September, 2015; \$915,593 SREL Investigators and Roles D. Aubrey (PI) External Collaborators and Affiliations P. Caldwell, USDA Forest Service R. Jackson, University of Georgia J. McDonnell, University of Saskatchewan C. Miniat, USDA Forest Service.

<u>Differences in the physiological response of</u> <u>Liquidambar styraciflua caused by season of</u> burn.

Funding Entity USDA Forest Service-Southern Research Station Start Date and Funding Amount August, 2015; \$60,000 SREL Investigators and Roles D. Aubrey (PI) External Collaborators and Affiliations J.J. O'Brien, USDA Forest Service Southern Research Station.

<u>Canopy complexity, physiological function, and</u> <u>ecosystem resilience of longleaf pine.</u> <u>Funding Entity</u> Tall Timbers Research Inc. <u>Start Date and Funding Amount</u> August, 2018; \$23,188 <u>SREL Investigators and Roles</u> D. Aubrey (PI) <u>External Collaborators and Affiliations</u> None.

Comparative hydrologic budgets and water use efficiencies of developing bioenergy plantations. Funding Entity USDA National Institute of Food and Agriculture Start Date and Funding Amount September, 2019; \$498,663 SREL Investigators and Roles D. Aubrey (PI) External Collaborators and Affiliations R. Jackson, University of Georgia.

Short rotation woody crop biomass and <u>nitrogen.</u> <u>Funding Entity</u> USDA Forest Service, Southern Research Station <u>Start Date and Funding Amount</u> July, 2018; \$100,000 <u>SREL Investigators and Roles</u> D. Aubrey (PI) <u>External Collaborators and Affiliations</u> None. Managing forests for ecological services and environmental sustainability. Funding Entity USDA Forest Service Start Date and Funding Amount May, 2019; \$479,480 SREL Investigators and Roles D. Aubrey (PI) External Collaborators and Affiliations None.

Kings Bay Rare, Threatened and Endangered Wildlife Surveys: Wood Storks and Wading Birds Funding Entity DoD Navy/LG2 Environmental Solutions Start Date and Funding Amount September, 2018; \$30,159 SREL Collaborators and Roles Lar1y Bryan (PI) External Collaborators and Affiliations None

Wood Stork (Mycteria americana) Species Status Assessment (SSA) Funding Entity U.S. Fish and Wildlife Service Start Date and Funding Amount June, 2019; \$20,000 SREL Collaborators and Roles A.L. Bryan (PI) and O.E. Rhodes (Co-PI) External Collaborators and Affiliations None

Evaluation of contact structure and disease dynamics in free-ranging wild pigs Funding Entity USDA-APHIS – Wildlife Services – National Wildlife Research Center **Start Date and Funding Amount** February, 2017; \$58,883.00 **SREL Collaborators** Dr. James C. Beasley (PI), Sarah Chinn (student), Dr. Peter Schlichting (postdoc) **External Collaborators and Affiliations** Dr. Toni Piaggio, USDA – Wildlife Services Dr. Kim Pepin, USDA – Wildlife Services Dr. Amy Davis, USDA – Wildlife Services Dr. Samantha Wisely, University of Florida Dr. Raoul Boughton, University of Florida

Dr. Kurt VerCauteren, USDA – Wildlife Services

Evaluation of crop damage by wild pigs Funding Entity

USDA-APHIS – Wildlife Services – National Wildlife Research Center <u>Start Date and Funding Amount</u> February, 2017; \$58,883.00 <u>SREL Collaborators</u> Dr. James C. Beasley (PI), Chris Boyce (student) <u>External Collaborators and Affiliations</u> Dr. Kurt VerCauteren, USDA – Wildlife

Services Steve Smith, USDA – Wildlife Services

Distribution and relative abundance of wildlife in Fukushima along a gradient of contamination and human land-use intensity,

<u>and spatial ecology of invasive civets</u> Funding Entity

University of Georgia, Office of International Partnerships, Office of the Vice President for Research

Start Date and Funding Amount June, 2016; \$15,350.00

SREL Collaborators

Dr. James C. Beasley (PI), Phillip Lyons (student), Hannah Gerke (student)

External Collaborators and Affiliations

Dr. Thomas Hinton, Fukushima University Dr. Kei Okuda, Fukushima University

¹³⁷Cs Activity Levels, Movement Behavior, and Efficiency of Vertebrate Scavengers in the Fukushima Exclusion Zone

Funding Entity

University of Georgia, Office of International Education

Start Date and Funding Amount

May, 2018; \$5,000

SREL Collaborators

Dr. James C. Beasley (PI), Hannah Gerke (student)

External Collaborators and Affiliations

Dr. Thomas Hinton, Fukushima University Dr. Kei Okuda, Fukushima University

<u>Effects of Chronic Radiation Exposure on the</u> <u>Health of Wild Boar in Fukushima Prefecture,</u> Japan

Funding Entity

Fukushima University, Institute of Environmental Radioactivity; University of Georgia, Office of International Education; University of Georgia, Graduate School; Environmental Radioactivity Research Network Center

Start Date and Funding Amount

May, 2018; \$17,936

SREL Collaborators

University

Dr. James C. Beasley (PI), Sarah Chinn (student)

External Collaborators and Affiliations

Dr. Thomas Hinton, Fukushima University Dr. Kei Okuda, Fukushima University Ms. Aryn Bordman, Colorado State University Dr. Thomas Johnson, Colorado State University Dr. Sami Pederson, Colorado State University Ms. Maggie Roberts, Colorado State University Dr. Rao Veeramachaneni, Colorado State University Dr. Jodi Flaws, University of Illinois Dr. Yui Nemoto, Fukushima Prefecture Dr. Diana Lafferty, Northern Michigan

<u>To initiate data collection from ongoing and additional feral swine field studies</u>

<u>Funding Entity</u> USDA-APHIS – Wildlife Services – National

Wildlife Research Center Start Date and Funding Amount

September, 2018; \$338,960.00 SREL Collaborators

Dr. James C. Beasley (PI), Lindsay Clontz (student), Allison Rakowski (Research Professional)

External Collaborators and Affiliations

Dr. Kim Pepin, USDA – Wildlife Services Dr. Nathan Snow, USDA – Wildlife Services Dr. Kurt VerCauteren, USDA – Wildlife Services

Wild pig management in Sumter and Francis

<u>Marion National Forest</u> <u>Funding Entity</u> USDA-USFS <u>Start Date and Funding Amount</u> May, 2018; \$30,259.00 <u>SREL Collaborators</u> Dr. James C. Beasley (PI) <u>External Collaborators and Affiliations</u> Mr. Donnie Ray, USDA – USFS

<u>Can waifs be used to restore viability of gopher</u> tortoise populations?

Funding Entity South Carolina Department of Natural Resources / USFWS / Riverbanks Zoo / Animal Welfare Institute Start Date and Funding Amount

May, 2017; \$90,424

SREL Investigators and Roles Dr. Tracey Tuberville (PI) Dr. Kurt Buhlmann (co-PI) Rebecca McKee (co-PI) External Collaborators and Affiliations Will Dillman, SCDNR Barry Kesler, SCDNR Andrew Grosse, SCDNR

Desert tortoise head-starting as a mitigation

strategy
Funding Entity
California Energy Commission
Start Date and Funding Amount
September, 2017; \$493,089
SREL Investigators and Roles
Dr. Tracey Tuberville (PI)
Dr. Kurt Buhlmann (co-PI)
External Collaborators and Affiliations
Dr. Brian Todd, University of California-Davis
Pearson McGovern, UGA
Dr. Mark Peaden, University of California-Davis
Carmen Candal, UGA
Collin Richter, University of California-Davis

Evaluation of head-starting as a recovery tool

for the Mojave desert tortoise

<u>Funding Entity</u> National Park Service

Start Date and Funding Amount June, 2013; \$450,000

SREL Investigators and Roles

Dr. Tracey Tuberville (PI), Dr. Kurt Buhlmann (co-PI)

External Collaborators and Affiliations Dr. Brian Todd, University of California-Davis

Head-starting to augment gopher tortoise

populations on protected areas in Georgia Funding Entity Georgia Department of Natural Resources Start Date and Funding Amount October 2013; \$141,158 SREL Investigators and Roles Dr. Tracey Tuberville (PI), Dr. Kurt Buhlmann (co-PI) External Collaborators and Affiliations

Dr. Terry Norton, Georgia Sea Turtle Center John Jensen, GADNR

Technical Expertise Requests in FY19

SREL Investigator

R. A. Kennamer <u>Date of Request</u> October 2018 <u>Requesting Entity</u> Augusta Regional Airport at Bush Field <u>Nature of Request</u> Wildlife hazard consultant for Augusta Regional Airport at Bush Field. Member of Augusta Regional Airport Wildlife Hazard Management Group.

SREL Investigator

Dr. S. Lance <u>Date of Request</u> January 2019 <u>Requesting Entity</u> Faculty member, University of Southern Mississippi. <u>Nature of Request</u> Develop genetic markers (microsatellites) for two species.

SREL Investigator

Dr. S. Lance <u>Date of Request</u> April 2019 <u>Requesting Entity</u> Faculty member, Bridgewater State University. <u>Nature of Request</u> Develop genetic markers (microsatellites) for one species.

SREL Investigator Dr. S. Lance Date of Request June 2019 Requesting Entity Faculty member, Virginia Tech University. Nature of Request Develop genetic markers (microsatellites) for one species.

SREL Investigator

Dr. James C. Beasley <u>Date of Request</u> February 2019 <u>Requesting Entity</u> International Atomic Energy Association <u>Nature of Request</u> Consult with Fukushima Prefecture, Japan

SREL Investigator

Dr. James C. Beasley <u>Date of Request</u> July 2019 <u>Requesting Entity</u> International Atomic Energy Association <u>Nature of Request</u> Consult with Fukushima Prefecture, Japan

SREL Investigator

Dr. James C. Beasley <u>Date of Request</u> September 2019 <u>Requesting Entity</u> Fukushima University, Japan <u>Nature of Request</u> Requested development of a workshop to provide information and presentations on research in my lab on the SRS

SREL Investigator

D.E. Scott <u>Date of Request</u> March 2019 <u>Requesting Entity</u> Georgia Cooperative Fish & Wildlife Research Unit <u>Nature of Request</u>

Gopher frog distribution and status on the SRS

SREL Investigator D.E. Scott Date of Request September 2019 Requesting Entity Susan Blas—SRNS-ACP Nature of Request Photos of Ash Plume Wetland (D-Area) for Brian Hennessey (DOE)

SREL Investigator D.E. Scott Date of Request September 2019 Requesting Entity Susan Blas—SRNS-ACP Nature of Request Distribution of metals in D-Area (Ash Plume

Wetland) sediments

<u>SREL Investigator</u> Drs. Tracey Tuberville, Kurt Buhlmann <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> SCDNR

Nature of Request

Serve as species expert on gopher tortoise population biology and reintroduction

SREL Investigator Drs. Kurt Buhlmann, Tracey Tuberville Date of Request FY2019 (ongoing) Requesting Entity USFWS Nature of Request Serve as species expert on Blanding's turtle population biology and reintroduction

SREL Investigator

Dr. Tracey Tuberville <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> Gopher Tortoise Council

<u>Nature of Request</u> Participate in working group and subsequent workshop on identifying range-wide conservation targets for gopher tortoises.

<u>SREL Investigator</u> Drs. Tracey Tuberville, Kurt Buhlmann <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> USFWS <u>Nature of Request</u> Participate in Species Status Assessment

Participate in Species Status Assessment for southern hognose snake (*Heterodon simus*)

<u>SREL Investigator</u> Drs. Tracey Tuberville, Kurt Buhlmann <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> Longleaf Amphibian and Reptile Conservation Project / UGA <u>Nature of Request</u> Participate in workshop and provide data records for conservation planning for reptile and

amphibian species associated with longleaf pine ecosystem.

SREL Investigator

Dr. Tracey Tuberville <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> Gopher Tortoise Council / USFWS

<u>Nature of Request</u>

Member of Gopher Tortoise Target Population Working Group for identifying and developing conservation targets for gopher tortoises across the species' range.

SREL Investigator

Drs. Tracey Tuberville, Kurt Buhlmann <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> Department of Defense / Eglin Air Force Base <u>Nature of Request</u> Provide guidance and technical assistance in developing gopher tortoise head-starting

developing gopher tortoise head-starting program and providing input on gopher tortoise translocation program at Eglin Air Force Base, FL.

SREL Investigator

Dr. Tracey Tuberville <u>Date of Request</u> FY2019 (ongoing) <u>Requesting Entity</u> Gopher Tortoise Council Nature of Request

Member of Gopher Tortoise Demographic Working Group. Help identify major gaps in demographic parameters and outline strategy for developing data warehouse for demographic data collected from throughout the species' range TASK 9. <u>SREL scientists will work closely with SRS personnel to assist DOE and other SRS</u> <u>contractors in making wise and informed decisions concerning land and facilities</u> <u>management. SREL will continue to publish its scientific findings in peer-reviewed scientific</u> <u>journals to aid the public and to assist DOE in making policy decisions by providing a basis</u> <u>of independent, verifiable science</u>

Please see SECTION VIII of this report for a list of SREL publications in FY19. Below we provide examples of specific activities that SREL personnel have conducted in FY19 to assist DOE and other SRS tenants with ongoing missions and to leverage federal funding provided to SREL to attract non-federal funding to conduct research activities on the SRS.

Department of Energy – EM

- SREL Director provided presentations to the Citizens Advisory Board on the Radiological Education, Monitoring and Outreach program, SREL's annual status and NERP Activities on the SRS
- SREL Director participated in meetings with various visitors from EM headquarters to provide an overview of SREL and its mission on the SRS
- SREL personnel participated in site visits with USDA personnel to evaluate potential research on wild pig control technologies on the SRS
- SREL leveraged DOE funding against UGA funding to conduct research on the development of pilot projects in proteomics/metabolomics at the SREL low dose facility to examine consequences of low dose exposures to aquatic species on the SRS
- SREL leveraged DOE funding and SRS site assets to obtain ~ 2.8 million dollars in new external funding during the FY19 fiscal year
- SREL personnel hosted multiple DOE personnel to tour SREL's analytical capabilities that might be used in support of SRS missions

Department of Energy – NNSA

- SREL personnel leveraged funding from NNSA to conduct research on biogeochemical cycling and efficiency of metal treatment of the HO2 wetland associated with the regulatory requirements of tritium production on the SRS
- SREL personnel leveraged funding from NNSA to conduct ecotoxicological research on amphibians utilizing the HO2 metal treatment wetlands to elucidate the biological effects of copper and other metals associated with tritium production on the SRS
- SREL personnel leveraged funding from NNSA to conduct studies of ecosystem health associated with the HO2 metal treatment wetlands
- SREL personnel leveraged funding from the NNSA to conduct research focused on the impacts of MOX construction on the viability of upper three runs creek
- SREL outreach personnel conducted extensive community outreach and education programs for NNSA and SRS

Savannah River Remediation

- SREL provided a support to SRR on technical aspects of saltstone weathering and radionuclide release over time
- SREL personnel provided technical support to SRR on addressing wildlife contamination issues

Savannah River Nuclear Solutions

SREL personnel leveraged funding from ACP to conduct radioecology research programs involving environmental characterization, contaminants in wildlife, rare species inventories and records, soil and water amendments, and tritium mitigation activities at the Mixed Waste Facilities on SRS

- SREL provided tours and presentations as requested to provide support to ACP during visits by regulators to the SRS
- SREL provided support to SRNS Corporate Communications by providing programs for 27 public tours to the general public or site visitors
- SREL provided 10 Wildlife Safety Talks to SRS site personnel

Savannah River National Laboratory

- SREL collaborated with Dr. Larry Lowe to provide research opportunities on the SRS in support of the SRNL's Minority Serving Institution Initiative
- SREL faculty collaborated with various SRNL scientists to accomplish a variety of research projects focused on environmental remediation and monitoring

US Forest Service

- SREL personnel worked with staff from the USFS-SR to provide funding to support a project to make historical rare species data easily accessible to SRS Forest Managers and to discuss methods to use digital technologies to collect, record, and make available rare species observations as they are encountered going forward.
- SREL personnel continue to inform USFS of habitat conditions at the Gopher Tortoise (*Gopherus polyphemus*) reintroduction site (Compartment 24).
- SREL personnel met multiple times with USFS personnel to discuss potential funding opportunities for SRS as a center for development of feral swine control methods
- SREL personnel worked with USFS personnel to plan and implement habitat management objectives for various Set-Aside areas on the SRS to facilitate environmental stewardship objectives of the site

TASK 10.SREL will provide stipend support to college undergraduates, graduate students, and visiting
faculty to conduct research on the Savannah River Site in association with ongoing
environmental research studies. The objective of the program will be to provide participants,
including minority students and Historically Black Colleges and Universities, with an
opportunity to pursue ecological research and training under the direction and supervision of
SREL scientific staff members

The objectives of the SREL Education Program are to (1) recruit and develop additional professionals to the environmental sciences and (2) enhance environmental awareness and research opportunities among undergraduate and graduate students with emphasis on conducting ecological research important to the DOE and Savannah River Site mission. We have made special efforts in the recruitment from underrepresented minority groups and our faculty members have worked with both students and faculty from Historically Black Colleges and Universities (HBCU) throughout the Southeast.

SREL has a long history of graduate and undergraduate education, training over 400 graduate and over 600 undergraduate researchers since 1967. Undergraduate students from more than 275 different colleges and universities have coauthored more than 170 peer reviewed research publications and more than 200 of these students have gone on to pursue careers in science. SREL offers students state-of-the-science laboratory facilities, a wide variety of natural and impacted habitats for field research, a diversity of faculty expertise, and more than sixty years of experience in ecological research. Since 1967, an average of six students per year have completed graduate students have won over 200 awards from regional, national, and international competitions at numerous professional societies and foundations. In recent years, SREL has undergone significant changes in administrative infrastructure and transitioned to self-supporting funding model. Throughout this transition period, SREL has maintained its commitment to student research and education as an integral component of its mission.

In FY19, SREL faculty and staff mentored and supervised over 87 graduate students (Table 10.1) from universities across the country. In many cases, this included formal involvement by serving as major advisors/co-major advisors and committee members for M.S. and Ph.D. candidates and in a few cases students received stipend support. However, support for students also included various activities in less formal relationships such as assistance at and access to field sites, use of field equipment, temporary lab space, as well as analytical and GIS resources for their studies.

During the FY19 fiscal year, an emphasis has been placed on finding creative ways to cost share graduate students by leveraging UGA funding with federal funding acquired through SREL's cooperative agreement with the Department of Energy. In addition, new emphases have been placed on enhancing participation of SREL Research Faculty in both graduate and undergraduate instruction. To these ends, SREL accomplished the following in FY19.

- SREL leveraged SRS site assets to acquire external resources to conduct UGA Maymester courses in wildlife ecology and genetics in May 2019
- SREL leveraged SRS site assets to acquire external resources to conduct a Spring Break course in Prescribed Fire during the spring of 2019
- SREL leveraged UGA funding against project specific funding from DOE and other sources to cost share over 55 graduate students all of whom have projects which will contribute to the knowledge base and needs of the SRS

- SREL leveraged DOE dollars to obtain salary support for 7 faculty members to provide instructional support to UGA departments as a means to maintain critical environmental expertise on the SRS
- SREL personnel successfully submitted a proposal to renew its National Science Foundation Grant to support a *Research Experience For Undergraduates* internship program for undergraduates in ecotoxicology for the summer of FY20. Details on the past success of this program are below:

<u>**REU: Radioecology**</u>

Funding Entity

National Science Foundation

Start Date and Funding Amount

April, 2015; \$302,000; May, 2018 (supplement) \$69,040

PI and co-PI's

2015-2017: Dr. J Vaun McArthur (PI, SREL), Dr. Tracey Tuberville (co-PI, SREL), Dr. Melissa Pilgrim (co-PI, USC-Upstate)

2018: Dr. Tracey Tuberville (PI, SREL), Dr. Stacey Lance (co-PI, SREL)

2019: Dr. Stacey Lance (PI, SREL), Dr. Tracey Tuberville (co-PI, SREL)

Objectives

For each participant, the primary research activity was an independent research project. Students worked with their mentors to develop the scope of their independent research projects and were involved in all stages of the research process—from articulating the hypotheses and project objectives, designing the study, collecting data, analyzing and synthesizing data, and communicating results to their colleagues and other on-site researchers. In addition to conducting independent research projects, students participated in recurring activities designed to provide them with the foundation needed to perform critical research activities during the REU program and in their future scientific endeavors. These include a radioecology seminar series, weekly professional development workshops and the Odum memorial lecture.

Summary of Research Activities

During the fourth summer of funding in 2018 we hosted 8 students using NSF funds and an additional student from other funds. Each student completed RAD Worker II training and job specific safety training. Each participant conducted individual independent research on a diversity of topics. Students presented their research as both platform presentations (at an SREL-hosted symposium) and poster presentations at a conference held on UGA main campus. In the summer of 2019, we attended a workshop focused on improving both the mentoring of – and professional development activities for - undergraduate researchers. We used this experience and a broadened theme (Interdisciplinary research in ecology, environmental chemistry and ecosystem health) to modify our NSF proposal substantially and resubmitted in August 2019.

Conclusions

1) The results from these studies are varied and cover a range of scientific import. Some of these studies may become stand-alone publications while others will be built upon either by other future REU students or by graduate students.

Major Impact(s) of Research

1) Twelve REU students have published peer-reviewed papers.

- 2) REU students have presented 23 platform and 37 poster presentations at scientific meetings.
- 3) Eleven REU students have pursued graduate programs in STEM.
- 4) Nineteen REU students have been employed within STEM fields.
- 5) Fifteen REU students have received awards for presentations or work in STEM.

Other Project Personnel

The following SREL personnel were mentors for these students: Dr. James Beasley, Dr. John Seaman, Dr. Tracey Tuberville, Dr. Stacey Lance, Dr. Guha Dhamarajan, Dr. O.E. Rhodes, Larry Bryan, and Dean Fletcher. Matt Hamilton assisted with program elements during the 10-week program. Margaret Wead provided administrative support throughout the year. Linda Lee provides website support.

External Collaborator

Dr. Melissa Pilgrim, USC-Upstate (2015-2017) Dr. Kristina Ramstad, USC-Aiken (2016-2018)

Products (Publications, Presentations, Technical Reports)

Listed below are the outcomes during FY19 for Radioecology REU. Undergraduate students funded by NSF are in bold italics with superscripts indicating their cohort (2015 cohort¹, 2016 cohort², 2017 cohort³). Other participating undergraduates are underlined. A list of comprehensive outcomes since program initiation can be found at: <u>http://srel-reu.uga.edu/outcomes.html</u>

Publications (Published or In Press) - ordered by cohort and last name of first student author

- Fulghum², C.M., E.R. DiBona³, J.C. Leaphart, A.M. Korotasz, J.C. Beasley, and A.L. Bryan. 2019. Factors affecting radiocesium (¹³⁷Cs) accumulation by fish in shallow waters within a legacy reactor cooling canal system. *Environmental International* 126:216-221.
- Leaphart, J.C., *K.C. Wilms*³, A.L. Bryan, and J.C. Beasley. 2019. Bioaccumulation of ¹³⁷Cs in anuran larvae utilizing a contaminated effluent canal on the U.S. Department of Energy's Savannah River Site. *Journal of Environmental Radioactivity* 203:25-29.
- Neff, E., A.L. Coleman, *R.W. Maness⁴*, *M. Tanelus³*, X. Xu, and G. Dharmajaran. 2019. Effects of methylmercury on mosquito oviposition behavior: maladaptive response to non-toxic exposure. *Science of the Total Environment* 667:248-254.

Publications (In Review)

- 1. Fletcher, D.E., A.H. Lindell, P.T. Stankus, N.D. Fletcher, *B.E. Lindell*², and J.V. McArthur. *In review*. Trace element accumulation in dragonfly nymphs and crayfish as indicators of constructed wetland effectiveness. *Environmental Pollution*.
- 2. Fletcher, **B.E.** Lindell², A.H. Lindell, P.T. Stankus, N.D. Fletcher, J.V. McArthur, J.C. Seaman. *In review*. Basins, beaver ponds, and the storage and redistribution of trace elements in an industrially impacted Coastal Plain stream. Environmental International.

Graduate Degrees earned - ordered by cohort, student last name

- 1. *Jarad Cochran¹* completed his MS degree in the Integrated Toxicology Program at UGA under the direction of Dr. John Seaman and began a PhD program at the University of Kentucky.
- 2. *Emily Dorward*¹ completed her MS degree in the Integrated Toxicology Program at UGA under the direction of Dr. John Seaman and began a PhD program at the University of Kentucky.
- 3. *Kyle Brown*² completed his MS degree in the Warnell School of Forestry and Natural Resources at UGA under the direction of Dr. Tracey Tuberville.

Graduate Programs in progress - ordered by cohort, student last name

- 1. Jarad Cochran¹ began a PhD program at the University of Kentucky
- 2. *Emily Dorward*¹ began a PhD program at the University of Kentucky
- 3. *Naya Eady¹* is enrolled in a PhD program in Cornell University's Biological and Biomedical Science Program.
- 4. Nat Fox^1 is in medical school at Case Western Reserve School of Medicine.
- 5. *Alexis Korotasz¹* is enrolled in a PhD program in the Department of Biology at Notre Dame University.

- 6. *Morgan Reed*¹ is enrolled in a MS program in the Department of Fish and Wildlife Conservation at Virginia Tech University.
- 7. *Sheldon Davis*² is enrolled in a MS program in the Department of Forestry and Environmental Conservation in at Clemson University.
- 8. *Emily Edwards*² has enrolled in medical school at Auburn University.
- 9. *Michaela Lambert*² is enrolled in a MS program in Forestry and Natural Resource Sciences at the University of Kentucky.
- 10. *Amelia Russell*² is enrolled in a MS program in Forestry and Natural Resources at University of Georgia, working with her mentor, Dr. Tracey Tuberville.
- 11. *Elizabeth DiBona*³ is enrolled in a MS program at Texas A&M-Corpus Christi.
- 12. *Melissa Lech*³ is enrolled in a MS program at Purdue University.
- 13. *Collin Tapolsky*³ is enrolled in a MS program in Mechanical Engineering at Embry-Riddle Aeronautical University.
- 14. *Ryne Maness*⁴ is enrolled in a MS program in the Biology Department at Georgia Southern University.

Student	Degree	University	SREL Faculty	Role
Seth Younger	Ph.D.	University of Georgia	Aubrey	Co-Advisor
Scott Oswald	Ph.D.	University of Georgia	Aubrey	Advisor
Elliot Lewis	M.F.R.	University of Georgia	Aubrey	Advisor
Joseph Hanson	M.S.	University of Georgia	Aubrey	Advisor
Mackenzie Dix	M.S.	University of Georgia	Aubrey	Advisor
Laura Fowler	M.S.	University of Georgia	Aubrey	Advisor
Jennifer McDaniel	Ph.D.	University of Georgia	Aubrey	Advisor
Ream Thomas	M.S.	University of Georgia	Aubrey	Advisor
Callie Oldfield	Ph.D.	University of Georgia	Aubrey	Committee
Karuna Paudel	Ph.D.	University of Georgia	Aubrey	Committee
Lauren Brockman	M.S.	University of Georgia	Aubrey	Committee
Monica Harmon	Ph.D.	University of Georgia	Aubrey	Committee
Luke Wilson	M.S.	University of Georgia	Aubrey	Committee
Emilee Poole	Ph.D.	University of Georgia	Aubrey	Committee
Chris Leaphart	Ph.D.	University of Georgia	Beasley	Advisor
Sara Chinn	Ph.D.	University of Georgia	Beasley	Advisor
Pooja Gupta	Ph.D.	University of Georgia	Beasley	Co-Advisor
Sara Webster	Ph.D.	University of Georgia	Beasley	Advisor
Eric Neff	M.S.	University of Georgia	Beasley	Co-Advisor
David Bernesconi	M.S.	University of Georgia	Beasley	Co-Advisor
Phillip Lyons	M.S.	University of Georgia	Beasley	Advisor
Chris Boyce	M.S.	University of Georgia	Beasley	Advisor
Hannah Gerke	M.S.	University of Georgia	Beasley	Advisor
Chelsea Titus	M.S.	University of Georgia	Beasley	Advisor
Lindsay Clontz	M.S.	University of Georgia	Beasley	Advisor
Cody Tisdale	M.S.	University of Georgia	Beasley	Co-Advisor
James Helton	M.S.	University of Georgia	Beasley	Committee
Wes Dixon	M.S.	University of Georgia	Beasley	Committee
John Grinder	Ph.D.	University of Georgia	Beasley	Committee

Table 10.1. SREL Graduate Student Program Participants, FY19
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Student	Degree	University	SREL Faculty	Role
Juan Sebastian Ortiz	Ph.D.	University of Georgia	Beasley	Committee
Darren Fraser	Ph.D.	University of Georgia	Beasley	Committee
Rebecca McKee	M.S.	University of Georgia	Buhlmann	Committee
Amelia Russell	M.S.	University of Georgia	Buhlmann	Committee
Carmen Candal	M.S.	University of Georgia	Buhlmann	Committee
Pearson McGovern	M.S.	University of Georgia	Buhlmann	Committee
Keysa Rosas-Rodriguez	Ph.D.	University of Georgia	Capps	Advisor
Christine Fallon	M.S.	University of Georgia	Capps	Advisor
Denzell Cross	Ph.D.	University of Georgia	Capps	Advisor
Kodiak Sauer	M.A.	University of Georgia	Capps	Committee
Rachel Gauer	Ph.D.	University of Georgia	Capps	Committee
Greg Jacobs	Ph.D.	University of Georgia	Capps	Committee
Suneel Kumar	Ph.D.	University of Georgia	Capps	Committee
Megan Hopson	M.S.	University of Georgia	Capps	Committee
Jeffery Beauvais	Ph.D.	University of Georgia	Capps	Committee
Eric Neff	M.S.	University of Georgia	Capps	Committee
Eric Neff	M.S.	University of Georgia	Dharmarajan	Co-Advisor
David Bernesconi	M.S.	University of Georgia	Dharmarajan	Co-Advisor
Pooja Gupta	Ph.D.	University of Georgia	Dharmarajan	Co-Advisor
Wesley Dixon	M.S.	University of Georgia	Dharmarajan	Co-Advisor
Julia Fredrick	Ph.D.	University of Georgia	Dharmarajan	Committee
Alec Thompson	Ph.D.	University of Georgia	Dharmarajan	Committee
James Helton	M.S.	University of Georgia	Dharmarajan	Committee
Julie Sanchez	M.S.	University of Georgia	Dharmarajan	Committee
Daniel Sullivan	Ph.D.	University of Georgia	Dharmarajan	Committee
Hannah Gerke	M.S.	University of Georgia	Gibbons	Committee
Sydney Hope	Ph.D.	Virginia Tech	Kennamer	Host
Will Thompson	Ph.D.	University of Georgia	Lance	Advisor
Cara Love	Ph.D.	University of Georgia	Lance	Advisor
Joshua Zajdel	M.S.	University of Georgia	Lance	Co-Advisor
Jordan Youngmann	Ph.D.	University of Georgia	Lance	Committee
Matthew Hale	Ph.D.	University of Georgia	Lance	Committee
Julie Ziemba	Ph.D.	University of Georgia	Lance	Committee

Student	Degree	University	SREL Faculty	Role
Elizabeth Shadle	M.S.	Virginia Tech	Lance	Committee
S. Simons	M.S.	University of Georgia	Martin	Advisor
C. Marshal	M.S.	University of Georgia	Martin	Advisor
A. Ward	M.S.	University of Georgia	Martin	Advisor
B. Kubecka	M.S.	University of Georgia	Martin	Advisor
J. Hill	M.S.	University of Georgia	Martin	Advisor
E. Prosser	M.S.	University of Georgia	Martin	Advisor
R. Gardner	M.S.	University of Georgia	Martin	Advisor
C. Tisdal	M.S.	University of Georgia	Martin	Advisor
J. Rectenwald	M.S.	University of Georgia	Martin	Advisor
M. Hazelbaker	M.S.	University of Georgia	Martin	Advisor
N. Wilhite	M.S.	University of Georgia	Martin	Advisor
J. Mohlman	M.S.	University of Georgia	Martin	Advisor
A. Colter	M.S.	University of Georgia	Martin	Advisor
T. Roberts	M.S.	University of Georgia	Martin	Advisor
T. Prebyl	Ph.D.	University of Georgia	Martin	Committee
C. Wakefield	M.S.	University of Georgia	Martin	Committee
A. Lohr	M.S.	University of Georgia	Martin	Committee
A. Burrow	Ph.D.	University of Georgia	Martin	Committee
H. Levy	M.S.	University of Georgia	Martin	Committee
M. Kunkel	Ph.D.	University of Georgia	Martin	Committee
T. Mezebish	M.S.	University of Georgia	Martin	Committee
K. McClearn	M.S.	University of Georgia	Martin	Committee
Emily Bertucci	Ph.D.	University of Georgia	Parrott	Advisor
Samantha Bock	Ph.D.	University of Georgia	Parrott	Advisor
Matthew Hale	Ph.D.	University of Georgia	Parrott	Advisor
Chris Elsey	M.S.	University of Georgia	Parrott	Advisor
Kristen Zemaitis	M.S.	University of Georgia	Parrott	Advisor
Josh Zajdel	M.S.	University of Georgia	Parrott	Co-Advisor
Kyle Brown	M.S.	University of Georgia	Parrott	Committee
Jarad Cochran	M.S.	University of Georgia	Parrott	Committee
Ashley LaVere	M.S.	University of Georgia	Parrott	Committee
Cara Love	Ph.D.	University of Georgia	Parrott	Committee

Student	Degree	University	SREL Faculty	Role	
Natalia Gelvez	Ph.D.	University of Georgia	Parrott	Committee	
James Helton	M.S.	University of Georgia	Rhodes	Advisor	
Wes Dixon	M.S.	University of Georgia	Rhodes	Co-Advisor	
David Bernesconi	M.S.	University of Georgia	Rhodes	Committee	
Pooja Gupta	Ph.D.	University of Georgia	Rhodes	Committee	
Natalia Gelvez	Ph.D.	University of Georgia	Rhodes	Committee	
Liyun Zhang	Ph.D.	University of Georgia	Seaman	Advisor	
Robert Thomas	M.S.	University of Georgia	Seaman	Advisor	
Emily Dorward	M.S.	University of Georgia	Seaman	Advisor	
Jarad Cochran	M.S.	University of Georgia	Seaman	Advisor	
Rebecca McKee	M.S.	University of Georgia	Tuberville	Advisor	
Kyle Brown	M.S.	University of Georgia	Tuberville	Advisor	
Pearson McGovern	M.S.	University of Georgia	Tuberville	Advisor	
David Lee Haskins	Ph.D.	University of Georgia	Tuberville	Advisor	
Amelia Russell	M.S.	University of Georgia	Tuberville	Advisor	
Carmen Candal	M.S.	University of Georgia	Tuberville	Advisor	
Rick Bauer	M.S.	University of Georgia	Tuberville	Committee	
Meghan Kelley	Ph.D.	Auburn University	Tuberville	Committee	
Kristen Zemaitis	M.S.	University of Georgia	Tuberville	Committee	
Chris Leaphart	Ph.D.	University of Georgia	Xu	Committee	
Meenakshi Agarwal	M.S.	Florida A&M University	Xu	Host	

TASK 11. <u>The participant will operate and maintain the SREL facilities on the SRS to efficiently and</u> <u>successfully perform the research, education and outreach programs described in this project</u> <u>description (Appendix A of the Cooperative Agreement)</u>

Facilities Maintenance

The Savannah River Ecology Laboratory (SREL), is the custodian of twelve DOE owned buildings with the largest of these being our 45,000 square foot main laboratory and office complex. We also have a 4,000 square foot radioecology laboratory located near Par Pond, four animal holding facilities, a greenhouse complex, two office buildings, and an assortment of utility buildings (maintenance shops, receiving building, and storage sheds). SREL is also currently the custodian of building 772-25B which is located in B-Area. 772-25B is an 8000 square foot building that contains 12 laboratories that are in various states of functionality and renovation.

At SREL we operate our own maintenance staff which consists of three full time technicians, three- part time temporary workers, and one full time custodial worker. This group is responsible for all ground's maintenance, custodial duties, routine infrastructure repair, and preventive maintenance duties for over 120 infrastructure assets. Our maintenance group also undertakes a limited number of fabrication projects in support of our research efforts. Our maintenance crew has expertise in the areas of welding-fabrication, vehicle repair, construction-renovation, electrical-HVAC, and equipment operation. This capability allows us to handle facility issues in an efficient and cost-effective manner.

As a partner here on the Savannah River Site, we strive to maintain our facilities in such a way that they comply with all the DOE guidelines for property use and safety standards. We have also worked to develop facilities that are not only aesthetically pleasing, enjoyable, and safe to work in, but facilities that lend themselves toward providing our researchers with the best possible environment to conduct their research. To that end, we have set aside significant parts of our overhead budget and dedicated many man-hours to the maintaining and renovation of our facilities.

In FY 19 our most significant facility improvement was the replacement of the roof on our main Laboratory-Office complex. This project was conducted by SRNS at the request of DOE, and they selected the roofing contractor and oversaw the project. This was a major effort which ultimately replaced just under 20,000 sq. ft. of roofing. The roof area of our main facility is composed of three distinct areas. The original structure, built in 1976, which had a two-layer spray on roof which covered 13,256 total sq. ft. of area, and two additions. The first addition was built in 1992 and totaled 2,500 sq. ft. of roof space, and the second was built in 1994 and had 4,100 sq. ft. of roof space. These two additions were constructed with a single layer black neoprene roof which is a totally different type of roofing compared to the original structure. All three of the roof areas were suffering from degradation and water intrusion. This degradation had led to water damage to interior SREL spaces, as well as safety concerns related to mold and water intrusion into electrical systems. The old roofing was completely removed and replaced with a new 30 year rated single layer neoprene roof which was coated with a white reflective coating to make the new roofing more energy efficient. This significant renovation should help to ensure the viability of our main complex for years to come.

The second major facility effort in FY 19 was the starting of the initial phase of the renovation of five of our laboratories. This included planning the layouts of the labs with affected researchers, selecting a contractor to supply and install the laboratory furniture, developing and approving a set of working plans, and having SREL maintenance personnel begin the necessary infrastructure modifications to prepare for the furniture and equipment install. These modifications to date included the removal of existing furniture in two of the five labs slated for renovation and the removal and replacement of vinyl flooring in these two labs. Since the overall renovation will include three new fume hoods, we took advantage of the roofing

renovation to incorporate three new roof penetrations into the new roof replacement. This pre-planning kept us from having to penetrate our new roofing system. This renovation project is currently ongoing and should be completed early during FY 20.

Over the last year we have also completed a number of other significant renovations to our facilities. Some of these significant projects include:

The repair of one of our storm damaged aviaries: During a windstorm event last year one, of our aviary pens was struck and significantly damaged by a falling tree. This is a custom-built facility, over thirty-five years old, and we had to do an extensive search to find a contractor capable of recreating the aluminum supports that constitute the frame of the structure. We were able to work with a greenhouse repair company from Ohio to create and install the aluminum supports necessary to make the repairs to this facility.

The installation of new fencing around one of our research ponds: SREL has six research ponds that have been traditionally used to hold research animals such as alligators, turtles, and various other reptiles. Only a couple of these ponds are currently usable due to deteriorated fencing. In an effort to start revitalizing this complex, we replaced the fencing around our front pond with heavy gauge, small mesh, chain link fencing. The enclosure was constructed to be able to handle variety of animals including large alligators and keep them properly contained.

Replacement of architectural plate glass windows. Our main building complex,737-A, was designed and constructed where most of its exterior walls in our office section are double paned vacuum sealed architectural glass. These panels are typically four-foot-wide by eight-foot-high and serve as the exterior walls for most of our offices. We have experienced a number of seal failures with these glass units, which leads to occluded windows that can not be cleaned, water intrusions, and air leaks. During the past year we replaced 15 of these glass panels.

Chair and meeting room furniture replacement. This year we focused on replacing many of the office and task chairs in our facility. We had not made any significant furniture purchases in 15 + years and we had a various assortment of worn and shoddy office and task chairs. We were able to replace over 90 chairs and bring consistency as well as an increased level of modern seating ergonomics to our facility. We also upgraded or main meeting room which was furnished with an assortment of furniture which ranged from forty to twenty-five years old. This effort included installing a new conference table along with new seating for twenty people.

The continued renovation of our faculty and staff offices: This year we completely renovated another three offices. This included re-carpeting, painting, furnishing, and making any other necessary repairs. These renovations marked our 36rd office renovation over the last five years. These offices marked the completed renovation of 65 percent of our office inventory.

Break room renovations: As the employee population of SREL has continued to grow over the last couple of years, it became apparent that our current breakroom layout was not appropriate for the new demand. As a result, we renovated this area and made the following improvements. We replaced all the existing cabinetry and countertop areas with new furniture that increased storage and counterspace. We added a bigger more functional sink and added additional and improved equipment (three microwaves and two new refrigerators). We also removed a wall to an adjoining area to increase the overall space for the breakroom. This renovation also included new paint and vinyl flooring.

Improvements to air handling infrastructure (HVAC): This year we replaced a 15-ton non- functioning HVAC in our B-Area Lab. This unit will condition over 60 percent of the 772-25B Lab complex and was a necessary first step as we plan to renovate several labs in this facility during FY 20.

Installation of LED lighting: SREL maintenance continued the phased replacement of the fluorescent lighting throughout our facility with new LED fixtures. To date approximately 45% of the old fixtures have been replaced with LED's and the new fixtures have dramatically improved the interior lighting through the facility. Our goal is to eventually replace all the lighting in our main facility with efficient LED lighting.

We also continued our emphasis on cleaning and proper organization this year. Our property coordinator is tasked to lead our efforts to clean our laboratories and storage facilities by disposing of any unneeded supplies and excessing any surplus equipment. We made significant progress in this area and we will continue to work diligently in the coming year to continue to improve our facilities in terms of proper organization and housecleaning. We also completed a major step in that effort this year by ordering and having ten 12 x 30-foot utility sheds installed at our main complex. Currently, every SREL faculty and most research professionals now have their own designated storage shed. This is a significant step in properly storing and caring for various field supplies and equipment and doing so in an orderly manner.

While much has been achieved this past year, we will remain institutionally committed to aggressively pursuing our goal of developing facilities that comply with DOE guidelines, as well as reflect positively on our staff and research efforts. To that end, we will continue to use our in-house maintenance staff and available funding to cost effectively maintain the DOE owned facilities that we occupy.

Environmental Health and Safety (EH&S) Program

The Savannah River Ecology Laboratory (SREL) continues to operate successfully under safety and environmental requirements and standards established by The University of Georgia, the SREL Safety Manual, and the Savannah River Site Policy Manual promulgated by the U.S., Department of Energy. These standards continue to address the hazards associated with SREL operations by permitting a focused effort on the health and safety issues most pertinent to SREL operations. SREL supports and promotes an integrated approach to SRS environmental health and safety issues as a signatory to the SRS Workplace Safety, Health and Security Policy and the SRS Environmental Management System Policy Statement.

SREL maintained a commitment of one, full-time safety support position (SREL EH&S Manager) during FY2019, with the addition of a second full-time safety support position (Environmental Safety Coordinator) in the 3rd quarter of FY2019. The SREL EH&S Manager serves as the manager of SREL's safety and environmental compliance programs. The SREL Environmental Safety Coordinator provides focused laboratory research related safety and hazardous waste management support to SREL science researchers. The SREL EH&S Manager interfaces with The University of Georgia's safety programs, The U.S. Department of Energy (Savannah River) Safety and Compliance oversight programs, and SRS Contractor Environmental Health and Safety Programs, Committees, and Professionals to implement the over safety and environmental compliance programs for SREL.

The SREL EH&S Manager functions as an interface with other SRS organizations in receiving and distributing applicable safety and environmental related Lessons Learned information. By integrating with other SRS organizations to share Lessons Learned information, SREL takes advantage of the collective experience and improvements identified by other organizations for similar work processes and controls at SREL. SREL's internal computer network was used to provide targeted safety information to specific groups in the laboratory. The SREL EH&S Manager electronically distributed **8 (eight)** targeted lessons learned and safety notices in FY2019 to specific worker groups at SREL. Additionally, in excess of 90 (**ninety**) SRS operational safety and environmental related announcements and notices were communicated to all SREL personnel.

The SREL EH&S Manager provided weekly reports of recordable personnel accidents or injuries to DOE-SR line management. SREL also provided monthly, SREL personnel work hour statistics to DOE-SR.

SREL personnel reported 0 (zero) work related, recordable injury/illness during FY2019. This represents a decreased injury/illness rate over the previous FY2018 reporting period (two recordable injuries).

The SREL EH&S Program continues to place an emphasis on safety and environmental training of SREL personnel. All new SREL personnel receive an initial, SREL-specific orientation on the topic of SREL safety and environmental programs, policies, and procedures in addition to the SRS required General Employee Training (GET). New SREL personnel also receive general SREL safety training and job specific safety training provided for by their SREL supervisor. Approximately **47 (forty-seven)** SREL personnel received this required training during FY2019. Additionally, SREL personnel received EH&S related training during FY2019 in the following functional areas as their job tasks required:

- Radiological Training Radiological Worker Training, Advanced Radiological Worker Training, Radioactive Sealed Source User Training, and Radiation Generating Device training
- Remote worker training in accordance with SRS remote worker requirements
- Georgia Right-To-Know Law (GRTK- HAZCOM equivalent) chemical specific training for UGA/SREL employees who utilize hazardous chemicals in the work place.
- Resource Conservation Recovery Act (RCRA) training for employees involved in the management, handling, or manipulation of hazardous or universal wastes.

SREL waste minimization and chemical disposal issues continue to be refined to promote sound environmental practices and support SRS environmental initiatives. Waste minimization techniques such as source reduction continue to be incorporated into experimental protocols, reducing the generation of chemicals wastes while supporting the SRS's pollution prevention efforts. SREL generated approximately **11 (eleven)** pounds of hazardous wastes in FY2019. **100 (one hundred)** percent of the hazardous wastes generated was from disposal of laboratory research process generated wastes. As part of SREL waste minimization efforts and to ensure that chemical hazards are addressed prior to purchasing chemicals, the SREL EH&S Manager reviewed and approved **159 (one hundred and fifty-nine)** separate chemical purchase orders made by SREL personnel.

SREL received no Notices of Violation in FY2019 as the result of external or internal reviews, inspections, or assessments. During FY2019, SREL's assigned DOE Facility Representative (FR) conducted periodic walk-down inspections of SREL operated SRS facilities in which minor safety issues were identified and promptly corrected. Additionally, SREL conducted assessments in the areas of chemical and radiological air emissions, community right-to-know, and the Georgia Right-to-Know law in compliance with state and federal requirements.

Savannah River Ecology Laboratory Analytical Services

The Savannah River Ecology Laboratory Analytical Services was established to assist SREL researchers with sample preparation and to provide in-house analysis of metals/metalloids and mercury from environmental samples. Fee for Service analysis by Inductively Coupled Plasma Mass Spectroscopy (ICP-MS) provides researchers with data on metals/metalloids. Total mercury analysis employing one of two Milestone DMA-80 Direct Mercury Analyzers is also available with a fee for service. Multiple research programs at SREL rely on metals and mercury analysis for a wide range of research projects through multiple funding agencies.

Six laboratories have dedicated workspace for all tasks associated with sample prep through analysis. One full-time position (SREL Analytical Services Manager) is dedicated to maintain these lab spaces and to operate and maintain the equipment. The manager maintains Standard Operating Procedures, Project Safety Appraisal Forms, and provides and documents Job Specific Training in sample preparation and equipment operations. In addition, the manager performs chemical coordinator duties: maintains all safety related

information including chemical inventories and safety data sheets. The manager coordinates equipment operation schedules and maintenance. These services allow students and other researchers to receive handson experience in sample preparation and equipment operation. In FY19, 16 people received training in order to utilize the analytical services labs.

Instrumentation, services and sample preparatory equipment include:

ICP-MS Analysis: The Perkin-Elmer NexION 300X is a fee for service ICP-MS. The Analytical Services manager maintains this instrument, performs calibrations, quality control checks, and analyzes samples for a fee. In FY19, we analyzed 3,965 samples that provided data for 12 research groups at SREL including graduate students, post docs, and principle investigators.

Mercury Analysis: Two Milestone DMA-80s measure total mercury following EPA Method 7473. These are fee for service instruments with charges per sample for analysis. The manager is responsible for maintenance, calibration, quality control checks, and data output. The manager provides training to students and researchers for operation of these instruments. In FY19, 12 researchers, including undergraduate and graduate students, technicians and principle investigators, performed total mercury analysis on 1,159 samples. The Brooks Rand MERX Methyl mercury analyzer is currently utilized by one research group. In FY19, approximately 200 samples were analyzed for methyl mercury.

Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES): The Perkin Elmer Optima 4300DV analyzes samples for trace and major elements. This instrument is available on a supply reimbursement basis with a small fee imposed for argon use. In FY19, there were no samples analyzed, however the instrument was maintained for availability if needed.

Sample Preparation for Metals and Mercury: Four laboratories house three chemical fume hoods, two laminar flow clean hoods, and two CEM microwave digestion ovens. These are used for acid digestions for metals analysis and for sample and calibration standards preparation. Four freeze dryers with a total capacity of 50 ports, 4 drying ovens, sample-grinding equipment, and an analytical balance are available for use for sample preparation. Consumables are supplied by either the researchers or are provided on a supply reimbursement basis in lieu of fees for lab use. Multiple research programs benefit from these dedicated lab spaces that allow for streamlined sample processing in a well-equipped setting while following SREL lab safety guidelines.

Personnel Trained in FY19Number of IndividualsGraduate Students3Undergraduate Students1Principle Investigators1Technicians/Research Professionals11Total16

Analytical Services FY19 Summary

	Number of	Number of	Number of
Equipment Description	Samples	Users/Research Groups	Days/Times Used
	Analyzed		
ICP-MS	3,965	Samples from 12 Groups	117 days
DMA-80 Mercury Analyzer	1,159	12 users, 10 groups	52
Methyl Mercury Analyzer	200	1 user, 1 group	n/a
ICP-OES	0	n/a _	0
CEM Microwaves (2)	n/a	6 users, 6 groups	42
Freeze Dryers (4)	n/a	19 users, 10 groups	245

Equipment Acquisition and Maintenance

Each year SREL reviews its capital equipment resources to ensure we maintain the analytical instrumentation as well as the laboratory and field equipment needed to meet the goals and objectives of our research programs. Regular review of our equipment infrastructure is important for maintaining and improving our research productivity, completing the tasks and objectives of our grants and contracts, and acquiring new equipment that employs technological advances needed to maintain the high quality of SREL's research programs. Based on input from the SREL research staff and prioritization by the Capital Equipment Committee the following equipment was approved for purchase by the SREL Director. The lists include a mix of new instruments as well as equipment upgrades and/or repairs that allowed us to best achieve our priority equipment needs within our budget constraints. The total expenditure for FY19 was \$97,190.

Description	Total Cost	Purpose	Programs Served
Minirhizotron Camera	\$17,000	In situ images of roots	Plant Ecology
Labconco Freeze Dryer	\$30,348	Metals and Radionuclide Analyses Preparation	Ecotoxicology Biogeochemistry Environmental Chemistry Wildlife Ecology
Microwave Digestion System	\$35,948	Sample Fish	Ecotoxicology Biogeochemistry Environmental Chemistry Wildlife Ecology
UV Water Purification System	\$5,894	Analytical Grade Water	Ecotoxicology Biogeochemistry Environmental Chemistry Wildlife Ecology
Laminar Flow Hood	\$8,000	Analytical Sample Prep	Ecotoxicology Biogeochemistry Environmental Chemistry Wildlife Ecology
Totals	\$97,190		

Table 11.1. SREL Equipment Purchases in FY19.

TASK 12.UGARF will be responsible for management and engineering services for the planning,
design, and construction of approved projects as may be required to repair, modify, or
upgrade existing facilities or construct new facilities, not to include line item projects,
necessary to support the UGARF scope of work, as approved by the Contracting Officer and
appropriate DOE program personnel. Funding for major repairs and new construction will be
provided by DOE

No new construction was undertaken in FY19. See Task 11 (above) for summary of facilities upgrades.

SECTION III. Cost Status Report

Provided to DOE-SR budget office monthly and final FY19 report was submitted on time.

SECTION IV. Schedule Status Report

No significant changes in the schedule of deliverables or achievement of milestones were experienced by SREL in FY19.

SECTION V. Changes in Approach or Goals

In FY19 SREL continued to implement a number of cost sharing initiatives with main campus units at UGA designed to improve accountability, facilitate the conduct of business, and focus resources and procedures within those areas deemed most critical to carrying out the mission of the laboratory. These initiatives include:

- Cost sharing of 6 tenure track faculty lines with UGA main campus units (3 housed at SREL and 3 housed at UGA)
- Addition of an Associate Director for Research at SREL funded solely off of funding leveraged from UGA
- Cost sharing graduate student stipends with UGA main campus units to leverage additional graduate students working on research issues on the SRS
- Leveraging research funding with UGA main campus faculty and with external funding agencies to increase SREL-based research activities on the SRS in mission critical areas such as radioecology and human wildlife conflict resolution
- Cost sharing support personnel salaries such as the Outreach Program and equipment costs with the Office of the Vice President for Research at UGA to increase the quality of SREL programs

In addition, the director of SREL has challenged the research scientists and staff at the laboratory to increase the proportion of total funding received by the laboratory from sources external to the SRS in an effort to both diversify funding streams for the laboratory and effectively leverage federal dollars to attract external funding to the SRS. In FY19, external funding (non-SRS or UGA dollars) totaled 41% of the laboratories externally funded budget.

SECTION VI. Actual or Anticipated Problems, Delays and Remedial Actions

Savannah River Nuclear Solutions has withdrawn their support for SREL participation in public tours on the SRS. As a result, the SREL director chose to redirect DOE-SR funding to cover the deficit and continue to provide SREL support for the SRNS program. In the latest Facilities Service Agreement with SRNS, SREL has indicated that it will continue to try to provide support for public tours as long as it (SREL) has the funding to support these activities.

SREL continues to work with SRNS to achieve a balance in Site Services that meets the needs of the laboratory as it increases in size and work scope to meet the needs of the SRS site tenants. Delays have occurred in delivery of services to SREL for a variety of activities despite the availability of funding. The inability to get these issues resolved has resulted in delays in research activities as well as unexpected costs to SREL's operating budget to prepare facilities for renovation or repairs. The SRS Interface Management Team has been helpful in resolving a number of these issues and with their help, there have been some success stories in FY19, despite these delays.

SECTION VII. Absence or Changes in Key, non-temporary Personnel or Team Arrangement.

Administrative Hired - Travis DeVault **Support Staff** Hired – Jina Scott-Phillips Hired – Cheri Summer **Tenure-track Faculty** No Change **Research Faculty** No Change **Postdoctoral Researchers** Separated – Fanny Coutelot **Research Professionals** Separated - A. Rakowski Separated – M. Hamilton Safety Hired – Rochelle Beasley **Outreach Personnel** Hired – Amanda Hurst Hired – Natalie Herrington Hired – Holly DeVault **Research Technicians** Hired – Abbey Riggs Hired – Morgan Shipiro Hired – Katherine McCallie Hired – Jeff Lott Hired – Tristan Mills Hired – Jason Obryhim Hired – Fabio Toledo Hired – Erin Peck Hired – Erin Spivey Hired – Mary Chapman Separated – J. Helton

SECTION VIII. Products or technology transfer accomplished: Publications, websites, collaborations, technologies, inventions/patents, other products

SREL faculty and staff added 51 new publications to the SREL reprint list in FY19

- 3473 Tomczyk, N. J., T. B. Parr, S. J. Wenger and K. A. Capps (2018). "The influence of land cover on the sensitivity of streams to metal pollution." Water Research 144: 55-63.
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- Daly, J. A., K. A. Buhlmann, B. D. Todd, C. T. Moore, J. M. Peaden and T. D. Tuberville (2018).
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SECTION IX. Special Accomplishments by Laboratory Personnel

- Jesse Abramns was awarded a Lilly Teaching Fellowship from UGA
- Stacey Lance was recognized as a Career Advocate For Students by the UGA Career Center
- James Beasley was recognized as an Early Career Fellow by the Ecological Society of America
- James Beasley received the Herrick Superior Teaching Award from the Warnell School of Forestry and Natural Resources at UGA
- James Beasley was awarded the Sarah H. Moss Fellowship
- Over 10 SREL graduate students won competative scholarships or received awards for presentations at regional, national or international meetings
- SREL research was highlighted in print, TV, and web-based media hundreds of times
- Numerous SREL faculty were asked to serve as peer reviewers on national funding panels for NSF, USDA, and other entities
- SREL Faculty were asked to give over 34 invited presentations to professional audiences in FY19