

# SOUTHEASTERN BIOLOGY

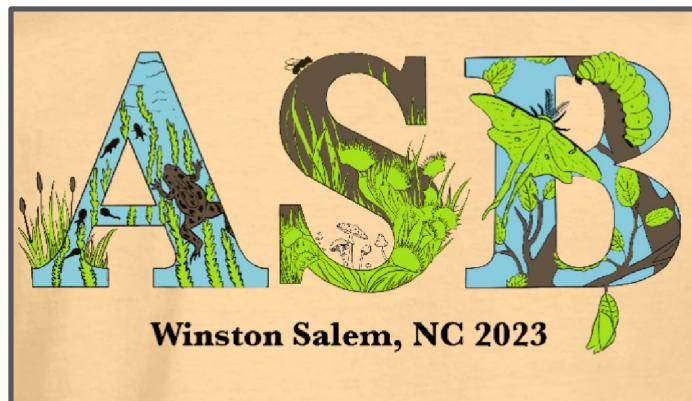


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## THE 84TH ANNUAL MEETING OF THE ASSOCIATION OF SOUTHEASTERN BIOLOGISTS

WINSTON-SALEM, NC  
23 MARCH - 26 MARCH 2023



T-shirt designed by Elijah Wells Campbell  
University, NC.

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The 2022-2023 Executive Committee.

Pictured from Left to Right: Jennifer Mandel, Matthew Heard, Chris Havran, Holly Boettger-Tong, Natasha Vanderhoff, Heather Joesting, Kimberly Hays, Lyndsay Rhodes, Chris Gissendanner, Charlie Horn, Christopher Brown. Not pictured: Hussein Mohamed, Veronica Segarra

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Goucher College  
Baltimore, MD



Heather Joesting  
Georgia Southern University  
Savannah, GA

### PURPOSE

The purpose of this association shall be to promote the advancement of biology as a science by encouraging research, the imparting of knowledge, the application of knowledge to the solution of biological problems, and the preservation of biological resources. The ASB has representation in Section G Committee of the AAAS. Varying types of membership are available to individuals and institutions. See inside back cover.

### TIME AND PLACE OF FUTURE MEETINGS

2024      Chattanooga, TN March 20-23

### The View from Here: A Message from the President

It is both an honor and a privilege to serve as the incoming President of ASB, and I am happy to report that both ASB membership and Annual Meeting attendance has rebounded, post-pandemic. Our 2023 Winston-Salem meeting attendance was a near-record 927 registrants, and the energy and vibe of our attendees was a welcome sign that we are moving forward. Kicking off the 84<sup>th</sup> Annual ASB Meeting was the R. H. Martin Plenary Speaker, Pulitzer Prize-Winning Novelist Ed Yong, who reminded us of the myriad ways that organisms on our planet sense the world and the role that anthropogenic activities play in interfering with these sensory adaptations. Work in the diversity of fields represented by ASB membership continues to reveal the mechanisms by which these and other adaptations contribute to the survival of fauna and flora of the Southeastern United States, and beyond. The world needs biologists to help navigate the host of planetary challenges we face, and the outstanding lineup of poster and oral presentations at our Annual Meeting in Winston-Salem reflected the excellent science being conducted by students and faculty across our region. These presentations offered all of us a chance to explore new ideas and exchange diverse, data-driven perspectives in a collegial, supportive environment that is beneficial for all scientists, but is especially vital for those at the beginning of their professional careers.

Our diverse membership is a strength for ASB, but we know that more can be done at our Annual Meetings and throughout the year to intentionally foster an inclusive environment for all. In Winston-Salem, the Human Diversity

Committee hosted, "Hitting the Ground Running with Your Graduate School Applications". Special thanks to Drs. Chinyere Knight, Jennifer Mandel and Veronica Segarra for making this insightful workshop a supportive pipeline for all future professionals. We also appreciate the continued collaboration of our affiliate organizations, (BBB, BSA, ESA, NABT, SABS and SHC), whose contributions are vital to the continued success of our Annual Meetings. The SABS mixer was particularly well attended in Winston-Salem and provided an outstanding opportunity for reconnecting; the fun continued in our evening Social at ROAR, with three floors of food, fun and fellowship. On Saturday evening, our Awards Banquet acknowledged a diversity of outstanding student presentations and allowed the Association to congratulate Dr. Matt Estep as the 2023 recipient of the ASB Meritorious Teaching Award, and Ms. Amy Allen from Southern Lee High School, as the recipient of the Lucrecia Herr Award for excellence as an outstanding high school biology teacher.

The Winston-Salem meeting, with a slightly different Thurs-Saturday format, would not have been possible without the dedicated work of the all-volunteer Executive Committee, and the continuing efforts of our amazing meeting planner, Shannon Oliphant-Gordon. In addition, special thanks to our Featured Institution, Catawba College, for their terrific volunteers and much-appreciated financial support. We are also extremely fortunate to have generous donors in Martin Microscope, Breedlove and Associates, Dwayne Wise, the PULSE Southeast Regional Network, and SIM BIO. I would also like to take a moment to gratefully acknowledge the outstanding leadership of our PastPresident, Dr. Chris Brown. His dedication to ASB and to providing an excellent experience for all attendees at our Annual Meetings in Arkansas and North Carolina was inspirational, and allowed our Association to both recover from the challenges of COVID and to thrive as we move forward. Prior to the 2023 meeting, he led the effort to renew our Leadership Guide, which clarifies the important roles that our committees play in the success of ASB. Please do consider volunteering for an ASB committee – it's an outstanding way to meet new colleagues and to shape the future of our organization. Finally, Thank You - for your support of ASB, for the excellence of your research, for your inclusive mentoring and for your dedication to the study of life. See you March 20-23, 2024, in Chattanooga, TN!

Holly Boettger-Tong  
ASB President

## **ASB 2023 R.H. Martin Plenary Session**

**Pulitzer-Prize Winning Journalist and Author  
Ed Yong**



Named "the most important and impactful journalist" of 2020 by Poynter, Pulitzer Prize winner Ed Yong is a science staff writer with The Atlantic. He was awarded journalism's top honor, the 2021 Pulitzer Prize for Explanatory Reporting, for his crucial coverage of the coronavirus pandemic. An accomplished speaker, Yong brings his vast scientific knowledge and engages his audiences through his insightful conversations about the pandemic, the animal kingdom, the challenges of science journalism, and more.

Ed Yong is the best-selling author of *I Contain Multitudes: The Microbes Within Us*, a groundbreaking, informative, and entertaining examination of the relationship between animals and microbes. His second book, *An Immense World*, takes a comprehensive look at the fascinating sensory worlds of animals. In addition to The Atlantic, his work has appeared in National Geographic, the New Yorker, Wired, Nature, New Scientist, and Scientific American, among others.

## About the R.H. Martin Plenary

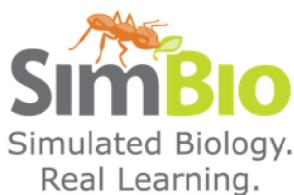
Robert H. Martin, founder and owner of Martin Microscope Company from 1946 until his death in 2006, was an avid supporter of ASB and of the sciences in general. He supported ASB for decades as a Patron member and sponsor of the ASB Student Research Award, and, with his passing, his family established what has become the Robert H. Martin Plenary Address fund to help cover the costs of bringing in highly qualified speakers for the ASB annual meeting.



## ASB Patrons and Sponsors



Southeast Regional Network



There are many ways to support ASB's mission to provide high-quality opportunities for biologists across the southeast to present their work, network with peers, and develop professionally. Find out more on our website:

<https://www.sebiologists.org/sponsors--exhibitors.html>



Breedlove, Dennis, and  
Associates, Inc.  
Winter Park, FL



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## Donors like you!

## 2023 Affiliate Organizations



*Southeast section*



*Southeast chapter*



*Districts I and II*



## 2023 Exhibitors



JACKSONVILLE STATE UNIVERSITY



College of Pharmacy & Health Sciences

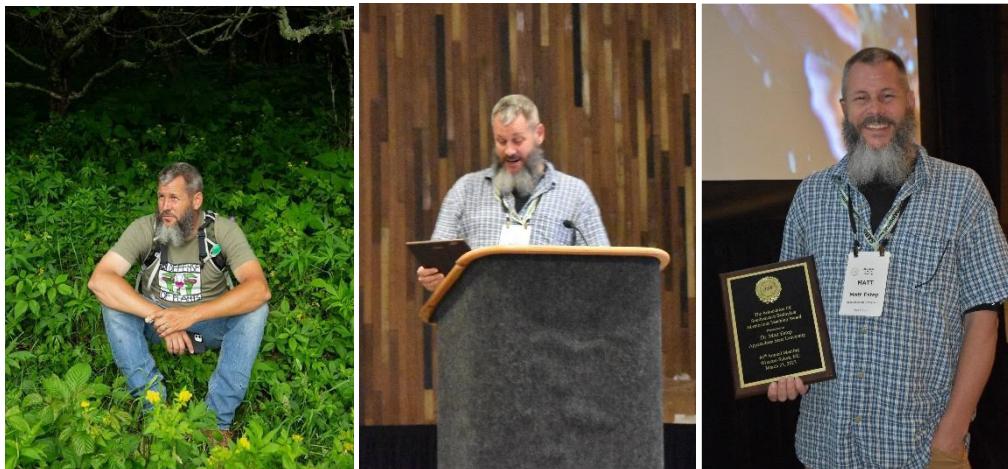


Districts I and II

## ASB PRESTIGIOUS AWARDS PRESENTED AT THE 2023 ASB MEETING IN WINSTON-SALEM

### CAREER AWARDS

**ASB MERITORIOUS TEACHING AWARD** **Dr. Matt Estep**  
Associate Professor, Director  
Appalachian State University



**ASB LUCRECIA HERR OUTSTANDING BIOLOGY TEACHER AWARD** **Amy Allen**  
Southern Lee High School  
Sanford, NC



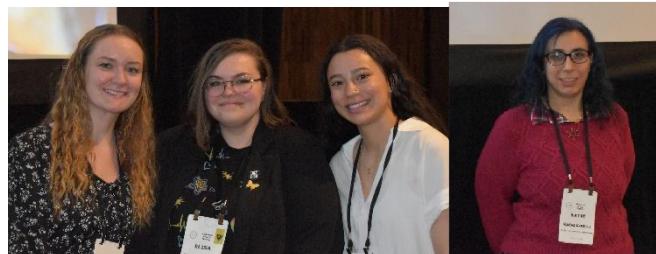
### ASB SUPPORT AWARDS

**THE LAFAYETTE FREDERICK Abigail Delepenha  
UNDERREPRESENTED MINORITIES University of Mary Washington SCHOLARSHIP**

**SUPPORT AWARDS FOR FIRST-GENERATION Barbara Comer, Georgia Southern University UNDERGRADUATE  
STUDENT MEMBERS OF Elisabeth Howansky, Lander University**

**ASB Jeilymar Jimenez, Lander University  
Georgia Spann, Lander University  
Natalie Salguero, Georgia Southern University**

**SUPPORT AWARD FOR GRADUATE STUDENT** Leigha Henson, MS, Appalachian State University  
**MEMBERS OF ASB** Sonja Barber, MS, Frostburg State University  
Maygan Palacios, MA, Georgia Southern University  
Peyton Jackson, MS, West Liberty University  
Garrett Billings, MS, University of Tennessee at Chattanooga  
Erin Gaylord, MS, University of Tennessee at Chattanooga  
Katia Coscia, PhD, Middle Tennessee State University  
Devani Jolman, PhD, Old Dominion University  
Alyssa Quan, PhD, University of Georgia  
Christina Harvey, PhD, Wake Forest University



### **ASB RESEARCH AWARDS**

**ASB STUDENT RESEARCH AWARD** Kathryn Greene, University of Kentucky  
**Sponsored by Martin Microscope** "Salamander Movement Propensity Resist Effects Of Supraseasonal Drought"



### **ASB PRESENTATION AWARDS**

**ASB STUDENT POSTER PRESENTATION AWARD** Sam Williams, Lander University  
– **ANIMAL BIOLOGY** "Shedding some light on artificial illumination's effects on mammal diel activity patterns"

**ASB STUDENT POSTER PRESENTATION AWARD** Cameron Braswell, Virginia Tech  
– **AQUATIC BIOLOGY** "The Effects of Varying Microplastic Morphologies on Crayfish-Annelid Symbiosis"

**ASB STUDENT POSTER PRESENTATION AWARD** Ruben Rosado, Florida Gulf Coast University

- **CELL AND MOLECULAR BIOLOGY** "Determining anti-cancer potential of proprietary compound against triple-negative breast cancer"

**ASB STUDENT POSTER PRESENTATION AWARD** **Shelby Blanchard & Tori Glazier**, Mercer University

- **MICROBIOLOGY** "Biofilm Inhibition in *Staphylococcus aureus* using Small Molecules as Competitive Inhibitors"

**ASB STUDENT ORAL PRESENTATION AWARD** **Aditi Patel**, Georgia Gwinnett College

- **ANIMAL BIOLOGY** "The fitness consequences of genetic variation for *Trichogramma kaykai* infected with parthenogenesisinducing *Wolbachia*"

**ASB STUDENT ORAL PRESENTATION AWARD** **Sydney Brown**, Georgia College & State University

- **AQUATIC BIOLOGY** "Uses of Diatom Community Analyses in Streams with Legacy Agricultural Uses in Middle Georgia"

**ASB STUDENT ORAL PRESENTATION AWARD** **Amber Ajamu-Johnson**, Augusta University

- **CELL AND MOLECULAR** "In vivo characterization of synthetic Notch signal BIOLOGY transduction"

**ASB STUDENT ORAL PRESENTATION AWARD** **Alexis Hoopman**, University of North Carolina at Greensboro.

- **MICROBIOLOGY** "Unintended Antibiotic Target: The (Honey Bee) Reproductive Microbiome"

**ASB DR. FLOYD SCOTT AWARD FOR THE BEST ORAL PRESENTATION AWARD IN HERPETOLOGY** **Erin Gaylord**, University of Tennessee at Chattanooga "Developing a Predictive Geospatial Habitat Model for a Rare Species of Salamander in TN: A Case Study for the Green Salamander (*Aneides aeneus*)"



## AFFILIATE AWARDS PRESENTED AT THE 2023 MEETING

**NORTH CAROLINA BOTANICAL GARDEN AWARD** **Skyler Fox**, Furman University "The importance of understanding genetic and clonal structure for species recovery: a case study using federally endangered *Sagittaria fasciculata*"

**SOUTHEAST CHAPTER OF THE ECOLOGICAL SOCIETY OF AMERICA – TRAVEL AWARDS** **Cameron Braswell**, Virginia Tech University  
**Brice Crum**, Virginia Tech University  
**Macy Gosselaar**, Mississippi State University  
**Cade Karminski**, Virginia Tech University  
**Cameron Lockett**, Virginia Tech University  
**Marykate McHale**, Frostburg State University  
**Htet Lin Naing**, Mississippi State University

**SOUTHEAST CHAPTER OF THE** **Gabriella LeFevre**, Austin Peay State University

**ECOLOGICAL SOCIETY OF AMERICA –** "Thistle come as no surprise, invasives exploit native

**EUGENE P. ODUM** pollinators: comparing the reproductive success of an invasive and native thistle in the subtribe Carduinae (Asteraceae)"

**SOUTHEAST CHAPTER OF THE  
ECOLOGICAL SOCIETY OF AMERICA –  
ELSIE QUARTERMAN-CATHERINE KEEVER  
AWARD**

**SOUTHERN APPALACHIAN BOTANICAL SOCIETY  
– RICHARD AND MINNIE WINDLER AWARD IN  
ECOLOGY**

**Amy Hessl, Andrew Liebold, and Morgan Leef.**  
Dendrochronological Reconstruction of the Historical Invasion of Balsam Woolly Adlegid (*Adelges piceae*) Feeding on Canaan Fir *Abies balsamea* ssp. *pahnerolepis* in the Central Appalachian Mountains. *Castanea* 87(1): 1

**SOUTHERN APPALACHIAN BOTANICAL SOCIETY  
– RICHARD AND MINNIE WINDLER AWARD IN  
SYSTEMATICS**

**Katherine Culatta, Alexander Krings, Lilian P. Matallana-Ramirez, and Ross Whetten.** Clarifying Taxonomic Boundaries in *Nuphar sagittifolia* (Nymphaeaceae): Insights from Morphology and Population Genetic Diversity. *Castanea* 87(1): 81

**SOUTHERN APPALACHIAN BOTANICAL SOCIETY  
– EARL CORE STUDENT RESEARCH AWARD**

**Megan Gauger**, University of Pittsburgh  
"Weather-induced Flower Closure in Spring Ephemeral Forest Wildflowers and its effect on Pollen Viability"

**Clayton Hale**, University of Georgia  
"Understanding the Impact of Climate Change on Phenological Mismatch between Spring Ephemerals and Overstory Leaf Out"

**Devani Joman**, Old Dominion University  
"Ecological Consequences of Hybridization in Highbush Blueberry across Environments in Virginia"

**SOUTHERN APPALACHIAN BOTANICAL SOCIETY  
– OUTSTANDING STUDENT ORAL  
PRESENTATION AWARD**

"Ecosystems in Transition: An Assessment of Plant Community Change in Pond Pine Pocosins in Coastal North Carolina."

**Maccoy Kerrigan**, NC State University  
**SOUTHERN APPALACHIAN BOTANICAL SOCIETY**

**– OUTSTANDING STUDENT POSTER PRESENTATION AWARD** "Preliminary data in the Species Delimitation of the Native, Southeastern Early Blue Violet (*Viola palmata* L.) species complex using morphological, ecological, and genetic

**Jonathon Osborne**, University of Southern Mississippi

**SOUTHERN APPALACHIAN BOTANICAL SOCIETY** **Dr. Alan Weakley**, UNC Herbarium, North Carolina Botanical Garden

**– ELIZABETH ANN BARTHOLOMEW AWARD**

**SOUTHEASTERN SECTION OF THE BOTANICAL SOCIETY OF AMERICA –** **Elizabeth Companion**, University of North Carolina Asheville "Reproductive output of closely-related sympatric pitcher datasets."

**OUTSTANDING STUDENT POSTER AWARD** plants (*Sarracenia* spp.) and their morphological hybrids" **IN PLANT SCIENCE**

**SOUTHEASTERN SECTION OF THE BOTANICAL SOCIETY OF AMERICA –** **Ben Brewer**, Appalachian State University "Assessing the Genetic Diversity and Structure of Gray's Lily

**OUTSTANDING STUDENT PAPER AWARD** (*Lilium grayi*)  
IN PLANT SCIENCE



**ASB  
PRESENTED  
MEETING**

**RESEARCH GRANTS  
AT THE 2023**

**ASB UNDERGRADUATE GRANT** **Sam Williams**, Lander University "Shedding some light on artificial illumination's effects on diel mammal activity"

**ASB GRADUATE GRANT**

**Gavriel Burger**, Florida Gulf Coast University "Identification of anti-metastatic stilbene compounds in a triple-negative breast cancer zebrafish xenograft model"

**ASB PROFESSIONAL GRANT**

**Lyndsay Rhodes**, Florida Gulf Coast University

"Understanding the role of stilbenes in EMT regulation of breast cancer migration"



## ASB Symposia

### SYMPOSIUM: Conservation, Natural History, and Systematics of Crayfishes - a Southeastern perspective A

Zachary Loughman, Department of Organismal Biology, Ecology, and Zoo Science, West Liberty University, West Liberty, WV and Zackary Graham, Department of Organismal Biology, Ecology, and Zoo Science, West Liberty University, West Liberty, WV

#### Symposium Introduction/Welcome

141 - Natural history and ecology of the slender crayfish (*Faxonius compressus*): an ecosystem engineer in the Western Highland Rim, USA

Zackary Graham, Zachary Loughman

*Department of Organismal Biology, Ecology, and Zoo Science, West Liberty University, West Liberty, WV*

Ecosystem engineers influence the physical environment which changes the distribution and availability of resources to other organisms. Based on their burrowing abilities, freshwater crayfish have been widely recognized as ecosystem engineers. Crayfishes construct burrows that range from simple, rudimentary depressions in aquatic systems, to complex, labyrinth-like terrestrial burrow systems that may be significant distances from a permanent body of water. Here, we investigate the ecosystem engineering and burrowing ability of the slender crayfish, *Faxonius compressus* which lives in aquatic environments characterized by an abundance of small cobble- and gravel-sized substrates. Without larger substrates to burrow under, we found that *F. compressus* construct extremely high densities of burrows across riffle, run, and pool macrohabitats. Based on the average number of burrows that we counted, a 5 x 100 m pool is estimated to contain an average of 25,860 burrows. We also conducted behavioral observations of this species to glean information on their natural history. *Faxonius compressus* regularly inhabits and competes for ownership of burrows which serve as a shelter from predators. Importantly, not only are these burrows used by *F. compressus*, but we documented other crayfishes and fish species utilizing these burrows. In total, we documented 2 other crayfishes and 6 fish species utilizing *F. compressus* burrows. We discuss the ecosystem engineering abilities of this species in relation to other crayfishes and suggest future avenues of research to elucidate this species' natural and life history.

142 - Crayfish invaders and their relationship with native symbionts

Cameron Lockett<sup>1</sup>, Cameron Braswell<sup>2</sup>, Robert Creed<sup>3</sup>, Bryan Brown<sup>1</sup>

<sup>1</sup>*Virginia Tech, Blacksburg, VA*, <sup>2</sup>*Virginia Tech, BLACKSBURG, VA*, <sup>3</sup>*Appalachian State University, Boone, NC*

Symbiosis is a component of the life history of most multicellular organisms. Symbiosis can affect growth rates, alter host mortality, and even affect immune-suppressant properties. However, the relationship between hosts and their symbionts is not a given. Multiple factors can disrupt the interactions between hosts and symbionts. The potentially disruptive effects of invasive species on native host-symbiont relationships is an emerging global change threat. Invasive species compete with natives for essential resources such as food and habitat. Another resource invaders may affect are endemic symbionts. This effect can be seen with the introduction of invasive crayfish to the Virginia Mountain Lake Region where invaders are disrupting the native hosts' relationships with symbionts, and even forming relationships of their own. In our study, we examined the prolonged effect of symbionts on invasive crayfish and identified the range of relationships that resulted from the pairings with 2 invasive hosts. In an aquarium experiment, we placed native symbionts (branchiobdellid worms) on the carapace of 2 invasive species, *Faxonius cristavarius* and *Faxonius viridis*. To limit the ability of invasive hosts to control symbiont abundance,  $\frac{1}{2}$  of each host species were subjected to dactyl ablation on the walking legs and compared to an unmanipulated control. We predicted that the invasive crayfish would serve as a suitable host for the native symbionts. The results of our study supported our predictions to varying degrees with *F. viridis* being more suitable as a host compared to *F. cristavarius*. In addition, our study showed that *F. cristavarius* had a higher degree of variation of percent mass growth between the experimental and control group. Suggesting *F. cristavarius* can receive symbiotic benefits under abnormal conditions.

#### 143 - Life History of Pristine Crayfish, *Cambarus pristinus*

Kendell Hamm

*Tennessee Technological University, Cookeville, TN*

The southeastern United States represents a global hotspot for freshwater biodiversity. Approximately one-third of the world's crayfish species are found in the Southeast with >80 species known from Tennessee. The pristine crayfish (*Cambarus pristinus*) is a rare species endemic to the Cumberland Plateau in Tennessee that is under consideration for federal listing under the U.S. Endangered Species Act of 1973. To inform conservation and potential recovery efforts, we conducted a 12-month life history study for two populations of pristine crayfish from November 2021 to October 2022. Two morphologically and genetically distinct forms of the species are known. The study populations were located in Pokepatch Creek, representing the Caney Fork form, and Camp Creek, representing the Sequatchie form. Pristine crayfish appear to become reproductively mature at/near one year of life upon reaching 18–19 mm TCL. Adult pristine crayfish have an average size of 23–24 mm TCL. Recently molted pristine crayfish were collected throughout the year; however, collection of recently molted individuals peaked during May at Pokepatch Creek and August at Camp Creek. A summer mass molting event where 100% of adult males molted to nonreproductive form occurred from May–July. Mating occurs throughout the fall and winter, females are ovigerous in the spring, and young of year recruitment occurs during the summer. Interestingly, Camp Creek trailed Pokepatch Creek by approximately one month for many aspects of life history events. However, this is likely due to prolonged colder water temperatures (°C) at Camp Creek. Both populations appear to have two age classes present throughout most of the year with the exception of summer months when young of year recruitment occurs. Pristine crayfish appear to have a longevity of 2–2.5 years with some individuals occasionally living up to 3 years.

#### 144 - Effect of an introduced crayfish on the native symbionts inhabiting native crayfish

Mary Massie<sup>1</sup>, Bryan Brown<sup>2</sup>, Robert Creed<sup>1</sup>

<sup>1</sup>*Appalachian State University, Boone, NC*, <sup>2</sup>*Virginia Tech, Blacksburg, VA*

Biological invasions are a major threat to native species. Invaders can cause reductions in native species via interactions such as competition, predation, herbivory and spread of disease. However, invading host species may also affect the abundance and diversity of symbionts living on native hosts. *Faxonius cristavarius*, an invasive crayfish, is now established in the North and South Forks of the New River in northwestern, North Carolina and its range now overlaps with those of three native crayfish in the genus *Cambarus*. *Cambarus* crayfish in the New River may host up to seven species of annelid worm known as branchiobdellidans. *Faxonius cristavarius* is a noncompetent host of branchiobdellidans and removes them quickly when they colonize. We conducted a crayfish survey in the New River to determine the diversity and abundance of branchiobdellidans on *Cambarus* spp. across a gradient of *F. cristavarius* relative abundance. There were significant, negative relationships between the relative abundance of *F. cristavarius* and worm numbers and richness. Significant reductions were also observed in several worm taxa. In a lab experiment,

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<sup>1</sup>*Virginia Tech, Blacksburg, VA*, <sup>2</sup>*Appalachian State University, Boone, NC*

As increased pressure from invasive species and anthropogenic pollutants continues to build, a greater understanding of native species' life histories is imperative to the preservation of healthy populations. Being a keystone species and ecosystem engineer, the survival of native crayfish populations acts as an important indicator of the overall health and stability of freshwater stream ecosystems. With the species of crayfish *Cambarus appalachensis* being described as recently as 2017, little research has been done in terms of their juvenile development and behavior. Our research sheds light on this issue by examining the change in mass, carapace length and behavior over the first six months of their development. From egg to new-of-the-year, the growth of these juveniles were photographed on a weekly basis, with important benchmarks in these animals' development such as pigmentation, burrowing, and aggressive behavior being observed to occur synchronously across 5 broods (families of hatchlings). In order to execute this experiment, 7 gravid females were collected from Sinking Creek, Virginia; of which, 5 produced viable broods of young. These broods were kept with their mother for the approximate month of gestation, and were then removed before being placed in an observation tank. Brood size ranged from 10 to 130 individuals, and survival rates ranged from 16% to 62% over the course of 6 months. Anthropogenic pollutants like microplastics, combined with the increased competition from invasive species present serious risks to young crayfish. This is why it is crucial to understand *C. appalachensis*' early life history and developmental strategies to protect native crayfish populations in the future.

we found that the presence of *F. cristavarius* in aquaria significantly reduced worm number on *Cambarus robustus* compared to a *C. robustus* occupying aquaria with a conspecific. The decline in worm abundance and diversity appears to be a hidden, effect of invasions and it is likely that many other native symbionts are also declining as a result of invasions.

145 - Exploring limitations of single methodology approaches to species delimitation and phylogenetics of North American crayfishes

Brooke Grubb<sup>1</sup>, Jeffrey W. Simmons<sup>2</sup>, Hayden Mattingly<sup>1</sup>, Carla Hurt<sup>3</sup>

<sup>1</sup>Tennessee Technological University, School of Environmental Studies, Cookeville, TN, <sup>2</sup>Tennessee Valley Authority, Fisheries and Aquatic Monitoring, Chattanooga, TN, <sup>3</sup>Tennessee Technological University, Department of Biology, Cookeville, TN

Crayfish taxonomy has historically been based on a few morphological traits. This is problematic as crayfish harbor morphological plasticity and cryptic forms. The increasing popularity of phylogenetic studies using mitochondrial data have aimed to circumvent the problem of using morphology to delineate species evolutionary relationships but they come with their own limitations: introgression, unresolved relationships, small sample size, expense of sampling and processing, etc. Each approach on their own lacks robustness needed to accurately delineate species boundaries and provide taxonomic validity for conservation needs. We explore the limit of mitochondrial phylogenetic analyses within the *Cambarus striatus* species complex when attempting to place a previously unknown taxon and the unresolved phylogenetic relationship between *Faxonius wrighti* and *F. yanahlinus* delineated by morphology. We incorporated a multi-marker genetic approach using single-nucleotide polymorphisms (SNPs) to provide a more robust framework for species delineation when used in conjunction with mitochondrial analyses. Species boundaries were explored using the generalized mixed Yule-coalescent (GMYC) method and Bayesian Phylogenetics and Phylogeography (BPP) method. We will discuss our preliminary results in the context of future species descriptions and the limitations of our current framework.

146 - Documenting Early Life History Strategies of the Conhaway Crayfish

Cade Karminski<sup>1</sup>, Cameron Braswell<sup>1</sup>, Bryan Brown<sup>1</sup>, Austin Gray<sup>1</sup>, Robert Creed<sup>2</sup>

147 - Ecology of the Lean Crayfish, *Cambarus strigosus* (Decapoda: Cambaridae)

Megan B. Stubbs<sup>1</sup>, Hogan D. Wells<sup>1</sup>, Zanethia C. Barnett<sup>2</sup>, Zackary A. Graham<sup>1</sup>, Zachary J. Loughman<sup>1</sup>

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The Lean Crayfish, *Cambarus strigosus*, was described by Hobbs (1981) as a narrow Georgia endemic burrowing crayfish found only in three counties within the Broad River Basin. Only two historical records from 1983 place *C. strigosus* outside of Georgia within the neighboring state of South Carolina. However, in 2019, populations of *C. strigosus* were discovered in Sumter National Forest, South Carolina, near the Georgia-South Carolina border. Little is known about *C. strigosus* aside from its original description. To elucidate the biology of this data-deficient crayfish, we performed a one-year study on the Sumter National Forest populations of *C. strigosus*. We split the populations into two open-field and two forested sites and completed five sampling events from February 2022-October 2022. Objectives included to (1) measure and compare the distribution and density of burrow surface activity using a 10x10 m grid, as well as T-square sampling method (2) model ecological correlates using maximum entropy (MaxEnt) modelling; and (3) gather natural history data. Analysis of burrow surface activity included measuring indices of aggregation using Spatial Analysis by Distance Indices (SADIE) software. MaxEnt modeling determined land-use type as the most informative ecological variable in the habitat suitability model. Basic natural history data, including morphometrics and commensal species, is also discussed. Ultimately, this study closes the knowledge gap on a primary burrowing crayfish and provides the first example of using SADIE on freshwater crayfish. Further studies may focus on life history or aim to ground truth the habitat suitability model to determine the full range of this previously understood Georgia endemic.

148 - Habitat associations and movement biology of the Big Sandy Crayfish (*Cambarus callainus*), assessed via radiotelemetry

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High resolution data involving the specific habitat requirements of crayfishes are lacking. This is recognized as a significant limiting factor in crayfish conservation and management. While significant efforts evaluating the life history of *Cambarus callainus* have been completed, a more detailed analysis of microhabitat utilization, habitat preference, and movement patterns remains incomplete. A study was completed in the Tug Fork River in Big Sandy, WV, where we assessed potential micro- and macro-habitat preferences and individual movement patterns of adult *C. callainus* individuals via radiotelemetry. A total of 29 animals were collected via kick-seine method, fitted with Holohil BD-2 transmitters, and released at their initial capture point across three phases (Phase 1: May-June 2022; Phase 2: June-July 2022; Phase 3: November 2022). Phase 1 animals were initially captured from an area of high anthropogenic activity, while Phase 2 and 3 animals were captured from a less disturbed area, farther upstream. Evidence of homing behavior throughout our study duration was limited, with most individuals maintaining a steady journey upstream, frequenting the same rock for just a short period before continuing additional upstream movement. Notably, one female traveled a total cumulative distance of 3.9 kilometers upstream from its initial capture location over a span of 34 days, averaging over 100 meters per day. Our study supports

previous findings of *C. callainus* preferring large, unembedded slab boulders that lack excessive sedimentation. The observed large distance travelled by individuals has direct implications for conservation and management efforts for *C. callainus* and exemplifies the importance of establishing baseline natural history data for crayfish from direct observation.

## LUNCH

### 149 - Tagging Method for Future Propagation and Release of the Imperiled Big Sandy Crayfish (*Cambarus callainus*) with a Surrogate Species, Conhaway Crayfish (*Cambarus appalachiensis*).

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Nearly one-third of all 669 crayfish species worldwide are threatened with extinction, 400 of which are native to the United States. The Big Sandy Crayfish (*Cambarus callainus*), native to the Big Sandy River basin within West Virginia, Kentucky, and Virginia, was listed as federally threatened in 2016 due to coal mining impacting their habitat. Propagation can be used to reintroduce and sustain declining populations of *C. callainus* within their native range. Captive care and release of captive-born young have been documented to be successful at both White Sulphur Springs National Hatchery and West Liberty University. Visible Implant Elastomer (VIE) is used frequently in small animals to identify individuals or groups of animals. Tagging stocked *C. callainus* with VIE can be used to identify individuals released in streams, record success rates, and track genetic lineage. Preventing inbreeding and outbreeding depressions in captive-raised animals is essential to ensure the native stock is genetically fit. Eightytwo Conhaway Crayfish (*Cambarus appalachiensis*) were used as surrogate species to monitor the effects of VIE tags. We found no significant mortality in tagged individuals with significant tag retention in all sizes and developed a protocol for future efforts with *C. callainus*. Thus, VIE may be a safe and inexpensive way to track released *C. callainus* alongside planned propagation and release efforts.

### 150 - Seasonal Microhabitat Use by Brawleys Fork Crayfish, *Cambarus williami*, a Tennessee State Protected Species

Tanya Khan, Hayden Mattingly

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*Cambarus williami* (Brawleys Fork Crayfish) is endemic to the Stones River and Collins river watersheds in central Tennessee. This species is listed as a state endangered (S1) species and is under review for federal protection under the Endangered Species Act. Determination of federal status will be facilitated by knowledge of ecological associations of *C. williami* across its distributional range. Our aim was to evaluate predictors of seasonal habitat selection and crayfish species assemblages associated with *C. williami*. Nine 200-m reaches in three streams (East Fork Stones River, Hollis Creek, and Mountain Creek) were sampled in the Spring (March 2021) and Fall (September-October 2021) seasons. Microhabitat conditions were measured, and crayfishes collected from six randomized 0.5-m<sup>2</sup> quadrat samples per 200-m reach in the Spring and Fall seasons. *Cambarus williami* was most abundant in riffle mesohabitats, moderately abundant in run mesohabitats, and least abundant in pool mesohabitats, and this pattern remained consistent across two seasons of the year. In both Spring and Fall seasons, there were no detectable differences in microhabitat use between adults and juveniles, or between adult males and adult females, or between Form I and Form II males. These results suggest that *C. williami* prefers riffle mesohabitat, although the species also occupies run and pool mesohabitats. Given the level of endemism the species displays, maintenance of quality riffle habitat within its range is likely important to its long-term persistence.

### 151 - The role of disease in the rapid expansion of an invasive crayfish

Sara Bolds, Lindsey Reisinger, Donald Behringer

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Infectious disease dynamics within a system can influence the establishment and spread of a non-native species during an invasion event. However, much is still unknown regarding the role of disease in crayfish invasions. To explore this mechanism, we examined disease prevalence and pathogen communities in a critically imperiled crayfish endemic to northeast Florida, *Procambarus pictus*, and an invasive congener, *Procambarus spiculifer*. Microsporidian infections in the abdominal muscle tissue ("porcelain disease") occur in both species, although infections in *P. pictus* were documented prior to the estimated introduction of *P. spiculifer*. We collected *P. pictus* and *P. spiculifer* with gross pathology indicative of microsporidian infection during trapping and net sweep surveys, and conducted behavioral assays to assess effects of the parasite on crayfish muscle tissue and survival. Crayfish were dissected and multiple tissue types fixed for molecular and histological analyses. Infected crayfish had slower righting response and increased mortality. Two species of microsporidia were detected in *P. spiculifer*, including a new species in the genus *Nosema*. The same *Nosema* species was also found to parasitize *P. pictus*, which indicates that pathogen spillover or spillback mechanisms are occurring. We are in the process of examining pathogen communities between species, and between native and introduced populations of *P. spiculifer*. This information will help predict future impacts of the *P. spiculifer* invasion, promote the conservation of *P. pictus*, and help guide efforts to control invasive crayfish to prevent pathogen transmission to native species.

## 152 - Systematic de-complexing of a widespread crayfish species complex

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*Cambarus* sp. C. is a widespread species complex, ranging from Columbia, South Carolina north to southeastern Pennsylvania. In 2018, we embarked on a large collaborative undertaking, using integrative methods to disentangle the *C. sp. C* complex, with an initial aim of identifying - and queueing for formal description - taxa of conservation concern. To date, we have systematically sampled nearly 1500 sites throughout the range of *C. sp. C*, and have generated a multi-locus molecular dataset of over 2000 individuals. Results of our (still) preliminary analyses indicate the potential for more than 40 taxa within the complex, only six of which are currently named. Biogeographic context has been crucial to our efforts to understand species limits, identify putative introductions, and reexamine the diversity of other Atlantic Slope crayfishes. Our sampling scheme allows us to determine conservation status in "real time", which in turn strongly influences how we prioritize our taxonomic efforts. 153 - Conservation and Taxonomic Assessment of an Undescribed Crayfish Species in Coastal Virginia

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been collected from nine localities. Phylogenies produced from the mitochondrial COI and 12S genes and the nuclear 28S gene suggest that the Virginia population of *C. aff. fodiens* may be a distinct species. Additional morphological and genetic data will lead to a better understanding of where the separation in classification lies among the Virginia population and provide insight to the management and conservation of these species. Ultimately, we seek to comprehend the genetic relatedness of the Virginia populations and provide a conservation and taxonomic assessment that will contribute to our understanding of global crayfish biodiversity.

154 - An Investigation of Underestimated Diversity in *Faxonius placidus*, *F. forceps*, and *F. durelli*.

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A major issue facing crayfish conservation is a limited understanding or underestimation of valid species that are currently unrecognized. These undescribed species are often grouped with sister species or not considered for review in management planning. Crayfish diversity could be lost before species complexes can be resolved and the implementation of conservation action. Mitochondrial barcodes and single nucleotide polymorphisms (SNPs) are used to illuminate the need for management and conservation action in previously unrecognized species by providing foundational information and identifying species boundaries in crayfish species complexes. Narrowly endemic species, that are currently recognized as a singular, wide-ranging species, are often at risk due to other invasive crayfishes, pollution, and stream disruption. Species such as *F. placidus*, *F. forceps*, and *F. durelli*, are especially problematic as various threats to each of these wide-ranging species can go unnoticed. These three species

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*Creaserinus fodiens* (Cottle 1863), commonly known as the digger crayfish, is a primary burrowing crayfish that inhabits complex burrows in wetlands, seasonal pools, wooded floodplains, and roadside ditches. Historically, *C. fodiens* ranges from Ontario, Canada following the United States down to Texas and through four Atlantic slope states including Virginia, Maryland, North Carolina, and South Carolina. The Atlantic slope populations are geographically isolated from other *C. fodiens* populations by the East Coast continental shelf, Blue Ridge Mountains, and Appalachian Mountains. Despite having a large geographical distribution, the Atlantic slope clades of *Creaserinus* spp. are poorly understood. To date, 118 sites within eastern Virginia have been sampled using minnow traps, hand excavation, and dipnetting. Of these, *C. aff. fodiens* has only <sup>2</sup>Auburn University, Auburn, AL, <sup>2</sup>United States Fish and Wildlife Service, Warm Springs, GA

Conservation of aquatic taxa is impeded by a lack of understanding of the relationships between physiology and thermal tolerance. To investigate the physiological basis of thermal tolerance we evaluated responses of three common, secondary-burrowing crayfish (*Procambarus clarkii*, *C. latimanus*, and *C. striatus*), and one state-threatened primary burrower (*C. harti*), to acute thermal stress using an adverse outcome pathway (AOP) framework. Objectives were to: 1) test for differences in thermal tolerance between species; and 2) trace physiological responses to thermal stress across multiple levels of organization (cellular to organismal). We evaluated the effect of temperature at the cellular level using an assay of respiratory enzyme activity (ETS), at the organismal level via respiration rate (MO<sub>2</sub>) and loss of equilibrium (LOE), and at the population level via upper thermal limits (UTL). Crayfish were acclimated to 25°C and then exposed to increasing temperature (2°C/h) until reaching UTL. Thermal performance curves were developed for ETS and MO<sub>2</sub>. For all species, ETS activity peaked first, followed by a peak in respiration, followed by LOE, and finally UTL. The most sensitive indicator of thermal tolerance was LOE with *C. latimanus* being the most sensitive and *P. clarkii* being the most tolerant. The UTL did not differ among species (range: 39.4-39.8 °C). Crayfish reached LOE 1-4 °C after MO<sub>2</sub>max and UTL 3-7 °C after MO<sub>2</sub>max. For all species, LOE occurred within a 96-73% reduction of ETSmax and UTL occurred with a 81-69% reduction in ETSmax. Results suggest that acute thermal thresholds protective of common species will be equally protective of a rare primary burrower. Understanding linkages

are found in the Cumberland and Tennessee river drainages, a region known for numerous endemic fish and mussel species that are often sympatric with unique populations of the previously mentioned crayfishes. We used nuclear (SNPs) and mitochondrial (CO1) markers for *F. placidus* and mitochondrial markers (CO1/16s) for *F. forceps* and *F. durelli* to investigate the potential undescribed diversity in these currently defined species. Regarding *F. placidus*, both the mitochondrial and nuclear based results revealed multiple undescribed species. Our results also provide genetic evidence that some of these undescribed species, previously thought to be *F. placidus*, are instead paraphyletic with respect to *F. shoupi*. This outcome shows that both future research and the taxonomic revision of common wide-ranging species are crucial to address uncertainties in crayfish conservation.

#### 155 - Effect of psychoactive substances on cardiac and locomotory activity of juvenile marbled crayfish *Procambarus virginalis*

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Pharmaceutically active compounds are common and increasing in the aquatic environment. Evidence suggests they have adverse effects on nontarget organisms, and they are classified as emerging pollutants for a variety of aquatic organisms. To determine the effects of environmentally relevant levels of psychoactive compounds on non-target organisms, we analyzed cardiac and locomotory activity in early developmental stages of marbled crayfish *Procambarus virginalis*. Responses to sertraline, methamphetamine, and a mixture (Mix) of citalopram, oxazepam, sertraline, tramadol, venlafaxine, and methamphetamine at a concentration of  $1 \mu\text{g L}^{-1}$  of each compound were assessed. On day four of exposure, cardiac activity was recorded for 5 min, and, on day eight, locomotory activity was recorded for 15 min. There was a significant increase ( $p < 0.05$ ) in heart rate in methamphetamine- and Mix-exposed juveniles compared to the unexposed control. No significant effect was observed with sertraline exposure. Velocity, distance moved, and percent time active did not significantly differ ( $p = 0.15$ ) in exposed and control animals. The findings revealed that low concentrations of chemicals and their mixtures can modify the physiological state of aquatic animals without outward manifestations (activity, distance moved, and velocity). Aquatic animals can be impacted earlier than is visible, but effects can potentially lead to substantial changes in populations and in ecosystem processes. Additional research to investigate chemical combinations, exposure systems, and organism physiological and molecular responses may provide evidence of broad impact of environmental pharmaceuticals.

#### 156 - Linking physiology and upper thermal limits in crayfish using an adverse outcome pathway approach

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between physiological responses and acute thermal stress may ultimately reduce the number of individuals and assays required for determining effects of acute thermal stress on species of interest via development of accurate AOP models.

## SYMPOSIUM: Conservation Genetics

Matt C. Estep, Appalachian State University, Boone, NC and Ashley Morris, Furman University, Greenville, SC

#### 157 - Genetic Diversity Within, and Hybridization Between, Two Imperiled North American Pitcher Plants

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Habitat loss, environmental modification, and direct exploitation have led to many plant species becoming rare or endangered. These activities have also reduced population genetic diversity, which significantly influences the ability to adapt to environmental shifts. *Sarracenia* species (North American pitcher plants) are rare, perennial carnivorous plants typically found in bogs with acidic, nutrient poor soils. *Sarracenia jonesii* (Mountain Sweet Pitcher Plant; federally endangered) and *Sarracenia purpurea* var. *montana* (Mountain Purple Pitcher Plant; federal species of concern) cooccur in some habitats, and the presence of phenotypic intermediates in these sites led researchers to suspect that hybridization has occurred. Although the intermediates have morphological traits of both *Sarracenia jonesii* and *Sarracenia purpurea* var. *montana*, this potential hybridization had not been investigated with molecular genetic tools. In addition, the diversity of parental populations remains unclear. The purpose of this research was to determine levels of genetic diversity in populations of these rare plants, then confirm or deny hybridization between the species. Tissue samples from the two *Sarracenia* species and their potential hybrids were non-destructively collected from three western North Carolina sites, and DNA was extracted. Two sets of informative microsatellite loci were used to analyze DNA extracts; one indicated population level diversity, while the other determined hybrid ancestry. Fragment analysis was used to assess allele sizes, and data were analyzed with the polysat and poppr packages in R. Genetic diversity within populations was relatively high, though it varied among sites and species. Phenotypic hybrids had alleles from both parental species. Hybrids overrepresented ancestry of the less common species at their sites of origin, perhaps due to pollinator behaviors. Results will help conservation biologists preserve both discrete species and make management decisions, particularly during plants' reproductive seasons.

158 - Genetic Contributions of Hatchery-Stocked Walleye to Douglas Reservoir

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Walleye (*Sander vitreus*) has been a popular sports fish for anglers in Tennessee for many years. However, some reservoir populations of walleye are in decline and fail to naturally produce enough offspring to sustain fishery desires. To support reservoir populations, the Tennessee Wildlife Resources Agency (TWRA) has been stocking Tennessee reservoirs, oftentimes yearly, with hatchery-raised fish. Captive rearing programs for walleye are costly, and the contribution of stocked fish to the existing population is not well known. The objective of this project is to determine the genetic contribution of hatchery-reared fish to reservoir populations of walleye in Tennessee. We used genome-wide Single Nucleotide Polymorphisms (SNPs) to estimate the genetic contribution of hatchery-reared fish to the existing walleye population at Douglas Lake, a popular fishing reservoir in Tennessee. We genotyped hatchery broodstock and the existing reservoir populations prior to stocking and after stocking was initiated. Results from this study will be used to assess population introgression of stocked fish and the effectiveness of TWRA stocking practices on improving walleye populations.

159 - The Effects of a Past Augmentation on the Fitness of the Federally Endangered Southern Appalachian Endemic *Geum radiatum* Michx.

Morgan Gaglianese-Woody, Matt C. Estep

Appalachian State University, Boone, NC

*Geum radiatum* Michx., or Spreading Avens, is a rare cliff-dwelling endemic in Rosaceae that is restricted to fifteen fragmented populations above 1500 meters along the North Carolina and Tennessee border. This long-lived perennial has been listed as federally endangered since 1990 and is considered imperiled (G2) in North Carolina and critically imperiled (G1) in Tennessee. *Geum radiatum*'s small and fragmented range and unique life history renders it vulnerable to extinction within the next several decades; this threat is exacerbated by the effects of anthropogenic climate change and habitat loss. There is therefore a dire need for integrative conservation management strategies that consider genetic factors. This project aims to estimate the fitness effects of a historical augmentation of a *G. radiatum* metapopulation to determine if a genetic rescue occurred. We first identified hybrids, native parents and augmented parents using genotyping technology. We then quantified performance differences among those genotypes using the demography data provided by the National Park Service and U.S. Forest Service. The results of this study will contribute to our understanding of transplantation as a conservation management strategy for *G. radiatum* and long-lived perennials alike.

160 - Conservation genetics in the endangered whorled sunflower *Helianthus verticillatus* (Asteraceae)

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<sup>1</sup>Center for Biodiversity Research, Memphis, TN, <sup>2</sup>University of Memphis, Department of Biological Sciences, Memphis, TN

Genetic diversity among and within populations of species is critical for survival in a changing world. Conservation genetics links studies of population genetic variation with conservation managers to provide scientific data for species management strategies. We study the conservation genetics of whorled sunflower, *Helianthus verticillatus* (Asteraceae), an endangered sunflower species endemic to the southeastern United States. Until about five years ago, only five populations were known to exist in Alabama, Georgia, and Tennessee, but additional populations of the species have been discovered in Mississippi and Virginia. Population genetic work using microsatellites revealed that the newly discovered Mississippi plants were genetically distinct from all other populations. To further investigate these new Mississippi individuals and compare them with the rest of the species, we used a combination of genome-wide SNPs and sequencing of phylogenetically informative loci to assess genetic diversity and relatedness on a small subset of individuals. We also included the closely related species, *H. grosseserratus*, using a collection from Shelby County, Tennessee from the University of Memphis Herbarium. This specimen was collected with only the top portion of the plant. Interestingly this sample fell within the *H. verticillatus* clade in some analyses. We report the comparative genetic analysis from this subset of samples and note that additional work will be required to further define the extent of genetic diversity in the whorled sunflower.

161 - Assessing the Genetic Diversity and Structure of Gray's Lily (*Lilium grayi*)

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*Lilium grayi* S. Watson, Gray's Lily, is a threatened perennial herb endemic to high elevations in the Southern Appalachians of Virginia, North Carolina, and Tennessee. *L. grayi* is an iconic species in the grassy bald plant community, also growing in bogs, mesic forests and wet meadows. *L. grayi* faces multiple challenges, including an invasive fungal pathogen, continued habitat loss, potential hybridization with the more common *L. canadense* and it has many small and seemingly isolated populations. Threatened species that occur in small and isolated populations are more likely to lose genetic diversity over time due to stochastic processes including genetic drift, thus molecular data from those populations is a critical research need. This study used 11 published *Lilium* microsatellite markers to evaluate the genetic diversity and structure of 2 *L. canadense* and 24

*L. grayi* populations, representing a majority of the 46 remaining extant *L. grayi* populations, including 6 populations thought to be experiencing introgression from *L. canadense*. In total 612 individuals were genotyped across the 26 populations, ranging from 3-57 individuals per population with all available individuals sampled in 19 populations. Calculated genetic diversity statistics include an across-population average expected heterozygosity of 0.338, average observed heterozygosity of 0.346, and average number of effective alleles of 1.806. Genetic structure analysis suggests an overall high level of gene flow between populations, as well as the existence of three separate genetic clusters of individuals. Alternatively, there was also high support for two separate genetic clusters of individuals. These data will be instrumental to Federal and State governments in determining the conservation status for this species and informing future management decisions.

#### 162 - Understanding genetic diversity and population genetic structure in western North Carolina American ginseng (*Panax quinquefolius*)

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American ginseng (*Panax quinquefolius* L., Araliaceae) is a long-lived herbaceous plant that is uncommon to rare in eastern deciduous forests of North America. This species is grown worldwide for commercial medicinal use. In the southern Appalachians, however, wild ginseng has been collected for centuries, primarily for export to East Asia, where an estimated 95% of wild ginseng is consumed for use in traditional medicine. Concerns about ginseng overharvest has led to its designation by CITES (Convention on International Trade in Endangered Species) as a species at risk of extinction, and caused the US Forest Service to suspend its distribution of ginseng harvest permits in the Pisgah and Nantahala Forests in 2021. Ginseng may be particularly vulnerable to extirpation events because harvesting generally removes larger, older individuals from populations, and plants take at least five years to produce seeds. Between 2014-2022, we sampled leaflets from 10-30 individuals per population in 22 western North Carolina ginseng populations that varied in harvest pressure, population size, and demographic structure. We are analyzing microsatellite diversity at seven polymorphic loci to characterize population genetic structure, estimate genetic diversity, test for gene flow among populations, and relate these factors to harvest pressures. Our findings will shed light on the evolutionary potential of this species in western North Carolina and support ongoing efforts to conserve this vulnerable, culturally valuable herb, in the face of climate change and continuing commercial demand.

#### 163 - The importance of understanding genetic and clonal structure for species recovery: a case study using federally endangered *Sagittaria fasciculata*

Skyler Fox, Ashley Morris

Furman University, Greenville, SC

*Sagittaria fasciculata* (bunched arrowhead; Alismataceae) is a semi-aquatic herbaceous perennial that inhabits marsh-like seepages of free-flowing water. This federally endangered plant is only found in two counties: Greenville County, South Carolina, and Henderson County, North Carolina.

Published genetic work about *Sagittaria fasciculata* is lacking, although understanding this plant's genetic structure is vital for species recovery and resiliency. Additionally, the reproductive strategy of a species can strongly influence its genetic structure. *Sagittaria fasciculata* has the ability to reproduce clonally via rhizomes as well as sexually via seed. Therefore, our goal in this study was to understand genetic and clonal structure within *S. fasciculata* and to analyze how these play a role in the management of the species. Plant material was collected for 162 *S. fasciculata* individuals across eight sites, spanning four of the five watersheds where the species is located. We screened a total of 73 loci, and 31 of these loci amplified cleanly and were polymorphic. Five of the 31 loci were selected for the current project. Our results show that overall genetic diversity of *S. fasciculata* is low. The species is highly dependent on clonal reproduction, but sexual reproduction still plays a role. Two sites, one on the Reedy River and another on the Enoree River, have no genetic variation, and each appears to be a single clone. Next steps for the conservation of this species will be discussed. In the future, we will employ high throughput sequencing of all microsatellite loci we have developed, expanding the spatial extent of our field sampling.

#### 164 - A tale of two bells: population and clonal structure in the *Shortia* complex of the Carolinas

Ashley Morris, Skyler Fox

Furman University, Greenville, SC

Oconee bells (*Shortia galacifolia* and *S. brevistyla*; Diapensiaceae) are an iconic component of the flora of the Blue Ridge of NC and Piedmont of SC. *Shortia galacifolia* is more widely distributed, known from Oconee and Pickens counties in SC, Transylvania and Jackson counties in NC, with *S. brevistyla* known only from McDowell County, NC. More than 60% of the range of *S. galacifolia* is thought to have been inundated by the creation of Lake Jocassee in the 1970's. The species persists along the margins of the lake and along associated rivers and tributaries. Limited genetic data have been published on this complex, but both species are presumed to be dependent on clonal reproduction by rhizomes or stolons. For this study, we developed novel nuclear microsatellite loci to evaluate clonal and population structure within and between species. A total of 63 loci have been screened so far, and 25 of these loci amplified cleanly in both species and were variable in at least one of the two species. An additional locus amplified cleanly for *S. brevistyla* but not *S. galacifolia*. Seven of these 26 loci were used for expanded sampling in the current study. We included three sampling locations for *S. brevistyla* ( $n = 61$ ) and seven for *S. galacifolia* ( $n = 111$ ). Our data clearly support recognition of these two taxa as separate entities, with only three alleles out of 35 shared between the two and each species exhibiting private alleles. *Shortia*

*brevistyla* was monomorphic at all loci included here, while *S. galacifolia* exhibited higher than expected genetic variation within and among sites. *Shortia brevistyla* appears to be a single individual, while clonality was limited within *S. galacifolia* at most sites. The implications of our findings will be discussed, along with additional insights from the remaining loci screened so far.

165 - Conservation Genetics of Timber Rattlesnake in Western NC: a beginning.

Matt C. Estep<sup>1</sup>, John Sealy<sup>2</sup>

<sup>1</sup>Appalachian State University, Boone, NC, <sup>2</sup>NC Timber Rattlesnake Conservation Project, Stokesdale, NC

The Timber Rattlesnake (*Crotalus horridus*) is the only rattlesnake of the temperate deciduous forests of eastern North America and faces growing and varied conservation challenges range-wide. These conservation challenges are exacerbated by the reproductive characteristics of female Timber Rattlesnakes which include late age at maturity and infrequent reproduction. North Carolina lists the Timber Rattlesnake as a Species of Special Concern. The greatest threat to the species in North Carolina is the loss and fragmentation of remaining habitats as a result of anthropocentric encroachment. The mechanism of this threat is the loss of genetic diversity due to population isolation (genetic drift) and inbreeding (loss of heterozygosity) due to decreases in population size from road mortality. This project aims to evaluate the genetic diversity and population structure of NC Timber rattlesnakes to understand their current status and to provide genetic data for future population management.

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## SYMPOSIUM: SISRIS: Supporting Inclusive and Sustainable (collections-based) Research Infrastructure for Systematics

Shawn Krosnick, Tennessee Tech University, Cookeville, TN and Andrea Weeks, George Mason University, Department of Biology and Ted R. Bradley Herbarium, Fairfax, VA

### WELCOME/OVERVIEW

181 - Discovering together "who dunnit?": building a borderless knowledge community

Deborah Paul

*Species File Group, Prairie Research Institute, University of Illinois Urbana-Champaign, Champaign, IL*

Knowing definitively who collects or identifies a specimen offers new benefits for both individuals and groups. Usually, digitized collections pivot around the "what" and include where and when. The who information ends up as non-standardized "text strings" making it difficult to index or search, which limits usefulness for everyone. If we know more about the who, we gain a searchable data asset offering many including collectors, collections staff, researchers, underrepresented groups, and the public, unique insights.

Unique identifiers for people provide the means to do this (e.g. Wikidata "Q numbers", ORCID "IDs"). Of course, not everyone associated with specimen vouchering has an identifier like this (yet). For current and historical collections, it turns out we can all work together to assign these people identifiers. Using Wikidata as a shared knowledge management tool means we can all contribute to implement effective knowledge transfer, reduce duplication of effort, and do this work openly allowing anyone who wants to, the chance to help. Then, using Bionomia software, anyone can help to link specimen records definitively to a person with a globally unique identifier making all sorts of new discoveries and metrics possible. To uncover tacit people information and its power, we need to find ways to promote identifiers, engage more people to do this work worldwide, store these IDs in our collection databases, and increase Wikidata entries for people associated with vouchered collections (Groom, et al 2022). Once these data exist, we can reveal hidden individuals and their contributions to science and compile individual and institutional-level metrics. In this symposium, you will discover who is doing this work, how it is being done, who benefits and how, along with outstanding community needs. With these data we can better understand our past, and illuminate the future, for people and the data.

182 - Connecting Scientists and their Specimens Through Bionomia

David Shorthouse

*Independent, Ottawa, ON, Canada*

Bionomia, <https://bionomia.net> was borne from a simple thought experiment - a diffuse idea that there is utility in stitching together digital tokens that represent natural history specimens and digital tokens that represent human collectors and determiners. Real science might be enabled by shining a light on this class of link, just as new knowledge emerges when lights are shone on other metadata elements in the grand envelope of natural history such as geographic places, taxonomic names, and time. But, there is clearly more than science at play when linking specimens to people. Anyone with a passing curiosity in genealogy, a desire to honour the memory of their academic advisor, or a wish to learn about the

characters that formed their local natural history museum is struck by something deeper here than mere links between tokens. Bionomia is a small chink of light that on occasion illuminates the darkness of colonial pasts, enlightens us about the unrecognized champions of natural history, and elevates our future scholars and natural historians. The visionaries behind the Integrated Digitized Biocollections (iDigBio), the Global Biodiversity Information Facility (GBIF), and other networks have permitted this new way to pivot on the names of people; it is simultaneously a hyper-local and a global prism. While newly emerging ideas and technologies like Digital Extended Specimens are inspirational and will absolutely result in more data flow, more data transactions, more data processing, and new science, let's be mindful as these unfold. Let's remind ourselves and our decision-makers that the narratives in our natural history collections are interconnected, holistic, human stories imbued with emotion. These are the engines of agency.

#### 183 - Digital data sleuthing and storytelling as tools to engage students with the unsung heroes of natural history collections

Olubunmi Aina<sup>1</sup>, Adania Flemming<sup>2</sup>, Jennifer Kovacs<sup>3</sup>, Shawn Krosnick<sup>4</sup>, Siobhan Leachman<sup>5</sup>, Makenzie Mabry<sup>6</sup>, Molly Phillips<sup>7</sup>

<sup>1</sup>Allen University, Columbia, SC, <sup>2</sup>iDigBio, Florida Museum of Natural History, Gainesville, FL, <sup>3</sup>Agnes Scott College, Decatur, GA, <sup>4</sup>Tennessee Tech University, Cookeville, TN, <sup>5</sup>Wikimedian and Data Curator, Auckland, New Zealand, <sup>6</sup>Florida Museum of Natural History, Gainesville, FL, <sup>7</sup>Bioquest, Gainesville, FL

Natural history collections (NHCs) are well-understood as repositories of invaluable data on living organisms, and massive efforts over the last twenty years have been directed at their digitization. However, one important type of specimen data has been overlooked and inconsistently applied: the names of people who collected and/or identified the specimen. Unlike Linnaean binomials, human names tend to be poor identifiers (not unique or stable), and many times only a single person is credited with collecting and/or identifying a species even when several people assisted. Names are frequently disambiguated over the course of someone's lifetime or across institutions as curators work to independently digitize specimen data in their collections. These issues perpetuate bias in who is credited as a collector or identifier, resulting in hidden figures that contribute to science but are never given proper attribution for their work.

Fortunately, new tools are available to facilitate attribution and improve the quality and consistency of collector and identifier name use, including ORCID, Bionomia, and WikiData. These tools are free and open, meaning anyone can help add and improve data about the people involved in documenting biodiversity. In the 2022 BIOME Institute, a working group formed (called The Hidden Figures Network) to develop educational modules that help university students discover hidden figures in NHCs. Users are trained in data-sleuthing techniques to ensure proper attribution of specimens collected and identified by these hidden figures. Students will help to rewrite the history of science as they work to give credit to people just like themselves. The workflow and associated activities will provide meaningful opportunities to bring awareness and highlight underrepresented scientists through personal storytelling. Perhaps even more importantly, students who participate in these modules can further continue the cycle of increased attribution as they share their skills with others.

#### 184 - An exception proves the rule: Lena Artz (1891–1976) and her legacy of botanical specimens.

Andrea Weeks

George Mason University, Department of Biology and Ted R. Bradley Herbarium, Fairfax, VA

Lena Artz is one of many thousands of individuals who documented the plant biodiversity of the southeastern United States in the mid-20th century. But her accomplishments as a pioneering explorer of the Massanutten Mountains, which were uncovered only because of ongoing herbarium digitization in Virginia, is a case study of how interrogating the legacies of poorly known collectors can advance botanical research in the present day. In 2019, the collection notebooks and professional papers of Lena Artz were discovered during the salvage of the Lord Fairfax Community College Herbarium. Despite being occasionally acknowledged in print as an early contributor to knowledge about Central Appalachian shale barren ecosystems, biographical information about Lena Artz or her professional accomplishments were largely undocumented at the time of the discovery. This presentation describes how archival research, gumshoe detective work, and the curation of Artz's ca. 2000 sheet exsiccatae across multiple institutions have worked synergistically to uncover the life and life's work of this hidden figure in botany. The results provide insight on why she, like many other collectors whose collections inhabit museums and databases, became forgotten. The results also reveal the time-value of her collections for contemporary studies of rare, threatened and endangered plant species, for capturing the biodiversity of landscapes long since destroyed, and for informing studies of global climate change. Lastly, I show how Bionomia can be used as a tool to improve digital data about specimens across institutions using the curation of Artz's exsiccatae as an example. I argue that assigning universal unique ID's to historical collectors and appending these to their digitized specimen records is a tractable challenge for the herbarium community to meet that not only will help us achieve the vision of the extended specimen concept but will make collections-based research more inclusive in the 21st century. 185 - Delving into the Lives of Herbarium Collectors

Carol Ann McCormick

University of North Carolina at Chapel Hill Herbarium, Chapel Hill, NC

Behind each specimen in an herbarium is a person who collected that specimen. In the past two decades, one of the joys of my role as Curatrix of the University of North Carolina at Chapel Hill Herbarium (NCU) has been illuminating collectors' professional and personal lives. This helps fill out the story of many specimens, revealing why the collector was at a particular site, or why the collector was interested in that plant (or fungus or fossil). Co-collectors noted on herbarium labels can reveal personal or professional ties. Going beyond the specimen, I use tools such as census data, school yearbooks, newspaper articles, Google Scholar, JSTOR, ProQuest, and genealogical databases (e.g., Ancestry.com) to document

collectors' lives which can then lead to a greater understanding of their botanical interests and motivations. Collaborating with librarians, archivists, and alumni records technicians is often particularly fruitful, as not all information is on-line. Herbarium database portals (e.g., lichenportal, sernecportal, mycortal) also allow me to follow a collector's whereabouts, interests, and co-collectors chronologically. Some collectors pose particular challenges: those who are outside of academia or those who change their name over the course of their career are particularly difficult to trace. However, unusual collecting localities, unexpected associates, and cryptic notes on specimen labels all add to the fun of the detective work.

I have posted ~ 200 biographical sketches on the Herbarium's website. These also serve a public outreach function: it's not uncommon for family members to find these sketches, to offer more information about their ancestor, and to become more interested in the Herbarium and NC Botanical Garden.

I will share how I've learned about some of my "favorite" collectors including F. Stuart Chapman, Frances Foust, Percy Gentle, and Ferdinand Blanchard.

## LUNCH

### 186 - Prolific plant collectors and the HBCU legacy: Charles Parker as a case study

Janelle Burke<sup>1</sup>, Dominique Pittman-Kidd<sup>1</sup>, Daniel Koenemann<sup>2</sup>

<sup>1</sup>Howard University, Washington, DC, <sup>2</sup>Clayton University, Orangeburg, SC

Charles S. Parker Ph.D., a botanist and former Chairperson at Howard University, was a prolific plant collector. Over his career, Parker collected over 2,000 specimens which included over 900 different species. He collected all over North America, though most of his collections were concentrated in the Pacific Northwest, Mid-Atlantic, and Southeast regions of the United States. His collections in Idaho and Washington contributed greatly to the "Flora of Southeastern Washington and of Adjacent Idaho". Notably, Harold St. John named two new taxa after Parker's collections: *Lathyrus parkeri* H.St.John and *Rosa spaldingii* var. *parkeri* H.St.John. After moving to Washington, DC in the 1930s, Parker continued his collecting and expeditions, building the collection of specimens at the Howard University herbarium (HUDC). Our study documents Parker's contributions to floristic work, and locations of the specimens of this pioneering Black botanist.

### 187 - Uncovering hidden figures at the Marshall University Herbarium through inter-disciplinary collaborations

Pamela Puppo<sup>1</sup>, Lori Thompson<sup>2</sup>, Lindsey Harper<sup>2</sup>

offered at the then State Normal School at Marshall College. This inter-disciplinary collaboration is helping us bring new awareness of the value of our collection, not only from the biological point of view, but also from a historical perspective.

### 188 - The State of Herbarium Backlogs: Perspectives from Bryophyte Collections

Robin Lewis<sup>1,2</sup>, Hannah Bendell<sup>3</sup>, Jessica Budke<sup>4</sup>

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In order to fulfill its role in the botanical community, an herbarium must have its specimens processed, identified, and accessioned so that botanists can consult these materials during their research. Many herbaria, however, struggle to keep up with the influx of specimens. As a result, specimens may accumulate while awaiting further processing, leaving the institutions with what is known as a 'backlog'; yet we know little about the content of such backlogs. In this study, we surveyed herbarium staff about their institution's backlog of unaccessioned specimens, focusing on the number and taxonomic lineage of backlogged specimens as well as the challenges associated with curating these specimens. We asked additional questions about unaccessioned bryophyte specimens in order to explore these collections in more depth. Eighty-seven staff members responded to our survey. Approximately 92.0% of staff indicated that their herbarium had a backlog with an average of approximately 30,372 unaccessioned specimens per herbarium. According to Index Herbariorum there are 3,426 active herbaria in the world, and thus we estimate that ~104 million specimens may be present in herbarium backlogs. While bryophytes only represent 9% of the accessioned specimens reported by our study participants, they are overrepresented in the backlog, comprising 16% of all unaccessioned specimens. According to our staff respondents, slightly more than half of backlogged bryophyte specimens are stored in field packets without labels and approximately three-quarters of the unaccessioned bryophytes are

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<sup>1</sup> Marshall University, Huntington, WV, <sup>2</sup>Marshall University Libraries, Huntington, WV

The Marshall University Herbarium (MUHW) in Huntington, West Virginia, is the second largest herbarium in the state with ca. 52,000 specimens. Most of the collection is composed of vascular plants, but MUHW also houses bryophytes, algae, fungi, plant fossils, and a small ethnobotanical collection. The herbarium also counts with a modest library, and a manuscript collection that includes correspondence from its first director, Dr. Frank Gilbert, dated back to the 1920's, and a herbarium book from 1896. With the help of archivists, we uncovered details of the early years of the Marshall University Herbarium and the people behind some of the documents present there. The Herbarium started around 1896–1897, years before its first director was hired in 1927. The 1896 herbarium book was made by Cora Klutz, who was probably a teacher taking the Botany class

not yet identified to species. In addition to describing the challenges associated with the integration of partially curated materials into an herbarium, staff also discussed other limitations. These obstacles included competing institutional priorities, labor shortages, and insufficient bryological expertise. Drawing on suggestions provided by our survey respondents, we offer suggestions regarding how best to accession specimens currently in herbarium backlogs.

189 - Strategies to enhance and sustain an established cyberinfrastructure

Zack Murrell<sup>1</sup>, Herrick Brown<sup>2</sup>, Michael Denslow<sup>3</sup>, Shawn Krosnick<sup>4</sup>, Andrea Weeks<sup>5</sup>

<sup>1</sup>Appalachian State University, Boone, NC, <sup>2</sup>SC Dept. of Natural Resources, Columbia, SC, <sup>3</sup>University of Florida, Gainesville, FL, <sup>4</sup>Tennessee Tech University, Cookeville, TN, <sup>5</sup>George Mason University, Department of Biology and Ted R. Bradley Herbarium, Fairfax, VA

Concepts regarding information infrastructure, or cyberinfrastructure, have evolved over the past thirty years as organizations and institutions developed best practices for “cyber-based information systems.” Lincoln Stein reported in 2008 on the state of affairs of the “biological cyberinfrastructure”, describing four areas of development needed to make data and tools available to enable computational analyses and to facilitate the exchange of knowledge from those analyses. SERNEC, the SouthEast Regional Network of Expertise and Collections, was funded through two National Science Foundation grants to mobilize the community of curators and herbarium affiliates in the Southeast USA to make herbarium specimens and associated data available to the public. The authors have been engaged in developing, maintaining and sustaining the four cyberinfrastructure areas outlined by Stein: data infrastructure, computational infrastructure, communication infrastructure and human infrastructure. The SERNEC effort has generated 5.3 million electronic specimen records to date. Most of the herbaria engaged in the project have fully incorporated the digitization process into their day-to-day workflows. We have demonstrated that a tiered network of herbaria of all sizes, and the people associated with them, has tremendous potential to generate a very large high quality dataset and to drive collaborations among associated researchers. In this presentation we report on our progress, with particular emphasis on the human infrastructure that generated the 15 million specimens housed in the 233 herbaria in the Southeast USA. We discuss the addition of Bionomia as a tool to both connect scientists with these specimens and to extend specimen value. We will explore strategies to engage our community to embrace Bionomia as a part of our growing cyberinfrastructure. The addition of Bionomia to our toolkit demonstrates the organizational value of maintaining and sustaining all four aspects of a cyberinfrastructure to maximize community productivity.

PANEL DISCUSSION

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**Southeastern Chapter of the Ecological Society of America-sponsored Symposium:  
Revisiting some of the plants that William Bartram wrote about in his 'Travels' to  
North Carolina, 1775 and 1776**

Charles Kwit, University of Tennessee Knoxville, Knoxville, TN

190 - Plants noted, and not noted, by William Bartram in and near current-day North Carolina

Charles Kwit<sup>1</sup>, Caleb Hickman<sup>2</sup>

<sup>1</sup>University of Tennessee Knoxville, Knoxville, TN, <sup>2</sup>Eastern Band of the Cherokee Indians, Cherokee, NC

During the years 1773 through 1777, William Bartram embarked on two separate treks that brought him into present-day North Carolina. Bartram documented the biodiversity he encountered in his ‘Travels,’ wherein he noted hundreds of plant species by common or Latin name. Many of these plants have subsequently been the subject of scientific investigations that have broadened our understanding of the natural world. It should be noted, though, that before Bartram’s travels in this area, the knowledge base of those and other plants was well engrained in Cherokee Indian (aniyunwiya) culture. For example, plants in and near the western portion of current-day North Carolina whose flowers grabbed Bartram’s attention on his first trek had longstanding uses by indigenous people; these include azaleas, rhododendrons, trilliums. Bartram’s second trek into current-day North Carolina entailed a more coastal route, where among many other plants he noted tulip trees. Indeed, thousands of plant species exist along the routes of Bartrams ‘Travels,’ and this means he prioritized his documentation to a selection of those present; sometimes Bartram fixated on plants cultivated and foraged by Cherokee Indians (e.g. corn, strawberries), and other times the mere flowering of plants (e.g. “mountain cluster Rose, blushing Rhododendron and fair Lilly of the valley”) resulted in documentation. But the notion of prioritizing plants by surveyors of the western world can come from a biased perceived importance, resulting in many missed species during this process. Surprisingly not noted by Bartram in this specific area was river cane, which was highly used by Cherokee people. Here we explore potential biases in Bartram’s selection of plants he describes and does not describe in his ‘Travels,’ from phenological, to consumptive, to overlooks of importance to Cherokee Indians, and we extend these ideas other cataloguing efforts that followed.

191 - A tale of two Trillium: differences in endemism, life history, and mutualism efficacy in sessile and pedicellate species

Chelsea Miller

*The Holden Arboretum, Kirtland, OH*

When one thinks of southern Appalachian cove forests, especially early in the year, an image of the damp green forest floor covered in white, yellow, pink or maroon trillium flowers may spring to mind. Trilliums are beloved by gardeners, botanists, conservationists, and researchers alike for their showy beauty. Indeed, these elegant plants were not lost on William Bartram as he explored present day Rayburn Co., GA, right before crossing into North Carolina, and noted the presence of *Trillium cernuum* and *T. sessile*. Eastern North America (ENA) constitutes a global biodiversity hotspot for *Trillium*, containing at least 35 of the approximately 50 species worldwide. In ENA, the southern Appalachian region is perhaps the richest in *Trillium* abundance and diversity. As a genus, *Trillium* is divided taxonomically into two groups based on the presence or absence of a pedicel: pedicellate species belong to subgenus *Trillium*, while sessile species belong to subgenus *Sessilium* (formerly *Phyllantherum*). In the southeastern U.S., the diversity of sessile trilliums is particularly high, and many sessile species are characterized by range-restriction and a high degree of endemism. This is especially striking when compared to their pedicellate relatives, several of which boast ranges as large as thousands of kilometers. Here, I present the results of a distribution study conducted for several members of the genus in ENA—including the two mentioned by William Bartram in his ‘Travels’ to North Carolina, *T. cernuum*, a pedicellate species, and *T. sessile*, a sessile species. Based on climatic suitability, sessile species were able to fill significantly less of their potential geographic distributions than their pedicellate congeners, suggesting some non-abiotic factor(s) may be limiting their ability to occupy larger ranges. I then discuss differences life history traits that might explain this finding, focusing on differences in seed dispersal efficacy among sessile and pedicellate trilliums.

192 - When do butterflies beat the birds and the bees? Exploring the pollination ecology of southeastern azaleas (Rhododendron spp.; Ericaceae) and beyond

Mary Jane Epps

*Mary Baldwin University, Staunton, VA*

With many key groups of pollinators facing global population declines, a central challenge of pollination biology is to determine which species of flower visitors actually contribute to a plant’s reproductive success. Previous work on the southeastern azalea *Rhododendron calendulaceum* (flame azalea) found that although many species visit its flowers, the vast majority of pollination is performed by the flapping wings of large butterflies in what is commonly viewed as an extremely rare reproductive strategy. Here, we investigated the pollination ecology of two other southeastern azaleas (*R. periclymenoides* and *R. viscosum*) to determine the relative roles of Lepidoptera and wing pollination in related plant species with slightly differing floral morphology. As with *R. calendulaceum*, large butterflies were found to be common visitors of *R. periclymenoides* (pinxter azalea) and these insects readily contacted both male and female reproductive parts with their flapping wings. Field experiments found that compared to freely accessible flowers, fruit set was 3.5 times lower in flowers from which large-bodied flower visitors (butterflies and occasional hummingbirds) were selectively excluded, and that their fruit set was no higher than that of flowers experimentally barred to all potential pollinators. By contrast, our field observations found that the smaller, white, and heavily scented flowers of *R. viscosum* (swamp azalea) were rarely visited by large butterflies, although a surprisingly diverse range of other visitors including night-flying moths frequently contacted both male and female reproductive parts as needed for pollination. Field experiments suggested that night-flying moths and day-flying visitors both contribute to pollination in this species, although results were not conclusive due to the high self-compatibility of our study plants. Beyond the genus *Rhododendron*, results of additional investigations into wing-mediated pollen transfer by Lepidoptera in other southeastern plant species offer clues of floral morphologies that may be predictive of this little-studied mode of reproduction.

193 - *Liriodendron tulipifera*: from Bartram’s Travels to Changing Forest

Lisa Kelly

*University of North Carolina at Pembroke, Pembroke, NC*

As one of eastern North America’s most unique and widely distributed hardwoods, *Liriodendron tulipifera* (yellow poplar or tulip tree) did not receive the attention we might expect from William Bartram’s *Travels*. The only other species (*L. chinense*) in the genus *Liriodendron* is native to East Asia, and the two share an ancient lineage that has been the focus of studies in angiosperm evolution. As true of the temperate deciduous forest during Bartram’s time, the geographic range of *L. tulipifera* is much the same today. Albeit, the forest is much younger, and *L. tulipifera* has become more abundant in the wake of both selective logging and the decline of *Castanea dentata* (American chestnut). Scientists now recognize considerable regional variation in *L. tulipifera* populations and possibly a subspecies in peninsular Florida. The species has become a popular ornamental, valued for utilitarian purposes, and the hybrid of *L. tulipifera* and *L. chinense* is planted extensively in China. A few old-age trees are alive today, offering a glimpse of the impressive height and beauty characteristic of *L. tulipifera*. It remains to be seen how this species will adapt as anthropogenic climate change and disturbances further reshape America’s forests.

194 - Current Ecology of Rivercane in Riparian Restoration

Alyssa Quan<sup>1</sup>, Beverly Collins<sup>2</sup>, Adam Griffith<sup>3</sup>

<sup>1</sup>*University of Georgia, Athens, GA*, <sup>2</sup>*Western Carolina University, Cullowhee, NC*, <sup>3</sup>*EBCI Cooperative Extension Services, Cherokee, NC*

Rivercane (*Arundinaria gigantea*), a native bamboo plant culturally important to Native American peoples, plays a major role in sustaining both cultural practices and habitat for various riparian species. Though once prolific throughout the Southeast, approximately 2% of rivercane's original distribution remains today. The loss of cane coverage has also led to a loss of botanical knowledge on rivercane over time. Current efforts to restore riparian ecosystems have led to growing interest in the reestablishment of canebrakes, as well as increasing collaboration between tribes in the Southeast and other organizations who focus attention on rivercane. As a native species, many restoration projects seek to utilize rivercane in order to repair riverbanks and provide native habitat to riparian animals, as well as maintaining cultural tribal practices. Proliferation of rivercane relies on successful transplantation of culms, due to the lack of existing rivercane stands. Future research on rivercane should seek to improve understanding of rivercane life history through combined forms of knowledge in order to develop sustainable management practices in concert with restoration goals.

195 - Cane on his brain: Bartram's observations of *Arundinaria* spp.

Adam Griffith<sup>1</sup>, Beverly Collins<sup>2</sup>, Alyssa Quan<sup>3</sup>

<sup>1</sup>EBCI Cooperative Extension Services, Cherokee, NC, <sup>2</sup>Western Carolina University, Cullowhee, NC, <sup>3</sup>University of Georgia, Athens, GA

In his extensive travels throughout North Carolina and the southeast, Bartram provided fairly detailed descriptions of *Arundo gigantea* (likely referring to *Arundinaria*). While he uses the binomial of this plant four times, his use of the term 'cane' approaches 50 in number and varies widely from cane forests and cane pastures to cane swamps and cane brakes. The variation in habitat and soils well described by Bartram and the known general location of these observations allow us to make inferences regarding specific species within the *Arundinaria* genus. This talk weaves Bartram's historical descriptions of cane with our contemporary understanding of the *Arundinaria* genus in North Carolina and the southeast with three distinct species: *A. gigantea* (giant or river cane), *A. tecta* (switch cane), and *A. appalachiana* (hill cane). We discuss the cultural significance of these species to southeast US Tribes with a particular emphasis on the Eastern Band of Cherokee Indians where a significant revitalization effort has increased knowledge about and respect for *A. gigantea*. Recent student research in natural and planted river cane stands has revealed cane growth characteristics important for use in traditional crafts that are sensitive to variation in neighbor density and variation in the amount and quality (red:far red ratio) of light. We discuss this research in the context of background information for ongoing and future river cane revitalization efforts.

## SYMPOSIUM: Joro Spider Summit

Patrick Cain, Georgia Gwinnett College, Lawrenceville, GA

196 - Introduction of the Jorō spider (*Trichonephila clavata*) to the southeastern U.S.: past, present, and future

Michael Sitvarin<sup>1</sup>, Angela Chuang<sup>2</sup>, John Deitsch<sup>3</sup>, David Nelsen<sup>4</sup>, David Coyle<sup>5</sup>

<sup>1</sup>Clayton State University, GA, <sup>2</sup>University of Florida Citrus Research and Education Center, Lake Alfred, FL, <sup>3</sup>Cornell University, Ithaca, NY, <sup>4</sup>Southern Adventist University, Collegedale, TN, <sup>5</sup>Clemson University, Clemson, SC

For nearly a decade, a non-native species has been spreading across the southeastern U.S. after its initial introduction to Georgia. The Jorō spider (*Trichonephila clavata*) arrived in approximately 2014 after traveling from its home range in Asia. This large, colorful species has captivated media attention, and widespread fear of its impacts spread faster than the species itself. Numerous scientific investigations are being launched, as even the fundamentals of its biology in this new geographical range remain to be documented. The spider represents an opportunity to engage the public in environmental awareness, simultaneously generating data needed by scientists to evaluate any ecological consequences of this introduction. It seems the Jorō spider is here to stay, though it remains to be seen how the region's native species may be affected by this newcomer.

197 - Range Expansion and the Ecological Impact of the Invasive Jorō Spider, *Trichonephila clavata*.

David Nelsen<sup>1</sup>, Aaron Corbit<sup>1</sup>, Angela Chuang<sup>2</sup>, John Deitsch<sup>3</sup>, Michael Sitvarin<sup>4</sup>, Harvey Salinas<sup>1</sup>, Emily Hamstra<sup>1</sup>, Logan Bateman<sup>1</sup>, Miguel Rivas<sup>1</sup>, David Coyle<sup>5</sup>

<sup>1</sup>Southern Adventist University, Collegedale, TN, <sup>2</sup>University of Florida Citrus Research and Education Center, Lake Alfred, FL, <sup>3</sup>Cornell University, Ithaca, NY, <sup>4</sup>Clayton State University, Morrow, GA, <sup>5</sup>Clemson University, Clemson, SC

The Jorō spider, *Trichonephila clavata*, is a large, colorful spider native to Asia. It was first reported in Georgia in 2014. Since then, Jorōs have spread throughout North Georgia and into South Carolina, North Carolina, Tennessee, Alabama, West Virginia, and Maryland. Their rapid spread, high local abundance, and status as one of the largest orbweavers in North America warrant more understanding of both their future distribution and impacts on native species. In this study, we surveyed 103 locations across North Georgia to track their spread and investigate if Jorōs negatively impact native orb weaving spiders. We also created several species distribution models (SDMs) to predict where Jorōs might spread in North America. We calculated three measures of the historical presence of Jorōs in an area: 1) distance from the center of the original 2014 observations and kernel density estimates of 2) years present at a location and 3) abundance of Jorōs at a location. We found consistent evidence that all three measures significantly predicted the species richness, Shannon's diversity index, and the inverse of Simpson's diversity index of a location. Our

SDMs suggest that Jorōs will continue to spread much further than their current range. In fact, our models suggests that the climate of the Northern USA is more similar to its native Asian habitat than where they are currently thriving in North Georgia, suggesting these spiders will likely be able to expand their range to even as far north as the Great Lakes Region and possibly beyond. Currently, Jorōs appear to be spreading more quickly toward the northeast than other directions, however, they are expanding in all directions. Future studies will continue to monitor their spread, evaluate the validity of our SDM predictions, and determine if the reduction in diversity of native spider species follows the spread of Jorōs.

#### 198 - What iNaturalist can tell us about Joro spiders and what Joro spiders can tell us about iNaturalist

John Deitsch<sup>1</sup>, Angela Chuang<sup>2</sup>, David Coyle<sup>3</sup>, David Nelsen<sup>4</sup>, Michael Sitvarin<sup>5</sup>

<sup>1</sup>Cornell University, Ithaca, NY, <sup>2</sup>University of Florida Citrus Research and Education Center, Lake Alfred, FL, <sup>3</sup>Clemson University, Clemson, SC,

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In the last year, the joro spider (*Trichonephila clavata*) has experienced a wave of media coverage in its non-native North American range. The resulting public awareness of this spider has created opportunities to monitor its presence and spread in the southeast U.S. in real time via community science (CS) data. However, CS data is subject to a variety of biases which can lead to inaccurate conclusions when not carefully considered. Using data from iNaturalist, we explored patterns of taxonomic bias in CS observations of joro spiders and native orb-weaver spiders (Araneidae) in eastern North America. We quantified observer behavior, from reporting to community identification of observations, for ~20 species of Araneids. We also analyzed how species' life history traits affected their representation in the iNaturalist dataset. Our results show significant taxonomic bias in the iNaturalist dataset. Morphological and behavioral traits influence reporting frequency, experience level of observers, number of community-made identifications on an observation, and the proportion of research-grade observations. Additionally, *T. clavata* is an outlier datapoint in many of these metrics – illustrating that iNaturalist users interact with this species differently than native taxa. For example, users are more likely to report multiple observations of *T. clavata* than native taxa. These results also indicate that caution is needed when using iNaturalist data to compare relative abundances of Joro spiders and native orb-weavers. However, (1) community science platforms like iNaturalist are invaluable tools for documenting the presence and range expansion of invasive species and (2) joro spiders are an excellent model species through which to explore the pros and cons of CS-generated datasets.

#### 199 - Joro spider research in Atlanta Georgia in the invaded region, and in Japan and Korea the native region

Robert Pemberton<sup>1</sup>, Jang Hoon Lee<sup>2</sup>, Makoto Tokuda<sup>3</sup>, Seizi Suzuki<sup>4</sup>

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The Joro spider, *Trichonephila clavata*, was first detected in the North America in 2014 in Georgia, where it has become abundant. It has spread to six other states and is predicted to spread widely in the eastern US. To learn of the Joro's occurrence and relative abundance in the Atlanta Georgia area, timed censuses of it and other aerial orb weavers carried out in 26 forests in the autumn of 2022. The Joro was present in all 26 forests, common in 24, was the most abundant orb weaver in 26, and the only orb weaver in 6. The abundance of the Joro may be due to the absence of specialist natural enemies in the invaded range. To begin to investigate this possible enemy release hypothesis, natural enemies are being sought in both the invaded and native ranges. In Georgia, 40 Joro females were held in outdoor cages for egg laying and 23 egg masses were laid. Twenty of these egg masses were enclosed in aluminum screen envelopes and attached individually to tree trunks to bait for potential egg parasitoids, a common type of specialist natural enemy of spiders. Four egg masses were placed in 5 different forests in Atlanta where the Joro is abundant. The egg masses will be collected in the spring before hatch and held for parasitoid emergence. Egg masses laid by wild Joro spiders in 3 areas of its native range (Kyushu in southern Japan, northern Honshu in Japan and in South Korea), will be collected and held for parasitoid emergence. The presence of parasitoids in the native range but not in the invaded range will give support to enemy release as possible reason why the Joro is abundant in Georgia.

#### 200 - Mud dauber wasps and Jorō spiders: Natural enemies in the United States

Erin Powell<sup>1</sup>, Lisa Taylor<sup>2</sup>

<sup>1</sup>Florida State Collection of Arthropods/Florida Department of Agriculture, Gainesville, FL, <sup>2</sup>Entomology and Nematology Department, University of Florida, Gainesville, FL

Adventive species typically face a different suite of natural enemies than what is found in their native range. Understanding what predators and parasitoids will attack an adventive species is important when considering the impact of the new species in the ecosystem. Natural enemies keep the new population from exploding and may be used in attempted eradication or control with targeted biological control agents. Females of *Sceliphron caementarium*, the black and yellow mud dauber wasp, hunt and paralyze spider prey (up to 25 per day) to provision their nests. At one field site in Otter Creek, Florida, the native golden silk orbweaver, *Trichonephila clavipes*, made up 73.37% of the population's prey with 9 of 30 wasp females exclusively specializing on this species. In 2016, we documented that *Sceliphron caementarium* was also hunting the adventive congener Jorō spider, *Trichonephila clavata*, in Jackson County, Georgia. Additional recent citizen scientist data from Lumpkin County, Georgia again confirmed that *Sceliphron caementarium* is hunting Jorō spider and wasp females may also specialize on this spider species. Spider-hunting wasps are likely one of the most important predators of Jorō spider in the United States. Predators in Jorō spiders' native range, other possible predators of Jorō spiders in the United States, and the role of citizen science will be discussed.

201 - Preliminary genetic analyses of the introduction of *Trichonephila clavata* in Georgia, USA.

Nicholas Mizera, James E Russell

*Georgia Gwinnett College, Lawrenceville, GA*

The Joro spider (*Trichonephila clavata*) is native in eastern Asia (China, Korea, Japan) and was introduced to northeast Georgia around 2014, representing the first record of the species in North America. A rapid population increase observed in the purported area of introduction has drawn attention to the distribution and potential invasive ecology of the species. The distribution and genetic structure of *T. clavata* in Georgia is unknown. Spiders from northeast Georgia and beyond have been collected and analyzed for genetic variation and population structure. Using mitochondrial genetic markers we investigate the genetic structure to address questions related to *T. clavata* introduction and current distribution. A single introduction event followed by rapid population expansion would result in limited genetic variation and population genetic structure, whereas multiple introduction events could result in a population genetic structure defined by local areas of introduction. Our preliminary research using phylogenetic analyses will test hypotheses related to genetic variation among *T. clavata* populations in Georgia.

202 - Molecular analysis of Joro spider gut contents and web carcasses to reveal trophic interactions

Erin Grabarczyk<sup>1</sup>, Kamal Gandhi<sup>2</sup>, Glynn Tillman<sup>3</sup>, Rebekah Wallace<sup>4</sup>, Colton Meinecke<sup>2</sup>, Caterina Villari<sup>2</sup>, Matt Elliot<sup>5</sup>, Brittany Barnes<sup>2</sup>, Jason Schmidt<sup>4</sup>

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Introduction of new species into novel ecosystems may alter trophic interactions; molecular gut content analysis can uncover changes in community structure. However, the patterns of interactions detected may depend on how dietary samples are collected and processed. Here, we used molecular gut content analysis via DNA metabarcoding of fecal samples, prey remains from webs (eDNA), and dissected guts to compare the diet composition and prey diversity of the newly introduced female Joro spider (*Trichonephila clavata*) in the southeastern United States. Arthropod prey DNA was amplified from each of the three material types. Prey remains from webs had the highest diversity, richness, as well as the highest proportion of prey reads relative to Joro predator reads. Recovery of prey reads from DNA of fecal samples and extracted gut content was low and both were overwhelmed by predator DNA. Although fecal samples and female guts had a similarly high proportion of Joro spider reads, fecal samples had higher prey diversity and richness. Moreover, prey DNA was detected from fecal samples several days after capture from the field providing a nondestructive estimate of previous trophic interactions. Our future studies will harvest new approaches to processing spider gut content data, and improve estimating of the ecological roles of the Joro in Southeast landscapes.

## ASB Presentations

### 1 - Effect of Diet on Adult-Egg Cannibalism and Gut Microbiome in Red Flour Beetles

Esther Okamoto, Jennifer Kovacs, Erica Harris

*Agnes Scott College, Decatur, GA*

The red flour beetle (*Tribolium castaneum*) is an ideal organism to study as it completes its entire life cycle in flour, which serves as a habitat, food, and a source of microbiomes. This makes it an easily tractable system for researchers to study various aspects of the beetle's biology and behavior. One of the behaviors that have been studied in this species is cannibalism, which occurs when individuals consume eggs, larvae, or other life stages of their own species. This behavior is known to occur under stressful conditions, such as inadequate nutrition or overpopulation. To study the effects of host diet and population density on the beetle's gut microbiome and cannibalism rates, two experiments were conducted. The first experiment isolated the role of the host diet by rearing flour beetles on different flour diets with varying nutritional content, such as whole wheat, high-gluten flour, teff, and oat. The second experiment isolated the role of population density by rearing beetles in different population sizes on the same flour diet. Adult-egg cannibalism rates were measured and analyzed, and the results indicate that stress type and cannibalistic behavior are correlated. The experiments suggest that a low-nutrient diet and high population density can lead to increased cannibalistic behavior in red flour beetles. These findings can help to shed light on the underlying mechanisms that drive this behavior and may have implications for understanding other organisms that exhibit similar behaviors.

### 2 - Exploring Spider Wasp (Hymenoptera: Pompilidae) Diversity in Cypress Grove Nature Park

Kelsey Crowley<sup>1</sup>, Jeremy Blaschke<sup>2</sup>

<sup>1</sup>*Union University, TN, <sup>2</sup>Union University, Jackson, TN*

Spider wasps in the family Pompilidae are stinging parasitoid wasps who utilize a great diversity of spiders as hosts. Because of complex tri-trophic interactions of parasitoid-host-host prey, the abundance and diversity of pompilids can offer useful information about the diversity and health of the ecosystem as a whole. The biodiversity present in the cypress swamp ecosystem of Cypress Grove Nature Park (CGNP) in Jackson, TN is largely unknown and the presence, abundance, and diversity of pompilids has never been investigated. To explore and begin to document the diversity of Pompilidae at CGNP, Malaise traps were set up in the park and samples were collected every 48 hours for 20 days. To identify pompilids, insects were first sorted by order, then the hymenopterans were sorted to family. Pompilidae were then further sorted to morphospecies. Representative specimens of each morphospecies were sent to the Canadian Centre for DNA Barcoding for genetic species identification. In total, 12,374 insects were collected and sorted. Of these, 56 were identified as pompilids and sorted to 29 morphospecies. Of these, six were positively identified to species based on >98% genetic similarity to pompilids within the Barcode of Life Database (BOLD) with confirmed species IDs. These included *Priocnemis germana* which parasitizes agelenid spiders and *Auplopus caerulescens* which has the unusual behavior of amputating the legs of its host. This is the first investigation of pompilid diversity at CGNP and therefore each of these species represents a new record for the park. The diversity of pompilids indicates a correlating diversity in their hosts (spiders) and reflects well on the insect biodiversity of CGNP as a whole. The low percentage of specimens identified accurately to species by DNA barcoding highlights the need for more comprehensive species records within BOLD and the importance of future research in pompilid systematics.

### 3 - Digital Reconstruction of the Bony Labyrinth of *Dipsalidictis krausei*: Anatomy and Functional Morphology

Katie Craun, Heather Ahrens

*High Point University, High Point, NC*

Oxyaenidae is a clade of archaic, carnivorous mammals known from the Paleogene of North America and Eurasia whose broader evolutionary relationships are poorly understood. The late Paleocene oxyaenid, *Dipsalidictis krausei*, is known from a particularly well-preserved specimen, which allows for the reconstruction of the bony labyrinth using high-resolution computed tomography (CT) data. The bony labyrinth serves as a proxy for the inner ear, a complex sensory organ responsible for hearing and balance. Thus, reconstruction of the bony labyrinth can provide insights into the hearing range and locomotor agility of extinct taxa, as well as serve as a source of phylogenetic data. Using Avizo Lite 9.4, we constructed the first digital endocast of the bony labyrinth of *D. krausei*. *D. krausei* possessed a highly coiled cochlea, large cochlear aspect ratio, and a secondary common crus. Among comparative Paleogene taxa, the overall morphology of *D. krausei* is similar to *Chriacus pelvidens*. We also determined the agility scores and hearing frequency range of *D. krausei* based on measurements of the semicircular canals, cochlea, and basilar membrane. The agility scores were lower than expected given the postcranial anatomy of *D. krausei*, either indicating that the agility scores were underestimated for *D. krausei* based on the mammalian regression or that methods used to measure body mass in oxyaenids should be reevaluated. The hearing frequency ranges calculated for *D. krausei* were similar to contemporaneous extinct taxa, as well as the extant aardvark and nine-banded armadillo. This information provides important first observations of the inner ear morphology of Oxyaenidae.

### 4 - Intrinsic and extrinsic factors predict local recruitment and natal dispersal distance in a resident passerine, the Eastern bluebird (*Sialia sialis*)

Sarah Hill, Lynn Siefferman

Appalachian State University, Boone, NC

Natal recruitment and dispersal distances can have important impacts on population dynamics and are often influenced by ecological and individual factors. I assessed the intrinsic and extrinsic determinants of local recruitment from a 3-year study of the breeding ecology of Eastern bluebirds (*Sialia sialis*) in the mountains of North Carolina. Predictors included: offspring sex and body condition, year and seasonality of nesting, and estimates of both habitat quality and nest site availability. I found that offspring were more likely to recruit and dispersed shorter distances when there was greater availability of nestboxes near their natal area. I also found that after fledging from territories with lower percentage of agricultural and grasslands and closer to forest edges, fledglings that weighed less for their body size were more likely to recruit, which may be explained by predation pressure soon after fledgling. Alternatively, larger bluebirds were more likely to recruit when fledged from sites with higher percent openness but tended to disperse further; this may be explained by competition for nesting sites rather than predation pressure. Overall, dispersal in this species is influenced jointly by intrinsic and extrinsic factors suggesting that individual quality, predation, and competition likely shape natal philopatry in resident passerines.

#### 5 - An open-source tool for conducting high-fidelity vibrational playbacks

Alana Evora<sup>1</sup>, Reginald B. Crocorth<sup>2</sup>, Jennifer A. Hamel<sup>1</sup>

<sup>1</sup>Elon University, Elon, NC, <sup>2</sup>University of Missouri, Columbia, MO

Animals communicate to exchange information about activities central to their survival and reproduction, like foraging and mating. Many animal species, ranging from invertebrates to elephants, communicate with vibrational signals, and recent research demonstrates that plants also detect and respond to substrate-borne vibrations. A playback experiment with substrate-borne stimuli is a simple and effective way to answer an array of questions about vibrationally-sensitive organisms. One challenge associated with such experiments is that the playback stimuli are filtered by both the playback equipment and the substrate. This filtering causes unwanted distortion to the stimulus received by the organism. A common solution to this challenge is to measure filtering by the substrate and compensate for it, such that when the stimulus is played through the experimental substrate, the amplitude spectrum of the recorded stimulus matches that of the original recording. One convenient tool for measuring and compensating for substrate filtering is a custom-written script in Matlab. Because proprietary software licenses impose economic barriers that can limit access to research, we have developed an analogous approach in Python, an open-source language. Here, we present an outline of the resulting Python script, which has three modules: (a) calculate substrate filtering and compensate for it, (b) calibrate playback amplitude, and (c) play back vibrational stimuli. We demonstrate its functionality, include sample code, and compare its output with that of the existing Matlab tool. The basic functions provided by this tool are a stepping stone towards increasing access to vibration research. Because it is open-source, we hope the script will be expanded upon by other researchers in the vibrational communication research community.

#### 6 - A Not So Silent Night: Anthropogenic Noise and its Impact on Bats and Insects

Juliet Spafford<sup>1,2</sup>, Emilie Patrick<sup>1,2</sup>

<sup>1</sup>Highlands Biological Station, Highlands, NC, <sup>2</sup>University of North Carolina at Chapel Hill Institute for the Environment, Chapel Hill, NC

With an increasingly urbanizing world, noise pollution has become an ever-pressing threat to wildlife reliant on vocal communication. As a bioindicator that relies on echolocation for their behavior, bats (Order Chiroptera) are especially susceptible to anthropogenic noise. However, little research has been conducted on the effects of such noise pollution on bats outside of the lab, as well as their common prey, insects. In this study, we measured the relationship between bat activity, insect abundance, and anthropogenic noise. At eight lakes and ponds in Western North Carolina, we monitored bat calls and insect presence while playing sonic and ultrasonic sound exemplars. We found a statistically significant correlation between bat activity and the sound exemplars, suggesting that anthropogenic noise, particularly ultrasonic sound, mask bats' communication. Across all our field sites, we collected 3218 insects of seven different orders, representing 4989 mg of biomass. Despite its limitations, our study is important in addressing the issue of anthropogenic noise pollution and its impacts on our environment and can hopefully be used in further research on this matter.

#### 7 - Predictive Species Distribution Modeling of Molluscan Agricultural Pests to Assess the Probability of Future Invasions in the United States in a Rapidly Changing Global Climate.

Kayla Hankins, Ali Baloochi, Austin Brenek, Austin Holmes, Christopher Randle

*Sam Houston State University, Huntsville, TX*

As a result of the explosive increase in the globalization of the world's economy, travel, and trade, the introduction of non-native invasive species, made either intentionally or accidentally, is a well-documented phenomenon. Industries such as the horticulture, pet, and live-food trades are all major culprits in the dispersal of non-native alien species around the globe. Invasive terrestrial gastropods pose a significant and understudied threat to US agriculture, native biodiversity, and public health. Thus, the objective of this research is to make use of publicly available occurrence data sourced from the Global Biodiversity Information Facility (GBIF) to generate a suite of predictive species distribution models, including the General Additive Model (GAM), Maximum Entropy (MaxEnt), Boosted Regression Trees (BRT), and a comprehensive ensemble model to better identify areas of potentially suitable habitat for high-risk invasive terrestrial mollusk species in the US under current climatic conditions as well as a variety of projected carbon emissions mitigation scenarios for the years 2050 and 2080. The results generated from this study will be used to better inform

quarantine measures, future threat assessments, long-term pest monitoring projects, and pest-management strategies to further aid in the prevention and early detection of the successful establishment of high-risk invasive terrestrial gastropods here in the US presently and in the future as our global climate continues to change.

#### 8 - Anthocyanins modulate feeding behavior and development in *Drosophila melanogaster*

Joshua Ekanem<sup>1</sup>, Morgen Story<sup>1</sup>, Zane Sobejana<sup>1</sup>, Isabella Allar<sup>2</sup>, Claire Welp<sup>2</sup>, Cecil Saunders<sup>2</sup>, Nicole Hughes<sup>1</sup>, Jackson Sparks<sup>1</sup> <sup>1</sup>*High*

*Point University, High Point, NC, 2Wake Forest University, Winston-Salem, NC*

Anthocyanins are water-soluble red or purple pigments in plants that act as visual cues to animals, protect against extreme temperatures, and serve as photosynthetic sunblock. Insects interact with anthocyanin-rich plants in many ways: feeding, egg-laying, sheltering, etc. While some studies have shown that anthocyanin-rich plant tissues affect insect feeding behaviors, comprehensive examinations of anthocyanin-insect interactions are needed. We use pure anthocyanins to test adult and larval chemosensory-dependent behaviors in *Drosophila melanogaster*. At least two common anthocyanins elicit robust appetitive responses in these flies. Further, anthocyanin-enriched diets increase the rate of development of multiple strains of flies. Our data suggests anthocyanins are important chemosensory cues affecting physiology in insects. Future experiments extend to genetic responses and non-model species.

#### 9 - New Wasp Records from the Ichneumonid Subfamily Cryptinae in Great Smoky Mountains National Park

Jake Ramgren

*Union University, Jackson, TN*

The subfamily Cryptinae is the most speciose subfamily of Ichneumonidae, with ~2800 species described in 274 diverse genera. Cryptinae wasps are mostly ectoparasitoids of lepidopteran cocoons and pupae, but some species also parasitize adult flies, hymenopterans, and beetles. Even though this diverse group is commonly encountered in a variety of ecosystems, the All-Taxa Biodiversity Inventory of Great Smoky Mountains National Park (GSMNP) has recorded 0 species so far. To begin correcting this disparity in recorded vs. actual species in the park, hundreds of ichneumonid wasps previously collected at Andrew's Bald from May 2001–July 2002 were sorted to morphospecies, identified to subfamily, and then 95 of these were sent to the Canadian Center for DNA Barcoding for genetic species identification. Of the barcoded specimens, eight were identified as members of the subfamily Cryptinae. Of these eight, three new park records were discovered: *Aritranis director*, *Cryptus albifrons*, and *Gambrus incubator*. *Aritranis director* is an invasive species from Europe that was collected in Quebec and Toronto in the 1930s and has since been found in the American Midwest. It is a parasitoid of lepidopterans. *Cryptus albifrons* is a parasitoid of known agriculture pests, such as *Helicoverpa zea* and *Grapholita molesta*. Three more specimens are potentially undescribed species from the genera *Trychosis*, *Xylophrurus*, and *Idiolispia*. This study adds three new park records for Cryptinae in GSMNP and demonstrates that DNA barcoding can accurately identify decades-old specimens that are difficult to identify based on morphology alone.

#### 10 - Multi-locus molecular phylogeny of the cricket-assassin wasps (Hymenoptera: Rhopalosomatidae)

Collette Truitt, Benjamin Scott, Jeremy Blaschke

*Union University, Jackson, TN*

Rhopalosomatidae (Hymenoptera: Vespoidea) are unusual wasps whose larvae develop as ectoparasites of crickets. Four extant genera are included in this understudied family: *Rhopalosoma*, *Paniscomima*, *Liosphex*, and *Olixon*. The elusive nature of these cricket-assassin wasps makes collection difficult and no genetically-informed phylogeny of the family exists. To construct the first molecular phylogeny of Rhopalosomatidae, 45 specimens representing 30 species and all four genera were gathered from collaborators and collections around the world. Genomic DNA was extracted and four genes (*POL2*, *EF-1 $\alpha$* , *COI*, and *VSP4B*) were amplified and sequenced. The diverse family Vespidae is currently considered sister to Rhopalosomatidae and two vespid species (*Dolichovespula* and *Polistes*) were included as outgroup taxa. The sequences were aligned and phylogenies were constructed using Maximum Likelihood and Bayesian analyses on each gene independently and on a concatenated dataset. The composite phylogeny confirms several longstanding morphological hypotheses including 1) the monophyly of all four rhopalosomatid genera, 2) the close affinity of *Rhopalosoma* and *Paniscomima* as sister taxa, and 3) the basal position of the distinct brachypterous form *Olixon*. The nuclear coding gene *VSP4B* was used here for the first time in rhopalosomatid phylogenetics. The effectiveness of *VSP4B* at accurately resolving deep nodes in the phylogeny mirrors previous work in the Vespidae and may indicate broad phylogenetic utility among the Hymenoptera.

#### 11 - Using DNA Barcoding to Identify High Priority Taxa (Hymenoptera: Ichneumonidae) in Great Smoky Mountains National Park

Emerie Landers, Victoria Seymour, Lydia Cordner, Scout Fluet, Hettie Peek, Jake Ramgren, Jake Phelps, Brayden Paulk, Jeremy Blaschke *Union University, Jackson, TN*

The All Taxa Biodiversity Inventory (ATBI) of Great Smoky Mountains National Park (GSMNP) seeks to document every species of living thing in the park. This effort is now over 20 years old, yet some taxa remain virtually untouched by taxonomists. Such “high priority” taxa include the

hyper-diverse parasitoid wasp family Ichneumonidae. Despite the positive and multifaceted effects ichneumonids have on their environment, such as efficiently limiting populations of agricultural pests and being markers for ecosystem health, only a small percentage of those collected in the park have been identified to species—due mostly to their complex morphology and overwhelming diversity. Recently, DNA barcoding has transformed biodiversity inventories, streamlining the process to be more efficient and accurate, and offering hope to identify high priority taxa awaiting taxonomic experts. To test the effectiveness of barcoding 20+ year-old specimens of Ichneumonidae and thereby document new species records in GSMNP, hundreds of wasp specimens from Andrew's Bald collected from April 2001–May 2002 were carefully sorted by undergraduate students into morphospecies. Ninety-five representative specimens were sent to the Canadian Center for DNA Barcoding where two sub-fragments of *COI* were amplified and assembled which increases the success rate of barcoding old samples. Of the 95 barcodes attempted, 84 were successful (>550bp). Of these, 46 were matched with a species name in the Barcode of Life Database (>98% sequence similarity). Among these successful barcodes were at least 30 new species records for the park. These exciting results contribute significantly to the ATBI by adding new park records for a high priority taxon (Ichneumonidae) and demonstrate the effectiveness of applying DNA barcoding to samples in long-term storage or those lacking immediate taxonomic expertise.

## 12 - Color and Habitat Preferences of Pollinators at Cypress Grove Nature Park

Elizabeth Shepherd

*Union University, Jackson, TN*

Pollinating insects fill many valuable roles in ecosystems by aiding in plant reproduction, maintaining biodiversity by connecting species, and meeting human food supply demands. Unfortunately, the abundance and diversity of pollinators are in decline due to habitat fragmentation, climate change, disease, and detrimental agricultural practices. Given their ecological importance and current vulnerability, gathering knowledge of local pollinator diversity is a high priority for conservationists. Consequently, the pollinator diversity within the unique lowland swamp ecosystem of Cypress Grove Nature Park (Jackson, TN) was investigated for the first time in the park's history. Insects were collected using 32 pan traps of various colors (yellow, white, blue, and red) established within a grid covering two different habitats (forest and meadow). In only ten days of sampling, 1,928 insects were collected representing at least 14 orders and 25 families. Specimens of Hymenoptera and Coleoptera were further sorted to morphospecies and the abundance and species richness of each color and habitat were examined. Overall, yellow pan traps captured the most insects (n=932), followed by white (n=407), blue (n=364), and red (n=223). Yellow also captured the most morphospecies (n=109), followed by white (n=48). However, red (n=38) collected more morphospecies than blue (n=29). More insects were collected in the meadow (n=1,251) than the forest (n=677). Conversely, the forest (n=102) captured more morphospecies than the meadow (n=94). Some specimens were found only in blue pan traps or the forest habitat. For example, the bees *Melissodes* sp. (n=2) and *Bombus impatiens* (n=1) and the butterfly *Lerema accius* (n=2) were captured only in blue traps and ichneumonoid and tephritis wasps were found primarily in the forest (n = 59) rather than the meadow (n=14). This demonstrates that using a variety of colors in diverse habitats can increase the number and diversity of species captured.

## 13 - Urban Forensic Entomology: The Case of the \$12,000,000 Ant Colony

Dr. Ed Mondor, BSc, MSc, PhD

*Georgia Southern University, Statesboro, GA*

Forensic entomology consists of three distinct fields: urban, stored-product pests, and medicolegal. As the founder and director of the only forensic entomology lab in Georgia, my lab focuses primarily on medicolegal cases, i.e., estimating the “time of death of human decedents using insect evidence, to assist in criminal investigations. Urban cases resulting in civil lawsuits, however, also occasionally arise. In this talk, I will discuss my role as a forensic entomologist in an urban case; a woman seriously injured from a falling tree limb in a residential neighborhood. I will describe the methodology used to determine the insect species involved, how those insects were related to the health of the tree, and how the accident then occurred. In sum, this talk will provide a better understanding of how a forensic entomologist can aid in court proceedings.

## 14 - A Multifactor Assessment for Determining the Impact of Anthropogenic Noise in Rural Areas across Upstate South Carolina

Katie Irsik, Dr. John Quinn

*Furman University, Greenville, SC*

Anthropogenic noise is underregulated across the United States, causing disruptions in natural soundscapes. The primary source of the anthropophony is generated by urban landscapes and traffic on roadways, both of which impact human health and the health of vocalizing wildlife, especially across rural and preserved landscapes. This study aims to create an accessible and easily replicable multifactor assessment of anthropogenic noise using a Geographic Information System (GIS) and parameters for field data collection. The assessment will determine a relative loudness threshold for the anthropophony across varying landscapes. The case study used to demonstrate the methodology found associations between the distance of a highway, the percent and density of tree cover, and the site's relative loudness (dBA). Trends in observed noise quality, landcover type/use, and road type/use are also seen in the data. The methods used in this study can be applied to statewide conservation plans, municipal habitat quality studies or could serve as the framework for starting noise regulations in rural areas. The factors discussed in this study could also provide insight into where noise-reducing infrastructure, like barriers, quieter machinery and sound absorbing materials should be built along roadways.

15 - Cattle pastures as carbon sinks: an analysis of tree carbon, beef production, and woody species diversity in temperate and tropical pastures.

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Forest restoration is the most effective natural climate solution, with the potential to sequester 37% of the carbon dioxide needed to reach the Paris climate mitigation goal. Cattle pastures offer an opportunity to increase global restoration, improve biodiversity, and maximize carbon storage through the adoption of management strategies that prioritize the incorporation of trees into pasturelands. Yet, the effect of increased trees on cattle production is not well-documented across biomes. Furthermore, the restoration potential of these landscapes as a byproduct of tree carbon remains to be studied. Therefore, the aims of this study were (i) evaluate the effect of increasing tree carbon (MgC/ha) on the pastoral stocking density (AU/ha), (ii) quantify woody species diversity ( $H'$ ) within pastures, and (iii) compare findings between farms in temperate and tropical ecosystems. The pastoral carbon storage in temperate ecosystems of Virginia and dry tropical ecosystems of Panama was estimated remotely and verified with in-situ plots. Woody plant species were quantified to determine diversity as a metric of ecological restoration potential within these systems. MgC/ha was higher in the tropical farms than the temperate farms ( $p = 0.006$ ), as was  $H'$  ( $p < 0.001$ ) and stocking density (AU/ha) ( $p = 0.015$ ). Farms in conservation programs had lower AU/ha than those with traditional management ( $p = 0.047$ ), but no significant differences in  $H'$  or MgC/ha. There was no relationship between stocking density and MgC/ha, which suggests pastures with more trees did not result in a decrease in beef production.  $H'$  was positively associated with increasing MgC/ha ( $p < 0.001$ ), regardless of region or management type. These findings demonstrate the potential for pastures to increase tree carbon and diversity without decreasing beef production. Moreover, such efforts support landscape restoration and offer potentially novel revenue streams through carbon credit programs.

16 - Species differences in dune building capacity

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Sand dunes are the first line of defense against coastal erosion from sea level rise and storms. Dunes are formed through the feedback between sediment, wind, and vegetation. It has been observed that different species create dunes with different size and shape, but species-specific mechanisms and speed of dune building is not well documented. The goal of this project is to quantify interactions between plant traits and dune development (e.g., shape and size) over time for dominant dune grasses. In Spring 2021 we planted 3 dune grass species (*Ammophila breviligulata*, *Uniola paniculata*, and *Spartina patens*) in recently deposited sediment on Hog Island, VA, an uninhabited barrier island. Virginia provides an optimum location to compare species as species distributions are overlapping in the region. We established 3 x 1 m plots ( $n = 5$ ) and planted 8 individuals per species in monospecific plots. Locations were chosen following known elevation for survival. Measurements of stem density, cover, height, accretion, and lateral expansion were taken in 2021 and 2022. After two growing seasons, all species accreted sand with no differences among species. Stem density varied seasonally and was highest in *Uniola*, but no effect on accretion was found. Percent cover increased in *Ammophila* and *Uniola* with sediment accumulation and *Uniola* plants grew taller. Lateral expansion was similar among species but increased >5-fold in one year. After two growing seasons, plant traits differ among species but have not altered sediment accretion. Future analysis will evaluate dune shape from digital elevation models and determine if species differences on dune growth rate and shape emerge over time.

17 - Variability & Elevation Drive Changes in Plant Community & Belowground Biomass in Coastal Dunes of the Outer Banks

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Under future climate change and sea level rise scenarios, Natural and Nature-Based Features (e.g. dunes) that protect coastal habitat and infrastructure will be exposed to increased wave energy and storm surge. Understanding how these forces will impact coastal dunes is necessary for their continued use as protective features. Coastal dunes develop through feedback between vegetation and sediment deposition, a process complicated by species-specific growth rates and responses to burial. Recent studies have documented above and belowground components of different plant functional types to be important for resisting erosion. Although dune building and erosion are known to be mediated by dune vegetation, the amount and distribution of plant belowground biomass within a dune represents a knowledge gap in coastal ecology and geomorphology. Our objectives were to quantify the belowground structure (e.g., plant roots, belowground stems and rhizomes) and aboveground composition of dunes across a range of management styles. We sampled belowground biomass across the dune profile at several sites representing multiple management histories in the Outer Banks, North Carolina, USA. Living belowground biomass was highly variable across sites and did not exhibit differences between managed and unmanaged dunes or among depths within 90 cm of the sediment surface. Plant community differences between management histories and among dune positions and sites occurred with larger sampling frequency. Elevation was a significant predictor of living belowground biomass, fine root surface area, soil organic matter content, living cover and species richness. These results underscore the importance of management and geomorphology on dune plant communities, with effects on species that may influence erosion resistance. Our findings will be incorporated into future numerical models used to predict dune response to sea-level rise and storms in order to better understand and manage dunes as natural protective features with climate change.

18 - Conservation of shrubland birds in working landscape

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With over 70% of the continental United States privately owned, conservation practices on private, working landscapes are important to achieve conservation goals. Shrub-scrub habitat is one most rapidly decreasing habitat types in eastern United States while also being one of the most important for avian populations. To identify critical thresholds in working lands, we conducted point counts and modeled occupancy of 10 different shrub bird species along a gradient of land use and land cover types. At a landscape scale, Eight species had higher occupancy probabilities with a greater percent of shrub cover within 1,000 meters. Specifically these eight species had a 100% occupancy probability with 5 percent of the landscape managed as shrub habitat. These results show the importance of working landscapes in conservation of avian species and the clear benefit of a small changes in landscape structure. More broadly, these results show the effectiveness of maintaining native habitats on working landscapes and potential ecological benefits of conservation-minded management of working lands.

19 - Using a collaborative research network to explore tree use by foraging sapsuckers (*Sphyrapicus* spp.), a keystone taxon, across diverse landscapes

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Research networks are valuable for exploring ecological questions at broad spatial scales and for engaging undergraduate students in hands-on scientific inquiry. This project uses a coordinated, network approach to investigate the characteristics of trees used by foraging sapsuckers (*Sphyrapicus* spp.), a genus of woodpeckers that primarily feeds on tree sap. Their feeding leaves behind rows or grids of small, round holes called sap wells, which can facilitate food resource access for other species. Project participants observed trees in their locality and recorded sap well data (e.g., abundance, location on the tree), tree data (e.g., species, diameter, bark type), and site attributes (e.g., urbanization index, ecoregion). Since the project was launched in January 2021 eight institutions have participated, including three that have used the iNaturalist app as an archiving database. Preliminary data analysis from two of these institutions indicates that the cardinal direction of sap wells was non-random, which may be caused by sapsuckers avoiding wind exposed tree faces or preferring sun exposed tree faces when feeding. Additionally, analysis of 181 tree observations from the North Carolina piedmont found that sap well occurrence did not significantly differ between native and non-native tree species, but among trees that had sap wells (n=111), abundance was significantly higher on non-native species compared to native species. A more comprehensive analysis of data from all participating institutions will enable us to examine these trends at a broader scale. Furthermore, this research network approach can be used to address macroecological questions, including the effects of urbanization on sap well occurrence and abundance, and the correlations between sap well abundance and the distributions of sap-well-using species, such as the rufous hummingbird (*Selasphorus rufus*).

20 - The potential for conservation grazing in coastal uplands of the northern Gulf of Mexico coast

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Along the Gulf of Mexico coast, natural resource managers struggle with managing coastal uplands due to front-end costs, prolonged maintenance, and habitat-specific ecological needs. Prescribed fire, mechanical removal of vegetation, and chemical treatments are commonly used to restore and maintain coastal uplands; however, there are challenges with the application of these tools (e.g., variable weather, urban development, etc.). A potentially less intrusive and a financially viable alternative is using livestock as a restoration and management tool. As part of a NOAA RESTORE Science Program Planning Grant, a survey was developed and distributed to natural resource managers with the goal of understanding the research needs, and logistical and environmental concerns related to using livestock for habitat management. Survey results show that >96% of respondents were interested in using livestock for habitat management, but <10% were aware of any information that could be used to inform grazing practices in coastal uplands. Generally small-sized cattle breeds (e.g., pineywoods) and goats were identified as the livestock with the most potential for environmental benefit and ease of containment. Items noted for further investigation included factors associated with implementation (e.g., which livestock type to use, grazing intensity, etc.), logistical considerations (e.g., fencing and rotational frequency), and the impacts of grazing on water quality, wildlife, vegetation, and livestock nutrition. Survey respondents overwhelmingly indicated that grazing would not be a standalone management practice and should be used in conjunction with other habitat management techniques (e.g., prescribed burns). This led the project team to develop two research objectives that need to be addressed regarding livestock grazing as a tool for coastal upland habitat management: 1) assessing the potential for livestock to spread and manage invasive and non-target species, and 2) determining the frequency and duration of livestock grazing needed to meet management goals across multiple habitat management scenarios.

21 - The Leftovers: A four year study of green ash (*Fraxinus pennsylvanica*) decline due to the emerald ash borer in a NC piedmont swamp forest

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The Emerald Ash Borer (EAB) is an invasive species to the central piedmont of North Carolina. Our goal was to measure the decline and patterns of change in a Green Ash (*Fraxinus pennsylvanica*) dominated swamp forest in the Catawba College Fred Stanback Jr. Ecological Preserve (FSJEP). In the fall of 2019 we established six, 200 m<sup>2</sup> plots using a nested plot design to measure changes in the canopy, sapling, and herbaceous vegetation.

We resampled the plots in fall 2020, 2021, and 2022. Our data indicated low diversity in the tree canopy, tree importance values in descending order, green ash, sweet gum (*Liquidambar styraciflua*), red maple (*Acer rubrum*), black willow (*Salix nigra*), American elm (*Ulmus americana*), and the American Sycamore (*Platanus occidentalis*). In Fall of 2019 green ash trees were visibly showing signs of EAB infection. In Fall 2019 no ash trees were standing dead, but by 2022, 78% of the green ash trees were dead. The importance value of the green ash canopy declined from 2019 (IV = 1.61) to 2022 (IV = 0.90). Canopy coverage declined strikingly from 73.1% to 5.78% from fall 2020 to fall 2022. Successional trends based on canopy mortality and sapling dynamics will be presented.

22 - Genetic divergence between common and marsh partridge peas (*Chamaecrista fasciculata*) reflects their distinct ecologies and histories

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The common Partridge Pea, *Chamaecrista fasciculata*, has been noted for its extensive intraspecific morphological and ecological variability. As a result of this variation, taxonomists have recognized variants as distinct species or varieties, but rarely have genetic data been considered to test the hypothesis that phenotypically variable taxa are evolutionarily distinct. In this study, we quantified genetic variation at 15 microsatellite loci in 11 populations from Virginia and Maryland to assess population genetic differentiation and structure of the widespread *C. fasciculata* var. *fasciculata* and the tidal freshwater endemic *C. fasciculata* var. *macroisperma*. We found lower allelic diversity and locus polymorphism in var. *macroisperma*, but populations of this variety harbored many more private alleles than those of var. *fasciculata*. Analysis of molecular variance indicated that 6% of the variation resides between the varieties, and Bayesian assignment analysis indicated strong support for two genetic clusters, which align with morphological identification of the varieties. Nevertheless, there also appears to be considerable admixture in all populations surveyed, with results suggesting multiple generations of hybrids and backcrossed individuals. Genetic bottlenecks were detected in some populations, which may reflect founder events from small numbers of seeds and the ephemeral nature of population persistence of *C. fasciculata*. Given the degree of genetic differentiation observed between the varieties, we suggest that they should continue to be recognized as distinct taxa and perhaps elevated to species-level. The infrequent occurrence of var. *macroisperma* in freshwater tidal wetlands combined with strong genetic differentiation of extant populations is of conservation concern for this taxon. To preserve genetic diversity of var. *macroisperma*, efforts should focus on maintaining all extant populations and preserving suitable habitat to allow for colonization of new populations.

23 - Ecological Niche Modeling for the Identification of Suitable Habitat of Dusky Arion (Arion subfuscus: Arionidae)

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*Arion subfuscus* (dusky arion) is one of the largest slugs of Arionidae and is a hermaphroditic, generalist feeder. This invasive species is native to northern and western Europe, especially the United Kingdom. However, it was introduced to North America via horticultural trade and can feed on

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The United States has the third highest agricultural output of any country, and exports far more food products than any other country. Global food security therefore depends heavily on agricultural practices in the United States. The Cooperative Agricultural Pest Survey (USDA-CAPS) has identified more than 30 priority pests of corn, cotton, grapes, small grains, and soybeans, most of them with broad distributions outside of the US. Ecological niche modeling (ENM) is a computational technique for predicting most suitable habitats for a species in a region. Niche models work by quantifying current distributions of species and deriving response functions that maximize correlation between species occurrence and environmental data, such as temperature, precipitation, soil type, or forest cover. These optimized functions can then be used to project the most probable sites in which those species may persist onto a different region. This may be a useful tool in predicting suitable habitat for future invasions of croplands. However, some of the assumptions underlying ENM are likely to be violated by invasive pests, namely the assumption of ecological equilibrium. In this study, we incorporate three tools, the General Additive Models, Maximum Entropy Models, and Boosted Regression Tree Models, to make predictions about habitat suitability of these pests in the US. Ensemble predictions consisting of AUC (Area-Under-the-receiveroperator-characteristic-Curve) weighted averages were analyzed to 1) develop a ranking of pests with the highest probability of finding suitable habitat in US croplands, 2) identify US regions most susceptible to new agricultural pests, and 3) to identify regions of the world from which pests and pathogens are most likely to enter the US.

25 - Predictive Distribution Modeling of Chrysodeixis chalcites (Tomato Looper)

stems and leaves of crops like corn, soybean, and wheat, the three most widely grown crops in the US. Occurrence points were obtained from the Global Biodiversity Information Facility, and sanitized by removing points outside of the native range, points without latitude and longitude, and duplicated points. Three ecological niche models, the Generalized Additive Model, Maximum Entropy, and Boosted Regression Trees were developed using 36 Climond environmental predictors, as well as elevation and human impact. Model prediction accuracy was assessed using Area Under the receiver operating Curve (AUC), and ensemble predictions of habitat suitability on the US were generated using AUC-weighted averages of model predictions. CLIMEX, a mechanistic model was also used to assess habitat suitability in the US.

#### 24 - The Good, the Bad, and the Ugly: Predictive niche modeling of pests and pathogens of major US crop commodities

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Nicholas Galle, Justin Williams, Christopher Randle

*Sam Houston State University, Huntsville, TX*

*Chrysodeixis chalcites* is an economically significant crop pest originating in Mediterranean Europe. As such, establishment in the United States could have severe economic repercussions. Recent surveys conducted in southern Ontario found increasing occurrences of *C. chalcites* on agricultural crops over successive years. Due to the proximity to the United States border, it is likely that *C. chalcites* has already invaded the United States and overwinters here. This poses a threat to American agriculture, namely soybean production, so it is important to determine where may be climatically suitable for *C. chalcites* establishment. Here we show that the eastern half of the contiguous United States is suitable for *C. chalcites* establishment, while the west coast is sparsely suitable. We reached this conclusion by using three mechanistic species distribution modeling techniques, boosted regression trees, Maximum Entropy, and generalized additive modeling, to predict most probable distribution by relating home-range occurrence and climatic conditions. From these models, we then generated an ensemble weighted by model performance. A mechanistic species distribution model, CLIMEX, was also used to predict where the species could occur based on climatic suitability. These results demonstrate that key areas of American agricultural output, such as the Mississippi River Valley and Midwest, are at risk for *C. chalcites* establishment. This study has implications for pest control and management strategies. Further work will compare the shift of most probable distribution under different climate change scenarios.

#### 26 - Estimates of online content reveal bias against amphibians and reptiles, especially those in decline

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In 2022, the International Union for the Conservation of Nature (IUCN) reported that over 40,000 species are at risk of extinction. Of those, about 3.2% are mammals, 3.3% are birds, 4.3% are reptiles, and 6.2% are amphibians. While mammals make up only a small portion of the total in-peril species, mammal-related papers comprised nearly 40% of the conservation publications over 15 years. Taxonomic biases are well documented in scientific literature, but it is unknown whether similar biases exist in the general public. As a proxy for public awareness, we considered the amount of internet content available on a species. We used Google hit-count estimates (HCEs)---the number of returned results a search engine provides for a query---to quantify the amount of available internet content for 2,163 species (classified as mammals, birds, reptiles, and amphibians). Then, we compared these quantities with various population trends and IUCN conservation statuses to identify potential biases. We found significantly more HCEs per species for mammals and birds than reptiles, and significantly more HCEs per species for reptiles than amphibians. Species with increasing populations returned larger HCEs than species with stable or decreasing population trends. Similarly, species of least concern returned higher HCEs than species listed as critically endangered, endangered, near threatened, or vulnerable. Our evidence suggests that in-peril amphibians and reptiles are likely underrepresented in the available internet content. While more research is needed to fully understand the relationship between a species' online presence and subsequent conservation actions, our results suggest that there is opportunity to enhance public awareness of these at-risk species by developing tailored online marketing campaigns.

#### 27 - Structural insights into the development of small-molecule inhibitors targeting the classical pathway of complement

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The complement system is a proteolytic cascade that, upon activation, act as the first line of innate immune defense against pathogens. Under normal homeostatic conditions, recognition of pathogen-associated molecular patterns (PAMPs) by complement leads to pathway activation, opsonization of bacterial cell surfaces, phagocyte recruitment, and bacteriolysis. *Borrelia* bacterial species, which traffic within the blood, have become models of complement inhibition through the expression of membrane-bound lipoproteins that our lab has previously shown to inhibit several different complement proteins. Using borrelial lipoproteins as a model of inhibition of the classical pathway of complement at the level of the initiating protease, C1r, we have developed a small-molecule inhibitor through a structure-guided approach for fragment-based drug discovery. While endogenous regulators are fine-tuned to control complement activation, dysregulation or aberrant activation is present as a driver and/or mediator of an ever-growing list autoimmune, inflammatory, and neurodegenerative pathologies. Using modes of inhibition similar to that of borrelial species, the optimization of a synthetic small-molecule compound may provide a promising therapeutic approach to classical pathway-mediated diseases.

We investigated the activity of a series of small-molecule compounds identified in a large-scale compound library screen and which were predicted to bind the C1r protease. Through several biofunctional assays, we identified two lead compounds that bind C1r and inhibit complement in a dose-dependent manner. Using molecular dynamics (MD) simulations, we were able to map out the favorable protein-ligand interactions and perform compound optimization through fragment replacement and synthesis. Newly synthesized analogs of our parent lead compound showed modest improvements in dose-dependent binding affinity and inhibition of the classical pathway, validating our structure-guided approach to fragment-based drug design. Additional routes of optimization will be explored to increase binding affinity and potency of our smallmolecule compound with the aim that it may provide a promising therapeutic approach to classical pathway-mediated diseases.

28 - The fitness consequences of genetic variation for *Trichogramma kaykai* infected with parthenogenesis-inducing *Wolbachia*.

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The Parthenogenesis-inducing (PI) form of *Wolbachia pipiensis* feminizes male arthropods and allows infected females to reproduce without mating (parthenogenesis). Many species are fixed for PI-*Wolbachia* and have lost the ability to reproduce sexually, resulting in clonal lineages dependent upon *Wolbachia* for reproductive survival. *Trichogramma kaykai*, a hymenopteran parasitoid native to the Mojave Desert, is one of the few species where PI-*Wolbachia* is not fixed, and infected females are still capable of reproducing sexually. We use this unique *T. kaykai/Wolbachia* symbiosis to investigate the fitness consequences of PI *Wolbachia* infection and the related loss of genetic variation associated with parthenogenesis. A curing protocol and mating scheme were developed with *T. kaykai* cultures that had been isolated as iso-female lineages for over 400 generations. Utilizing the retained sexual function of infected *T. kaykai* we were able to introduce genetic variation in both infected and cured experimental cultures to test two general hypotheses: 1) Genetic variation results in increased fitness, and 2) *Wolbachia* infection results in decreased fitness. Our preliminary results suggest there are significant costs associated with infection and absence of genetic recombination. We are currently testing these hypotheses with different genetic lines to assess genotypic effects on fitness. These results are discussed in terms of ecological conditions and evolutionary consequences for *Wolbachia*-infected populations.

29 - Alveolates (dinoflagellates, ciliates and apicomplexans) and Rhizarians are the most common microbial eukaryotes in temperate Appalachian karst caves

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The purpose of this study was to survey the eukaryotic microbiome of two karst caves in the Valley and Ridge physiographic region of the Appalachian Mountains. Caves are known to harbour eukaryotic microbes but their very low densities and small cell size make them difficult to collect and identify. Microeukaryotes were surveyed using two methodologies, filtering water and submerging glass microscope slides mounted in periphytometers in cave pools. The periphyton sampling yielded 13.5 times more unique amplicon sequence variants (ASVs) than filtered water. The most abundant protist supergroup was Alveolate with large proportions of the ASVs belonging to dinoflagellate, ciliate and apicomplexan clades. The next most abundant were Rhizarians followed by Stramenopiles (diatoms and chrysophytes) and Ameobozoans. Very few of the ASVs, 1.5%, matched curated protist sequences with greater than 99% identity and only 2.5% could be identified from surface plankton samples collected in the same region. The overall composition of the eukaryotic microbiome appears to be a combination of bacterial grazers and parasitic species that could possibly survive underground as well as cells, cysts and spores probably transported from the surface.

30 - Surveillance for antibiotic-resistant *E.coli* in wildlife feces from areas disturbed by agricultural and residential land uses in Appalachian Kentucky

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Microbial communities exposed to antimicrobials can select for antimicrobial resistance (AMR) genes via processes such as horizontal gene transfer. Increasing antibiotic pressures can enhance AMR gene expression in livestock, wildlife, and environmental flora, which may increase AMR among microbial communities. We aimed to assess correlations between land use types and AMR rates among *E.coli* in wildlife and livestock. *E.coli* was assessed due to their ubiquity in animals and waterways. Samples (n=51) from Kentucky roadkill, manure, and scat from roadways, farms, and forested areas were thrice swabbed for *E. coli* detection. Swabs were incubated in ColiGlow broth tubes containing no antibiotic, tetracycline, and ciprofloxacin. After incubation, tubes fluorescing under UV light were presumed *E. coli* positive and resistant (if from treated broth). Isolates obtained by streak plating on mTEC agar were then plated on general media for loading Microscan® urine panels. Isolates were analyzed for species identification and resistance to 25 antibiotics including ciprofloxacin and tetracycline. The percentage of AMR-positive isolates was positively correlated with agricultural land uses. 100% and 76% of isolates from high-intensity agricultural samples were tetracycline- and ciprofloxacin-resistant, respectively. These results corroborate other studies documenting AMR within agricultural microbial communities impacted by antibiotic use and manure spreading practices.

31 - Investigation of the effects of novel anti-biofilm compounds on biofilm production in *Staphylococcus aureus* Nor-A wild-type and mutant strains

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Quorum-sensing is a density-dependent intercellular communication mechanism utilized by bacteria to restrict specific gene expression to high cell-density conditions where collaboration is most beneficial. Once a bacterial population reaches a threshold, one intercellular response is the production of a surface-coating virulence factor called biofilm. Biofilms prevent a host's immune system from detecting and eradicating them. Recent studies indicate that efflux pumps may be involved in the process of biofilm production. These pumps are transmembrane transport proteins that extrude various substances, including toxins, quorum-sensing molecules, and biofilm components. The NorA efflux pump in *Staphylococcus aureus* is the most biofilm-related pump studied, as subsequent overexpressor (SA-1199B) and knockout (SA-k1758) strains have been developed to analyze the relationship between NorA activity levels and biofilm production. Known NorA efflux pump inhibitors (EPI), to date, cannot be used clinically due to their bactericidal and/or bacteriostatic effects, which significantly harm our normal flora. Thus, discovering novel compounds that act on the NorA efflux pump without bactericidal and bacteriostatic effects is essential for maintaining a positive symbiotic relationship with bacteria. NorA EPI structures were obtained through a literature search and were compared to the structures of 853 previously designed quorumsensing analogs. Three were chosen for further testing: Proline trans-3,5-Difluorocinnamic acid (Pro 61), Proline trans-3-nitrocinnamic acid (Pro 68), and Tyrosine trans-cinnamic acid (Tyr 16). Biofilm was stained with crystal violet and absorbances were recorded (550 nm). Disc diffusion and Ure dilution assays qualitatively studied the bacteriostatic and bactericidal effects of our drugs. The bacterial growth curve assay (600 nm) studied changes in bacterial growth patterns. Minimum inhibitory concentrations were determined ( $\mu\text{g/mL}$ ) for ideal biofilm inhibition. Pro 61, Pro 68, and Tyr 16 significantly reduced biofilm in SA-1199 and SA-1199B, but not in SA-k1758, which are similar effects of known NorA EPIs reported in the literature.

### 32 - Is Spider Silk Antibacterial? The Bacteria and Hygiene Aspects of Spider Webs

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Spider silk has been reported to have antibacterial properties and has been used historically as a first aid wrap. Spider webs were sampled and placed in broth, and after plating had bacterial growth. Direct contact of silk webs onto 3M Petrifilm also resulted in bacterial growth where the silk was in contact with the agar. Next Gen analysis of orb webs demonstrated over 300 bacterial OTU's (species). These results indicate that spider webs do represent a collection device of aerial bacteria, but at the same time that spider silk is not directly antibacterial. Spider webs can prevent the movement of water through the silk and this could be the reason the web was useful for first aid. While silk is probably not antibacterial, the preparation of silk for testing may be the reason for reports of antibacterial activity of spider webs.

### 33 - Cannabinoids Attenuate Inflammation Associated HIV-1 Reactivation

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As of 2021, there were 38.4 million people living with HIV-1 (PLWH), a retrovirus that infects, replicates, and establishes latency/persistence in both CD4+ T cells and macrophages. Despite the success of antiretroviral therapy (ART) in the management of HIV-1 infections, chronic inflammation persists in PLWH, thus contributing to the development of comorbidities. Cannabis (*Cannabis sativa*) is a substance that has been used to treat inflammatory disorders for millennia. While research shows 2-3 times higher use of cannabis in PLWH compared to the general population, our understanding of the potential role of cannabinoids in regulating HIV-1 associated inflammation and latency is not well understood. Here, utilizing pharmacological, biochemical and single-cell flow cytometry-based assays, we show that cannabinoids suppress PKC agonist mediated latency reversal in a CD4+ T cell model of HIV-1 latency through inhibition of the inflammatory mediator NF- $\kappa$ B. We demonstrate that the cannabinoid receptor 2 (CB2) agonist, GW 842166X, attenuates NF- $\kappa$ B activity in response to PKC agonists by suppressing both phosphorylation (S536) and acetylation (K310) of the NF- $\kappa$ B p65 subunit, post-translational modifications (PTMs) necessary for activation. Taken together, we observe a role of CB2 targeted signaling in the attenuation of HIV-1-associated inflammation and provide rationale for further study of this pathway for the development of therapeutic strategies designed to ameliorate chronic inflammation in PLWH.

### 34 - The historical distribution of an incipient invasive fern, *Cyrtomium fortunei*, in the eastern United States.

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*Cyrtomium fortunei*, is a native east Asia but has recently begun to be recognized as an invasive fern in the United States. Previous research has suggested that *C. fortunei* may have been cultivated in the United States since 1866, and according to herbarium data, the fern has been spreading, slowly at first throughout the eastern United States. As new records of *C. fortunei* continue to be documented, the distributional expansion appears to be up the mid-Atlantic and northeast US from the southeastern US. *Cyrtomium fortunei* demonstrates a range of continual morphological variation that can result in misidentifications of the species. It is suspected that *C. fortunei* might have a relation to the cut flower industry because in early collections it is reported as cultivated in yards and growing as an apparent adventive plant in cemeteries. Therefore, raising the questions of where was *C. fortunei*'s invasive origin and its relation to the cut flower industry? 302 total records of data were collected on the occurrence of *C. fortunei*

in the Southeast using online herbarium data from SERNEC that included 25 herbaria, iNaturalist, the Smithsonian National Museum of Natural History, and the New York Botanical Garden. That data was then used to visualize the earliest to most recent occurrences of *C. fortunei* in the United States to determine its potential origin and spread throughout the eastern US.

35 - A Vascular Flora of the Walls of Jericho in Jackson County, Alabama

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The Walls of Jericho (WOJ) is a 5,062-ha tract of land purchased in 2004 by the Alabama Chapter of The Nature Conservancy through the Forever Wild Land Trust Fund and incorporated into the greater Skyline Wildlife Management area. The WOJ occurs on the southern edge of the Cumberland Plateau at the transition to the Plateau Escarpment, an area characterized by sandstone caps transitioning into jagged karst limestone valleys. According to the species-area curve model developed for the Cumberland Plateau (Huskins and Shaw, 2010), we estimated the WOJ to contain ~765 species of vascular plants. A total of 20 trips were made during the initial field season in which we documented ~410 species, representing 94 families and 198 genera. Here we provide an update and summarize our findings from the initial field season and report ongoing efforts to more fully document this protected area.

36 - *Diervilla* (bush-honeysuckle, Caprifoliaceae) contains three species based on morphology and RAD sequence analysis but shows admixture throughout its range

Katherine Mathews

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While three species of *Diervilla* are currently accepted in most floras and checklists, confusion among what constitutes the species still exists and has led to misidentification of herbarium specimens, uncertainty in field identification, and lumping of quantitative character measurements between species. To clarify the taxonomy of *Diervilla*, I collected and obtained plants from throughout its range in eastern North America, examined and mapped herbarium specimens, reviewed and scored morphological characters, and sought new characters to aid in species identification and delimitation. I generated a ddRAD-Seq dataset from field-collected and herbarium leaf samples since single-gene sequencing was insufficient to resolve a *Diervilla* phylogeny, signaling either lack of species differentiation or post-speciation admixture. As a result of these new data and analyses, the three currently accepted species are confirmed, quantitative character ranges are clarified, and new inflorescence and vestiture characters are described to help distinguish among them. Hybridization may have occurred among all three species, possibly following Pleistocene range contraction and expansion, as shown by Structure admixture plots.

37 - New evidence about Michaux's plant introductions

Charlie Williams

*AMIS, the Michaux Society, Charlotte, NC*

On behalf of a succession of French governments, French botanist André Michaux (1746-1802) traveled and explored throughout eastern North America between 1785-1796. He was the first European-trained botanist to visit many areas and is remembered because of his hundreds of names and descriptions of new species in his *Flora Boreali-Americana*, the first flora of North America. He is also remembered because he is credited with introducing several favorite garden plants, including *Camellia japonica* through his Charleston, SC garden. Documentary evidence supporting some of his plant introductions and evidence about on how they were accomplished was uncovered during the research for *André Michaux in North America Journals and Letters, 1795-1797*, Univ. of Alabama Press, 2020.

38 - Quantifying the National Park Herbarium Specimens at the University of Tennessee at Chattanooga Herbarium (UCHT).

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*University of Tennessee at Chattanooga, Chattanooga, TN*

Tennessee is home to 13 herbaria, of which 12 are associated with universities and one is a federal herbarium in the Great Smoky Mountains National Park (GSMNP). While GSMNP is somewhat unique among national parks in having a herbarium, many others have herbarium specimens spread across untold numbers of herbaria, making direct use by National Park Service (NPS) biologists or others difficult prior to specimen digitization. Through use of the University of Tennessee at Chattanooga Herbarium (UCHT), it became apparent to curators that it contained a relatively high number of specimens collected from a number of different national parks. Most of these specimens were collected in the 1950s/1980s. The purpose of this project was to: locate all of the specimens within UCHT that were collected within the boundary of a national park or battlefield (not national forests); database them in regard to the history of any scientific name changes, the park in which they were collected, and NPS barcodes that are separate from UCHT barcodes/catalog numbers; segregate them into separate brown-stripe folders; record any damage on a scale from 0-3; and then analyze trends among the specimens collected within these national sites. To date we are approximately 50% through the 50k-specimen herbarium. We have identified ~1000 specimens collected from within 23 national parks of three countries (US, Canada, and Kenya) and 16 US

states. The national site with the highest number of specimens is the Chickamauga and Chattanooga National Military Park. Specimen quality is generally high with 87% in excellent condition with no signs of degradation and 13% with moderate damage, and four specimens with significant damage.

#### 39 - Salamander Movement Propensity Resist Effects Of Supraseasonal Drought

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*University of Kentucky, Lexington, KY*

Movement can act as an effective strategy used by amphibians to avoid detrimental environmental conditions, particularly drought. However, due to the unpredictable nature of droughts, evaluating the patterns and consequences of movement has rarely been investigated. In 2007-2008, the southeastern United States experienced a supraseasonal drought that resulted in 110-yr low flow levels amongst the first-order streams. In this study, 61 months of mark-recapture data collected from one first-order stream were used to examine the effects of drought on the movement frequency distribution, survival, and growth rates of adult *Desmognathus fuscus* (Northern Dusky Salamander). We hypothesized that salamanders would demonstrate a higher propensity to move during supraseasonal drought conditions and that moving salamanders would experience higher survival and growth rates. We found that salamanders were more likely to move immediately after the drought compared to the pre-drought and drought conditions. Although movement frequency was low during the drought, survival was higher for individuals who moved during drought conditions in comparison to individuals who remained in their original capture location. Although our model did not detect a trend, salamanders experienced slightly higher growth in the post-drought conditions compared to drought and pre-drought conditions. In addition, during the postdrought, salamanders that moved had slightly higher growth rates compared to salamanders who remained in their original capture location. Our results suggest that adult salamanders were potentially displaying an adaptive movement strategy to resist drought conditions by moving away from affected (i.e., dry) areas within the study stream. In addition, movement was likely utilized to access replenished resources in other areas after the severe effects of the drought ended. Therefore, both in-stream and riparian barriers that impede movement may inhibit resilience of streams amphibians during severe droughts.

#### 40 - Investigating summer occupancy of small terrestrial salamanders (genus *Plethodon*) in the Southern Appalachian Mountains

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*Appalachian State University, Boone, NC*

As habitats become more fragmented and amphibian assemblages change, understanding what environmental factors influence the detection and occupancy of *Plethodon* salamanders is becoming increasingly important. Previous research has found that salamander habitat requirements may be restricted to specific environmental parameters. We investigate the regional spatial occurrence of *Plethodon cinereus* and *P. richmondi* in the southern Appalachian Mountains, on 40 plots surveyed three times during the months of June, July, and August of 2021. We evaluated 17 covariates to explain both occupancy probability and detection probability for each species. Detection for *P. cinereus* was best explained by cm of precipitation. *P. richmondi* detection was best explained by the interaction of relative humidity and days since last rain. Occupancy probability for *P. cinereus* ( $\text{psi}=.46 \pm 0.1$ ) was best explained by latitude while longitude best explained *P. richmondi* occupancy ( $\text{psi}=0.41 \pm 0.08$ ). Ultimately, detection and occupancy of both species appear to be influenced by moisture levels. This data will serve as the foundation for future models across seasons and to understand how these species coexist within terrestrial ecosystems.

#### 41 - Quantifying Habitat Parameters for the Southern Appalachian endemic, *Plethodon welleri*

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*Plethodon welleri* is a small-bodied salamander species that is endemic to select mountain tops in the Southern Appalachian Mountains and considered threatened across its entire range. Initial descriptions indicated that this species was a high-elevation, spruce-fir specialist only found above 1500m. However, recent observations have documented populations outside of this range. The goal of this study is to assess environmental parameters for habitat preferences and build a comprehensive dataset on species detection and occurrence of *P. welleri*. Preliminary analysis from our 2021 field season showed population occurrences across an elevation gradient of 700-1600m and in a variety of forest structures. In 2022, sites were expanded across the North Carolina and Tennessee portion of *P. welleri* range, and population and environmental data was collected March-November. This data is being used to model occupancy for this species throughout 3 active seasons. We expect that the most influential environmental covariates will change by season, as lower temps allow populations to emerge with less threat of desiccation. This study will provide foundational data for this species and other salamander species in this region. Having access to baseline knowledge for threatened species is necessary for the development of effective conservation strategies and comprehensive management plans.

#### 42 - Developing a Predictive Geospatial Habitat Model for a Rare Species of Salamander in TN: A Case Study for the Green Salamander (*Aneides aeneus*).

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University of Tennessee at Chattanooga, Chattanooga, TN

Green Salamanders (*Aneides aeneus*) are a secretive and cryptic species that have low detection probabilities which is primarily driven by their occupation of arboreal habitats. This aspect of their life-history makes their populations not well studied throughout much of their range. Our foci are four-fold and include: 1) to locate potential populations, 2) to better recognize habitat parameters, 3) to identify suitable habitat, and 4) to gain insight for future studies on their distribution. Incorporating geospatial tools is a critical yet necessary step in developing predictive models. Such models can aid in determining prime habitat while offering insight on their status. MaxEnt is a geospatial software that can be used for modeling habitat suitability for a variety of species. In short, it requires data on the species occurrence in order to predict suitable habitat. MaxEnt's versatility creates an optimum distribution research application for developing predictive geospatial models for rare salamander species. Our baseline data was provided by the Tennessee Wildlife Resources Agency (TWRA) so that predictive geospatial models for *A. aeneus* could be generated. After processing the model, we randomized points to focus on natural areas and this yielded 87 potential study sites; and, of those, 49 were surveyed. Each location had the landscape and habitat structure assessed and we searched for the target species. Of the 49 sites, we found 46 to have suitable habitat in the immediate proximity; and of these 18 had confirmed presence for the target species representing 39 individuals. *A. aeneus* is considered by TWRA to be an uncommon species, and its management benefits greatly by the incorporation of predictive geospatial models in order to avoid extirpation. We plan to continue our validation efforts to further improve the utility of this model for this species and its associated stakeholders.

#### 43 - Interactive Effects of Hatching Variability and Experimental Warming on Larval Wood Frogs (*Rana sylvatica*)

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Climate change is an anthropogenic-induced shift to warmer average temperatures globally. As temperature increases, so does variability in precipitation, which directly affects the timing of phenological events in many taxa. In response many amphibians have begun breeding earlier. Similarly, many larval amphibians experience faster growth in response to higher temperatures and faster drying of temporary ponds. Consequences of earlier breeding and increasing temperatures for larval amphibians are increased mortality due to frost exposure and reductions in size and fitness at metamorphosis. To measure the interactive effects of hatching phenology and temperature on larval wood frogs (*Rana sylvatica*), we performed a mesocosm experiment. Phenology treatments were 14 days apart and temperature treatments were either ambient or heated at +4.5°C above ambient. Tadpole growth rate was faster in heated and early hatching treatments, however, tadpoles in later treatments showed compensatory growth by day 65. Mean time to metamorphosis was shortest in late treatments. Both temperature and hatching phenology had a synergistic effect on time to metamorphosis with mean size at metamorphosis being smallest in heated treatments. Mean survival was highest in heated treatments. In conclusion, larval wood frogs experience faster growth in response to higher water temperatures; however, there is a tradeoff in size. Wood frogs from our populations appear to be highly plastic in response to environmental change, however many anuran species may lack that adaptive response. Thus, it is imperative more research be done to determine how additional amphibian populations might respond to both large and finescale anthropogenic changes.

#### 44 - Evaluation of Nutritional and Health Status in Captive Eastern Indigo Snakes (*Drymarchon couperi*) in Response to Formulated Sausage Diet

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The federally threatened Eastern Indigo Snake (EIS; *Drymarchon couperi*) is an active ophiophagus snake native to the southeastern United States. The Orianne Society built the Orianne Center for Indigo Conservation (OCIC) to establish a captive breeding and head-start program in collaboration with other conservation-focused organizations. EIS are housed in outdoor or indoor enclosures and given whole prey diets rotating between domestic rodents, chicks, and freshwater fish. While outdoor enclosures alleviated a drop in female fecundity, which is associated with indoor enclosures, there are still concerns regarding the occurrence of dystocia, colloquially referred to as eggbinding, in the OCIC's colony. A retrospective study by Bogan et al. (2021) identified virgin breeding as one contributing factor but recognized other vitamins and minerals could contribute to the health of gravid females. Additionally, studies on elephants and swine indicate that the nutritional status of an animal could correlate with reproductive health and output. As such, we are evaluating the blood chemistry and circulating plasma levels of fat-soluble vitamins (D,E) and minerals of EIS consuming mixed whole prey, whole prey sausages, or the experimental sausage that replicates the nutritional profile of free-range EIS diets over

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The phenomenon of woody plant encroachment into grasslands has rapidly increased in coastal areas in the southeastern US in the last few decades. Previous studies at the Virginia Coast Reserve, a Long-Term Ecological Research site, show that native shrub, *Morella cerifera* is expanding into grasslands due to warming temperatures. It has been observed that shrubs establish in grassland and not bare sand, however it is unknown if there is a level of grass cover that is beneficial for *M. cerifera* establishment. We hypothesize that low grass density ( $\leq 70\%$  cover) will change the microenvironment and increase the establishment of *M. cerifera* seedlings, whereas high grass density

the course of one year. Snakes have responded well to the experimental sausage and differences in vitamin E levels and other blood chemistry parameters have been observed between whole prey and sausage groups.

45 - Bog turtle (*Glyptemys muhlenbergii*) demography and movement in Shady Valley, Tennessee

Timothy Calhoun

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The bog turtle (*Glyptemys muhlenbergii*) is a federally threatened freshwater turtle species found primarily in wetlands within the Appalachians. There is only one known natural meta-population of these turtles in Tennessee, a handful of wetlands within one valley in northeast Tennessee. In recent decades the Nature Conservancy has worked to restore and expand these wetlands. The area is believed to have approximately 40 bog turtles in its endemic population, although there has not been a recent intensive capturing effort to estimate the size of the population. Last summer (May – August 2022), I conducted capture-mark-recapture surveys at two of these wetlands to determine the population density and demography after the restoration efforts. I used live traps, camera traps, and random walk probing surveys to capture individuals. I also attached radio transmitters to six individual turtles to assess movement through the wetland using a passive telemetry grid of receiver nodes installed at one of the wetlands. I used a spatially explicit capture-recapture modeling approach with package SECR in R to assess population density. My population estimates from this model were consistent with the expected population size. I also found a diverse age structure among captured individuals ranging from 2-yearold juveniles to decades-old adults, as well as recorded movement events into the new restored wetland habitat.

46 - High grass density aids in the survival of *Morella cerifera* on Hog Island, VA

Natasha Woods<sup>1,2</sup>, Ayleen Mexquititla<sup>1</sup>, Julie Zinnert<sup>2</sup>

(>95% cover) will impede establishment. To address the impact of grass density on *M. cerifera* seedling establishment 3m x 3m observational plots (n = 10) were set up and monitored for natural occurrence of *M. cerifera* seedlings with two levels: high grass density (95-99% grass cover) and low-density grass plots (40-70% grass cover). Within these observational plots the following biotic and abiotic measurements quantified including seedling number, species richness, temperature, chlorides, and light availability. Experimental plots were established adjacent to each of the observational plots (n=10) and five seedlings were transplanted per plot. After four weeks seedling survival was quantified. Results show that there were no naturally occurring *M. cerifera* seedlings in the high-density observational plots; however, after four weeks there was 88% survival in high density plots compared to 80% survival from the low-density plots during the summer. High grass density appears to be important for the survival of *M. cerifera* seedlings during the summer months but may limit seedling establishment.

47 - Rock Climbing Effects on Lichen Species Richness and Diversity on Boulders

Tyler Pesce

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Cliff habitats are diverse but understudied due to inaccessibility. Cliff vegetation is often left out of biodiversity surveys and management plans. In the last 30 years, accessibility and concern about these cliff habitats has increased partly due to the rising popularity of rock climbing. However, research about the impacts of climbing on vegetation is limited, especially on boulders. This research fills this gap by studying rock-climbing effects on boulder vegetation, primarily lichen. Species richness, Shannon's diversity, and percent cover were measured before being climbed and after being climbing 50 times. There was a significant decrease in lichen coverage before and after climbing, however there was no difference in species richness. Shannon's diversity increased slightly from  $H' = 1.99$  to  $H' = 2.05$  with the presence of climbing. However, this may be due to increased evenness in climbed plots. Climbing should be allowed on Bailey Mountain Preserve as long as management guidelines are followed. This includes yearly vegetation surveys on the boulders, leaving some of the vegetation on top of the boulder intact, sustainable trails, and recommending that climbers only remove vegetation on the holds they are using.

48 - Description of *Tsuga caroliniana* Engelm. communities in the southern Appalachian Mountains of western North Carolina, USA

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Southern Appalachian forests contain unique microhabitats for ground layer vegetation species due to their varying topography and geologic history, among other characteristics. There are several hundred state-listed and many federally listed species of concern in North Carolina, many residing in the Southern Appalachian region. Ground layer species play an important role in ecosystem functions such as seedling regeneration, nutrient cycling and wildlife resources. Consequently, the retention or restoration of species composition and diversity of the ground layer vegetation community has been a growing concern for researchers. Silvicultural management techniques developed to promote oak regeneration are often used

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Carolina hemlock (*Tsuga caroliniana*) (CH) is endemic to the southern Appalachian Mountains. Extant populations are under threat from the spread of introduced hemlock woolly adelgid (*Adelges tsugae*) and information necessary to inform CH conservation and future restoration efforts is limited. In this study, we characterized the structural and compositional characteristics of five CH communities to achieve the following objectives: (1) Assess similarities in structure and composition of spatially disjunct CH communities and identify how composition varies in response to physiography and other ecological attributes; (2) Examine recruitment patterns of CH over time; and (3) Quantify how the importance of CH and species diversity and richness vary within and across the overstory and regeneration. At all but one site, CH possessed the greatest importance value, and the average age of CH ranged from 48 to 65 years. Across the five sites, the CH overstory was dominated by two age classes: 41 – 60 and 61 – 80 years, suggesting current CH overstories were facilitate, in part, by the loss of American chestnut (*Castanea dentata*). Overstory (stems ≥5 cm DBH) species composition was distinct at each site, while that of the regeneration layer (stems <5 cm DBH) layer was similar across sites. Relative density of CH in the regeneration layer was low, averaging only 3% of stems. As mortality of overstory CH continues due to hemlock woolly adelgid, maple and oak species that currently dominate the regeneration layer are poised to recruit into the overstory, suggesting that underplanting may be needed to ensure CH remains a component of these landscapes over time. Coupled with previously published descriptions of CH communities, the data presented here can be used by managers to guide restoration activities that restore and maintain the variability in structure and composition of these unique and endemic communities across the landscape.

#### 49 - Ground Layer Vegetation Response to Oak Regeneration Treatments in Southern Appalachian Forests

Brandy Benz<sup>1,2</sup>, Jodi Forrester<sup>2</sup>, Tara Keyser<sup>1</sup>

resistant lifeforms (SHRUBS, VINES) in the shelterwood/fire and repeated prescribed fire treatments. We are testing the effects of treatments on repeated measurements of ground-layer species cover and biomass over a decade following initial treatment establishment. We use multivariate analyses to evaluate the treatment versus microhabitat effect on species cover and to evaluate long-term trends in successional turnover in response to the silvicultural treatments.

#### 50 - Thistle come as no surprise, invasives exploit native pollinators: comparing the reproductive success of an invasive and native thistle in the subtribe Carduinae (Asteraceae)

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Invasive plants often have higher reproductive outputs than their native counterparts conferring a competitive advantage. Pollinator declines may exacerbate disparities in reproductive output by negatively impacting native plants more than their generalist invasive counterparts.

To determine how reproductive success varies with pollinator community, we conducted a pollinator exclusion experiment for two functionally similar species: an invasive (*Carduus nutans*) and native thistle (*Cirsium discolor*) in Clarksville, TN. We manipulated pollinator access to flowers by completely (screened) or partially excluding (nets that prevent large pollinators) pollinators. We evaluated pollinator visitation rates and composition influenced reproductive success.

No pollinators visited screened flowers of either plant species, so complete pollinator exclusion resulted in a higher probability of total reproductive failure (screened: 54.8%; unscreened: 6.5%). When flowers produced at least one viable seed, any level of pollinator exclusion resulted in lower germination success relative to the control. The invasive *C. nutans* attracted more pollinators, pollinators spent more time on the flowers, and flowers produced more viable seeds, resulting in overall greater reproductive success than native *C. discolor*. More taxonomic groups (2.4 mean) visited control flowers (no pollinator exclusion) than netted flowers (1.5 mean), and visitors spent more time on control flowers (control: 11.9 mean minutes; netted: 3.6 mean minutes); however, species richness did not vary between the native and invasive thistles. *C. nutans* also achieved a greater incidence of autonomous self-pollination (screened invasive: 61.1%; screened native: 23.1%), suggesting that factors besides pollination biology determine differences in reproductive success between species. Higher levels of self-pollination may allow the invasive *C. nutans* to easily establish new populations from low numbers of propagules. Our study provides evidence that invasive *C. nutans* enjoys greater reproductive success while attracting a different pollinator community than native *C. discolor*.

#### 51 - Ecosystems in Transition: An Assessment of Plant Community Change in Pond Pine Pocosins in Coastal North Carolina

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in Southern Appalachian forests. One such study is the “Regional Oak Study” being conducted by the US Forest Service in the Cold Mountain Game Lands of western North Carolina. It is important to consider the consequences that species-targeted management techniques can have on the growth, survival, and diversity of the ground layer vegetation community. The objective of this study is to compare how silvicultural management techniques, such as repeated prescribed fire, oak shelterwood thinning with herbicide, or shelterwood harvesting followed by prescribe fire alter the productivity and diversity of the ground-layer vegetation community. We hypothesize overstory reduction and/or prescribed fire treatments will promote greater biomass production by increasing available light following canopy reduction and/or fuels reduction post-fire. We further hypothesize that species composition will shift from more shade-tolerant, fire-sensitive lifeforms (HERBS, FERNS) to more shade-intolerant, fire-

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Sea level rise, increased intensity and frequency of hurricanes, and changing climatic regimes are threatening forested ecosystems along the central Atlantic coast of North America, putting these forests at risk to extreme degradation. This rapid degradation has led to the formation of "ghost forests"—recently transitioned shrub and grass marshes with still-standing dead trees overhead as plant communities change over the span of just a few years. As such, researchers find it difficult to maintain current estimates of community composition, stand structure, and standing carbon stores given the incredibly rapid rate of transition within these ecosystems. This study resampled historic plots established by previous research and established new permanent plots for future study with the intent to assess community composition and standing carbon stores. Thirteen plots focused on pond pine pocosins in the northern area of the Alligator River National Wildlife Refuge in Dare county, North Carolina, were sampled, three of which were sampled in 2003 and five of which were sampled in 2015. Community classification within these plots was done with the Guide to the Classification of the Natural Communities of North Carolina (4th Approximation). Given that these coastal forest conditions are expected to further degrade as climate change continues to impact the globe, this study allows for future research within these permanently marked plots and intends for more regular assessment of these ecosystems in transition. Sequential data from these ecosystems could then potentially be used to predict the trajectory of other coastal forests at risk to previously mentioned hydrologic stress and subsequent transition. Such serial data would be of value to land managers, both public and private, as they contend with climate change and how it will impact management decisions mitigating such transition.

#### 52 - Trait Variation of Highbush Blueberry Hybrids and Progenitors across sites in Virginia

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The evolutionary importance of hybridization has long been debated among plant scientists and was only recently accepted as common and influential throughout the plant phylogeny. Controversy surrounding natural hybridization is primarily due to a lack of supporting evidence caused by difficulty studying hybridization in natural settings. The process of identifying hybrids from morphological traits is complicated and their taxonomic distinction is commonly questioned. Because hybrids and their progenitor species are an exception to the commonly accepted species rule of reproductive isolation, additional evidence is necessary to support the taxonomic distinction of all three taxa. We will showcase the challenges of studying hybrids in a natural system by investigating a proposed hybrid highbush blueberry, *Vaccinium x Marianum*, and its progenitor species, *Vaccinium fuscum* and *Vaccinium formosum*, across five environmentally variable sites in the Coastal Plain of Virginia. Various functional traits were collected from the taxa at every site. Presence of the three taxa varied at each site, though *V. x Marianum* was often more abundant. Morphologically, *V. x Marianum* exhibits different combinations of traits that are intermediate between the parent traits. Functional traits, including plant maximum height, specific leaf area, leaf dry matter content, leaf thickness, and leaf area, were analyzed using two-way ANOVA tests with taxa and collection location as factors. Collection location has a significant effect on all functional traits, while taxon only has a significant effect on leaf thickness. This may indicate little difference between *V. x Marianum* and its progenitors in their functional roles, and that environmental factors may be more indicative of functional traits than taxon, reflecting context-dependency. Additional evidence, e.g., from genetic markers, may be necessary to support the taxonomic distinction of *V. x Marianum*.

#### 53 - Mycorrhizal Associations Found With the Native Venus Flytrap

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*Dionea muscipula*, a plant commonly known as the Venus Flytrap, is found endemic to southeastern North and northeastern South Carolina. Several populations of native Venus Flytrap's are known to exist in the nutrient poor peat bog conditions in and around Lewis Ocean Bay Heritage Preserve (LOBHP) in northeastern South Carolina. Growing development in the Myrtle Beach area has caused loss of habitat in and around LOBHP impacting survival of the native Venus Flytrap. Conservation efforts have focused on relocating native Venus Flytraps into adjacent habitat. This is only a temporary solution and does not account for aspects of soil microbiome that may strongly influence Venus Flytrap growth, development, and survival. Fungal composition found in wetland soils are distributed in intermittent spatial areas thus may be an important driver for soil microbiome selectivity to native plant communities. It is presumed that Venus Flytrap's do not require mycorrhizal associations due to evolution of the leaf-trap for increased nutrient acquisition from otherwise, nutrient poor soils. However, related carnivorous plants, such as the sundew, were shown to form mycorrhizal associations. The presence of mycorrhizae in Venus Flytrap roots would suggest that conservation efforts must account for soil microbiome prior to re-location of plants. Specificity of mycorrhizae to LOBHP soils may further impact conservation practices. We investigated root structure and surrounding soil of native Venus Flytrap roots for evidence of mycorrhizae associations and identification of mycorrhizal species in the soil microbiome. Findings support that Venus Flytraps form arbuscular mycorrhizal associations. These associations may be influenced by prescribed burns that increase nutrient availability in the LOBHP soils.

#### 54 - Stuck in a rut: habitat characterization of critically imperiled Ludwigia ravenii (Onagraceae)

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The critically imperiled wetland obligate *Ludwigia ravenii* Peng (S1G1) is broadly known from wet roadside ditches, ruts, and pine savannas. Extant occurrences are currently limited to 22 populations across its strictly Coastal Plain range. Detailed habitat requirements are poorly understood, presenting a knowledge deficit that must be addressed for successful conservation efforts. The objectives of this research were to quantitatively assess habitat characteristics for utility in predicting occurrence, identifying suitable habitat for reintroduction, and delineating critical habitat. To assist in distinguishing habitat features, a comparative study was designed to include two common congeners with overlapping ranges: *L. alternifolia* and *L. pilosa*. Habitat variables (vegetative composition, ditch physical features; dimensions, slope, aspect, soil characteristics, and canopy cover) were recorded for 108 plots across the current range of *Ludwigia ravenii*. To describe physical habitat, bisecting transects, and one square meter quadrats were established, centered on target stems of *Ludwigia*. Preliminary NMDS results suggest differentiation in vegetative composition between occurrences of *L. ravenii* and focal congeners, driven by features such as pH and elevation. *Ludwigia ravenii* occurred in statistically more acidic conditions with higher bryophyte cover than congener sites.

#### 55 - Distribution and habitat associations of the Shenandoah Mountain salamander, *Plethodon virginia*

Paige Pettry

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Nature's biodiversity is currently undergoing a "sixth mass extinction" largely driven by human activity, of which amphibians in particular are experiencing rapid decline. Amphibians are declining at a rate faster than that of birds and mammals, and over a third are currently at risk of extinction. Many terrestrial salamanders of the family Plethodontidae are at risk due to sensitivity to rising temperatures and habitat loss. *Plethodon virginia*, known as the Shenandoah Mountain salamander, is one such species endemic to part of the Valley and Ridge region of Virginia and West Virginia. The purpose of this study was to generate a GIS model of the probability of occurrence of *P. virginia* for use by state and federal agencies in their biological evaluations. *P. virginia* presence data was collected using daytime cover object searches and nighttime visual encounter surveys during the months of May and June 2021 after rainfall of at least 0.1 inches. The data collected, as well as previously collected presence data, was used to create a GIS layer in ArcGIS using the Presence-Only Prediction (MaxEnt) tool. The resulting model may provide a needed tool for land and wildlife managers and aid in the conservation of an endemic species.

#### 56 - Understanding the significance of soil microbiome and salinity in the growth of *Spartina alterniflora*

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Coastal salt marshes are highly productive and important ecosystems that are globally in decline. Along the Eastern and Gulf coast of the United States, salt marshes are dominated by *Spartina alterniflora*, which plays a vital role in marsh growth and maintenance. Due to its ecological importance, it is often planted as a biotic component in marsh restoration projects (e.g., living shorelines). In natural marsh environments, the rhizosphere of *S. alterniflora* supports a diverse microbiome, including microbes that enhance growth through nutrient fixation and detoxification of harmful compounds present in the soil. Furthermore, *S. alterniflora* and its associated microbial community grow in estuarine water, which can range in salinity. However, traditional nursery production of *S. alterniflora* include growth in a potting soil mixture and freshwater, and nutrients are supplied via commercial fertilizers, which do not mimic natural salt marsh conditions. The purpose of this research was to examine the effect of the addition of salt marsh soil and growth in brackish water on the growth of nursery-produced *S. alterniflora*. Seedlings propagated from locally collected seeds were grown in a full-factorial design in a combination of soil (salt marsh soil inoculation or potting soil only) and water treatments (brackish and freshwater). Growth was assessed weekly, and biomass was measured after 16 weeks. Plants grown in freshwater had significantly greater growth rate and leaf chlorophyll content compared to plants grown in brackish conditions. Additionally, plants grown in inoculated soil had a significantly greater growth rate, plant height, leaf chlorophyll content, and total biomass compared to plants grown in potting soil. Results suggest that the presence of salt marsh microbes increased growth and productivity of *S. alterniflora* in nursery production. These results will facilitate the development of the nursery production of native *S. alterniflora* for coastal salt marsh restoration efforts in Georgia.

#### 57 - Disturbance and Diversity: Lessons from Lichen Richness

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Diatoms are robust bioindicators, with species-level identification yielding meaningful water quality analyses of stream ecosystem health. Species-level identification requires high-levels of training to delimit morphological variation within microbial populations. The goals of this research were to document diatom communities in low-impact streams in the southeast, investigate seasonal and long-term variation, compare results with previous research in the area, and provide a diatom voucher flora for future use in research in this area. Two low-impact streams in middle Georgia were sampled February and April 2022, in which both sites met regional reference criteria for nutrients set by the EPA. Historical data was further analyzed through archived slides from 2011 (Tobler Creek) from the Georgia College Natural History Museum. Tobler Creek is a recovering agricultural stream on the site of Andalusia farm and has been in decades of recovery and is now a National Historic Landmark recognized by the National Park Service. Murder Creek, located within the Oconee National Forest, was monitored long-term by the USGS NAWQA program.

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Ecological disturbance is on the rise due to anthropogenic factors including habitat degradation and deforestation, yet the effect of disturbance on small but diverse and ecologically important species such as lichens remains understudied. This talk will discuss the effects of habitat quality on lichen species richness at a regional scale: the Southern Appalachian Mountains, a lichen biodiversity hotspot. We used a comprehensive data set (n = 892 species) collected across 208 one-hectare plots from northern Alabama to North Carolina, to determine which of a suite of environmental variables exerts the largest impact on species richness. Generalized linear models indicate that habitat quality, scored in the field on a 10-part scale, is the most important environmental factor influencing lichen richness. We then compared habitat quality and lichen richness using linear regression. We chose subsets of lichen functional groups including reproductive mode, growth form, lobe width, size, photobiont type, substrate, chemical protection, and physical protection. All groups show a strong positive linear correlation: as habitat quality increases, species richness increases. This indicates that, unlike other taxa, Southern Appalachian lichens do not follow the Intermediate Disturbance Hypothesis, at least at this regional scale. This finding also has important conservation implications. If we intend to support high lichen species richness, we must focus on protecting older, more intact forests.

#### 58 - Inverse settlement patterns of oyster spat (*Crassostrea virginica*) and barnacles (*Chthamalus fragilis*) at two southern Georgia salt marshes

Maris Guzman, Thomas Hancock

*Middle Georgia State University, Macon, GA*

The decline of eastern oyster (*Crassostrea virginica*) populations has been a cause for concern in recent years. This is due in part to the potential loss of the myriad ecosystem services provided by these animals. Such services include marsh stabilization, water filtration, habitat creation, and serving as a food source for the estuarine ecosystem. Although receiving less attention, barnacles (*Chthamalus fragilis*) are also important components of the Georgia salt marsh system, providing many of the same ecosystem services as oysters. Little is known about the settlement patterns of either oyster spat or barnacles in southern Georgia salt marshes. This study was initiated to better understand these settlement patterns and to determine if correlations exist between settlement and environmental factors. Three sites were selected on Sapelo Island and three sites on Jekyll Island in relation to the upland-estuary-sound gradient. Settlement racks were established at each site. Tiles were collected every six weeks and number of oyster spat as well as barnacles was determined. Additionally, water temperature and salinity were measured continuously at each site via data loggers. Oyster spat settlement was correlated with higher salinities and more open, ocean-influenced sites while barnacle settlement was correlated with lower salinities and more closed, upland-influenced sites. These correlations may reflect differences in feeding mechanisms and physiology exhibited by oysters and barnacles.

#### 59 - Uses of Diatom Community Analyses in Streams with Legacy Agricultural Uses in Middle Georgia

Sydney Brown<sup>1</sup>, Kalina Manoylov<sup>2</sup>

Reserved non-digested composite samples were analyzed, confirming that diatoms were the dominant algal group in the community. Cleaned from organic material, diatoms were identified to species-level and enumerated. Standard community analyses and comparisons revealed that in from 2011 to 2022, Tobler Creek experienced an increase in species richness ( $X^2=42.1$ ,  $p<0.05$ ) and diversity ( $X^2=207.4$ ,  $p<0.01$ ). Additional diatom-specific metrics were assessed, Multi-Metric Index (MMI) and Trophic Diatom Index (TDI), to measure overall ecosystem and trophic conditions of waters, respectively. There was no significant difference in TDI thorough time within-stream, but MMI showed degraded community health due to sedimentation. Between-streams TDI was higher in Murder creek, reflecting higher levels of measured nitrate-nitrogen. Results of this research confirm that continuous monitoring and taxonomic updates are required in diatom research until a baseline for all regions has been developed.

#### 60 - Analysis of fecal bacteria in the Wheeling Creek watershed

Emily Huff, James Wood

*Department of Organismal Biology, Ecology, and Zoo Science, West Liberty University, West Liberty, WV*

Fecal bacteria contaminating surface waters is a persistent problem impacting human health. While most fecal bacteria found in the fecal waste of warm-blooded organisms are nonpathogenic their presence in surface waters can indicate the presence of pathogenic bacteria or viruses and can cause gastrointestinal illnesses to humans. *Escherichia coli* (*E. coli*) has been recognized as the best indicator of fecal contamination when evaluating surface waters, however, it is difficult to distinguish between human and other warm-blooded organisms using *E. coli*. Conversely, the human gut bacteria *Bacteroides* is a fecal waste associated gene marker the EPA has utilized to determine sources of fecal material. We monitored water quality and identified predictors of *E. coli* in the Wheeling Creek watershed, a tributary to the Ohio River, in the northern panhandle of West Virginia, at seventeen sampling locations. We measured water temperature, pH, dissolved oxygen, conductivity, chloride and turbidity in the field and used an IDEXX system for *E. coli* quantification. Source tracking samples were also taken to determine the source of fecal material with the use of *Bacteroides*. Our results indicate *E. coli* concentrations are increasing in the downstream direction of Wheeling Creek and generally show a positive correlation with turbidity. Results of the *Bacteroides* study are forthcoming.

61 - Analysis of Cyanobacterial Community Composition in *Podostemum ceratophyllum*

Ashley Vankirk<sup>1</sup>, James Wood<sup>2</sup>, Nicole L. Garrison<sup>2</sup>

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*Podostemum ceratophyllum* is a vascular macrophyte that grows attached to stable substrates in shallow, swift-water streams and rivers throughout eastern North America. The plant is considered a foundation species because it provides shelter for benthic organisms and contributes to detrital and trophic food webs. *Podostemum ceratophyllum* appears to be declining throughout much of its native range and is assumed to have declined substantially from historical locations due to changes in water chemistry, habitat alteration, and anthropogenic impacts. The strong attachment between *P. ceratophyllum* and stable substrates is facilitated by the epithelial tissue (haptera) which securely fastens the plant to the substrate, but this connection has been observed to be weaken in degraded habitats, triggering detachment and loss of the plant. The exact attachment mechanism of *P. ceratophyllum* is still unknown, but it is hypothesized that cyanobacteria aid in the attachment of *Podostemum* to stable substrate. Understanding the biofilm community composition in healthy *Podostemum* populations could aid in identifying habitat characteristics needed for the conservation of the plant. Previous research confirmed the presence of cyanobacterial biofilms via microscopy; however, the specific species of cyanobacteria were not identified. To further analyze the relationship between *Podostemum* and cyanobacteria, we collected 20 *Podostemum* covered rocks from the Youghiogheny River, PA and Middle Island Creek, WV and examined the microbial community associated with epilithic attachment, focusing on genetic characterization of cyanobacteria found on haptera using three molecular markers (*nifH*, 16S rRNA, and PC). 62

- Water-level Fluctuation Effects on Yellow Perch Reproduction in a Hydropower Reservoir

Stuart Welsh<sup>1</sup>, Kyle Matt<sup>2</sup>, Dustin Smith<sup>3</sup>

## 63 - Conservation amid the RCRA/CERCLA remedial process

Susan Blas

*Savannah River Nuclear Solutions, Aiken, SC*

Lower Three Runs (LTR) is one of five major Savannah River Site (SRS) stream systems that discharge into the Savannah River in South Carolina. The surface water bodies and wetlands that define the LTR stream system have been under a Resource Conservation and Recovery Act/Comprehensive Environmental Response, Compensation, and Liability Act (RCRA/CERCLA) remedial investigation because of the potential impact to human health and the environment as a result of decades of site operations. Investigations concluded that a remedial action is necessary due to the presence of cesium-137 in sediment/soil, and cesium-137 and mercury in fish tissue that may pose a threat to human health and the environment. Collaboration with the Department of Energy, Environmental Protection Agency, and the South Carolina Department of Health and Environmental Control resulted in a final cleanup decision for LTR. As documented in the Record of Decision (ROD), the selected remedy for the upper portion of LTR is: (1) Land Use Controls (LUCs) with Monitored Natural Recovery (MNR), (2) Excavation, Treatment and Disposal of Principal Threat Source Material Sediment/Soil (at one location), and (3) Maintain Water in Ponds for two pond systems. The selected remedy was approved by the public, marks a substantial cost avoidance when compared with other remedial alternatives, and establishes long-term protection and monitoring for system.

The remedial decision reached for the LTR watershed was a positive step in environmental stewardship, protecting research opportunities, waterfowl flyways, and ecological habitats and wildlife associated with the LTR stream system. The selected remedy is a testament to collaboration with state and federal stakeholders that culminated in substantial cost savings along with the protection of approximately 30 miles of canals/stream system and over 3,000 acres of aquatic habitat while ensuring the protection of potential human receptors to the low levels of radioactivity in the LTR system.

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Water level fluctuations alter reservoir ecosystems, causing direct and indirect effects on fish populations. We examined relationships between water level fluctuations and spawning characteristics of Yellow Perch in a Central Appalachian hydropower reservoir. Daily presences of egg masses were recorded on artificial spawning structures at two sites for the spring spawning seasons of 2019 and 2020. Spawning structures were placed at different distances from the shoreline, spanning water depths with and without the potential for dewatering based on the lowest permitted levels for lake elevation drawdowns. Generalized estimation equations were used to analyze egg mass presence and six covariates: Secchi disk depth, distance to the shore, water temperature, water depth, lunar illumination, and lake level fluctuation. We also examined the proportion of egg masses in potential dewatering zones based on the minimum lake elevation drawdowns permitted for March and April. Data supported an additive effects model of year + water depth + lunar illumination + water temperature. The predicted probability of egg mass presence was negatively associated with water depth and lunar illumination and positively associated with water temperature. A year effect, in part, reflected a betweenyear difference in the timing of spawning, where the number of egg masses during April exceeded that of March in 2019, a relationship that was reversed in 2020. During the 27-day spawning period in 2019, 52% (54 of 104) of egg masses had the potential to be dewatered, whereas 70% (30 of 43) had the potential to be dewatered in the 22-day spawning period of 2020. Our results could have direct implications for fishery and hydropower management, as data on the characteristics and timing of spawning of Yellow Perch relative to water level fluctuations may help inform decisions regarding management of fish populations and lake level drawdown regulations.

## 64 - Analyzing Basic Morphometrics to Aid in Taxonomy of Federally-Listed Mussels from the Pea River Watershed in Southeast Alabama

Patricia Gordon, Victoria Zlotea, Zora Osborn, Jonathan Miller *Troy*University, *Troy, AL*

Freshwater mussels are the most endangered fauna in North America, with about 70% of the species considered at-risk, endangered, or extinct. Proper taxonomy is difficult within this group, although other resources to assist in identification may be utilized. Our research focus was to analyze basic morphometrics (Length:Height) to aid in taxonomy of mussels, specifically federally threatened and endangered species from the Pea River watershed. Our analysis primarily focused on the federally-listed species *Ptychobranchus jonesi*, *Hamioia australis*, *Pleurobema strodeanum*, and *Fusconaia burkei*, with comparisons of ratios between morphologically similar species. Comparison of Length:Height for *H. australis* and *P. jonesi* resulted in about half of the samples overlapping while *P. strodeanum* (88%  $\leq 1.5$ ) and *F. burkei* (78%  $\geq 1.6$ ) showed promising results. Some drawbacks include that we only measured mussels from the Pea River and major tributaries, thus results from the current study do not account for varying morphology from mussel species in our study occurring in other habitat types and/or watersheds. Further analysis of such variables across watersheds and habitat types, although likely resulting in variation from our study, should be analyzed to determine locations and species where such results may help provide supporting information for species descriptions.

65 - Land use across eastern North America is reflected in the elemental composition of the lotic macrophyte *Podostemum ceratophyllum*

James Wood

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Stream salinization and nutrient enrichment can alter aquatic food webs by alleviating stoichiometry constraints on primary production. Land uses that facilitate salinization may contribute toxic metals that can enter and possibly bioaccumulate in aquatic food webs. Some macrophytes can bioaccumulate metals at sub-lethal and lethal concentrations, potentially inducing toxic elements into food webs. We investigated the effects of land use on the elemental composition of the widespread riverine macrophyte, *Podostemum ceratophyllum*; a plant that grows in swiftwater habitats from Georgia, USA, through Canada in eastern North America. *Podostemum* is consumed by a variety of vertebrate and invertebrates (i.e. crayfish, Canada Geese, turtles) but the effects of land use on the elemental composition of the plant are unknown. We collected *Podostemum* from 91 locations distributed between Georgia and Maine in order to determine the nutritional quality of the plant and assess the influence of land use on its elemental composition. Our results indicated that increasing concentrations of  $\delta^{15}\text{N}$ , Cd, Cu, K, Li, Mg, and P were correlated with increasing urban development, while  $\delta^{13}\text{C}$ , Co, Cr, Mo, Ni, Sr, C:P, and N:P were positively correlated with increasing forest cover. Using PCA, axis 1-3 accounted for 51.3% of the variation in the dataset with axis 2 correlated with the nutrients P, Mg, Na, K, and Li. Potentially toxic concentrations of Cd, Co, Cr, Cu, and Ni were found in some samples. Our data indicates that land use can influence basal resource quality and toxicity in eastern North American rivers.

66 - Resolving evolutionary relationships in the ragworts: phylogenomics, divergence time estimates, and biogeography of *Packera* (Asteraceae: Senecioneae)Erika Moore-Pollard<sup>1,2</sup>, Jennifer Mandel<sup>1,2</sup><sup>1</sup>*University of Memphis, Department of Biological Sciences, Memphis, TN*, <sup>2</sup>*Center for Biodiversity Research, Memphis, TN*

*Packera* is a genus in the sunflower family that contains about 64 species and varieties endemic to North America. *Packera* is known to have a complicated evolutionary history due to hybridization and polyploidy, making it difficult to reconstruct phylogenetic relationships within the group. Previous research used sequencing methods such as nuclear internal transcribed spacers (ITS) to reconstruct phylogenetic hypotheses, but ultimately produced poorly resolved trees highlighting a lack of sequence divergence within the group, providing little information on the evolutionary relationships within this complex genus. Here, we generated nuclear and plastid phylogenies of *Packera* and related Senecioneae taxa using next-generation sequencing (NGS) to determine their evolutionary relationships, estimate the age of all tested taxa, and trace historical biogeographic patterns. We found that the nuclear and plastid phylogenies were highly incongruent, with the nuclear tree presenting higher resolution than the plastid tree, though still showing some areas of low resolution. The nuclear tree showcased that geography may play a large role in the evolutionary relationships within *Packera*. An apparent lack of plastid diversity may explain why the plastid tree was unresolved. The origin of *Packera* was dated as older than most other studies—at around 19.2MY – 25.9MY during the late Oligocene to early Miocene. Though these dates differ from prior phylogenetic estimates, they align well with previous geohistorical predictions that *Packera* speciation and diversification events coincided with the changing geography and climate in North America. Finally, *Packera* likely originated in the western United States or Mexico then diversified north and east into the rest of North America and Russia, further coinciding with previous geohistorical predictions.

## 67 - A meta-analysis to determine if concentrated animal feeding operations cause increased antibiotic resistance in the environment

Tab Viana

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Bacteria resistant to antibiotics threaten human health, limiting options for treating infections. Over-treating animals with antibiotics, even when not sick, contributes the spread of antibiotic-resistance (AR) genes. Commonly, concentrated animal feeding operations (CAFOs) provide antibiotics for growth rather than treatment of disease. It has been reported that AR bacteria/genes have been found near CAFOs. This study performs a meta-analysis of literature reporting AR genes found in media (soil, water, manure) near CAFOs. The goal was to determine if there was evidence of AR consistently higher near CAFOs compared to background levels. Literature was sampled on antibiotic resistance and CAFOs by performing a Google Scholar search using a combination of keywords and Boolean modifiers (“antibiotic resistance” AND “Concentrated Animal Feeding Operations” OR CAFOs AND PCR). The search resulted in 989 papers; the first 300 papers were analyzed. Only the primary papers collecting data from media near CAFOs and reporting AR in bacteria or the genes associated with AR were accepted. 2 sets of criteria were applied, leaving 34 papers for analysis. The first analysis determined if the paper compared data to negative control; 24 papers were found to have control samples. A second analysis examined whether statistical evidence reported increased AR associated with CAFOs. Of those 24 papers, 22 reported a statistical increase in AR in at least 1 AR gene/antibiotic compared to samples collected outside the influence of the CAFO. Results suggested no significant relationship between the CAFO type and whether there is a consistent increase in AR. Control samples are not as standard as expected to confirm CAFOs’ impact on nearby media, which is consistent with the higher resistance genes/antibiotics found. These data contribute to understanding the impact of CAFOs on AR and may help enact policies to control the use of antibiotics for purposes other than treating disease.

#### 68 - Effect of PDE5A on Left Ventricle Function Following Arteriovenous Fistula Creation

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Cardiovascular disease (CVD) is the leading cause of mortality among hemodialysis patients. Arteriovenous fistula (AVF) creation impacts cardiac structure and function. The Phosphodiesterase 5A (PDE5A) enzyme is overexpressed in the heart following AVF creation and may adversely impact cardiac remodeling by acting on the NO/cGMP pathway. We hypothesize that overexpression of PDE5A in the cardiomyocytes will result in adverse cardiac remodeling after AVF creation as compared to wildtype mice. AVFs were created in age matched PDE5A overexpression and wildtype mice. Echocardiography was performed at baseline and 1-, 7- and 21-days post-surgery. Left ventricular (LV) end-diastolic diameter (LVEDD), left ventricular end-systolic diameter (LVESD), and end-diastolic volume (EDV) were increased at 21 days in all AVF mice from baseline and in PDE5A compared to wildtype mice at 21 days post AVF creation. An increase in LV/body weight (LV/BW) ratio was also observed following this same trend. Masson’s Trichrome and Picosirius Red staining of LV showed an increase in collagen surrounding the perivascular areas of all AVF mice tissue with increased perivascular collagen observed in PDE5A mice compared to wildtype at 21 days post AVF creation. In conclusion, cardiomyocyte-specific PDE5A overexpression mice demonstrated increased adverse LV remodeling in comparison to wildtype mice following AVF creation. Our future studies will evaluate mechanisms of AVF-induced cardiovascular structural and function for PDE5A mice at longer time points and in the setting of Chronic Kidney Disease (CKD).

#### 69 - The role PVR, a growth factor receptor, plays in thermal nociception in Drosophila

Ishani Chattopadhyay, Andrew Bellemer

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Nociception is the sensory nervous system’s behavioral and physiological response to noxious stimuli. This noxious stimulus can present itself in various forms such as mechanical, thermal, chemical or UV radiation, and can trigger pathways that elicit a response behavior. This research project utilizes a *Drosophila* model of thermal nociception in order to understand the role that a growth factor receptor, Pvr, plays in thermal nociception. This model organism is utilized to make genetic manipulations that modify genes involved in nociception. Pvr is a cognate receptor related to the platelet derived growth factor (PDGF) and vascular epithelial growth factor (VEGF), that regulates several physiological processes in humans and other vertebrates. Previous experiments have established that Pvr is required in nociceptive sensory neurons for regulating baseline mechanical nociception, however, the role of Pvr in thermal nociception is not well understood. This project aims to knockdown and overexpress Pvr in the multidendritic neurons of *Drosophila* and assess the thermal nociception phenotypes that result. The thermal nociception assay demonstrated that manipulations of Pvr do not produce a significant defect in baseline thermal nociception. However, Pvr overexpression caused behavioral hypersensitivity to weak noxious stimuli. The hypersensitivity caused due to Pvr overexpression suggests that Pvr receptor activation may act to sensitize sensory neurons to noxious stimuli. In order to better understand the role of Pvr in sensitization, a tissue-damage assay will be performed using UV radiation to induce nociceptive hypersensitization in *Drosophila* with Pvr knockdown.

#### 70 - In vivo characterization of synthetic Notch signal transduction

Amber Ajamu-Johnson, Paul Langridge

*Augusta University, Augusta, GA*

Notch is a highly conserved cell-surface receptor found in all animals, that transduces signals between cells through interactions between the ligands, such as Delta, found on neighboring cells. Notch activation plays a role in cell proliferation, cell death, specific cell fates, and cell differentiation.

Notch is characterized by an intracellular domain that acts as a transcription factor and its extracellular domain composed of multiple epidermal growth factor (EGF) repeats and a negative regulatory region (NRR). Once the ligand on the surface of a neighboring cell binds to Notch, a cleavage within the NRR domain initiates the activation process. Importantly, force is required to pull apart the NRR and allow the activating protease access to the cleavage site. This force is provided by the endocytosis of the ligand via a select pathway involving the endocytic adapter protein Epsin. Using *Drosophila* Notch, we have produced synthetic receptors predicated on the mechanism of Notch activation but with heterologous binding regions. By replacing the NRR region of our synthetic Notch receptor with heterologous domains from a range of different sources, previous work has identified several interesting potentially force sensitive domains and testing in the wing for receptor activation in response to ligand. To gain further evidence that these domains are indeed sensitive to force, we performed an assay removing the capacity of the ligand to be endocytosed via Epsin and therefore to exert activating force on the receptor. We find that many of the protein domains do require endocytosis of the ligand via Epsin to at least get full activation, indicating that these domains are indeed cleaved in response to force. This finding suggests that many proteins may be cleaved because of force in a way analogous to the NRR and that this might have important roles in the native function of these domains.

#### 71 - Antiviral Potential of Fungal Endophytic Compounds against Adenovirus 5

Alexandra Wright

*Mary Baldwin University, Staunton, VA*

Across countries, there has been a surge in infectious diseases, and there is a significant lack of antivirals readily available to treat viruses. It is hypothesized that Cyclosporine A & Mycophenolic Acid show antiviral activity against replication-incompetent Human Adenovirus 5 (Ad5). Adenovirus 5 causes human infection and serves as an excellent model virus for double-stranded DNA non-enveloped viruses. Cyclosporine A & Mycophenolic Acid were chosen for this study because prior research has shown that both drugs inhibit a viral protein that is involved in replication expression. HEK 293 Mammalian cells will be infected with Ad5 and treated separately with varying concentrations of both antivirals. The expression of green fluorescent protein (GFP) will be detected via fluorescent microscopy and a cell count will be performed to quantify the number of cells positive for GFP. It is hypothesized that GFP expression will be lowest in trials with high concentrations of drugs and that GFP expression will be highest in trials with low concentrations of drugs or no drugs. Practically, this result would indicate that the antiviral compounds successfully inhibited the expression of the reporter gene and would likely inhibit viral replication in a replication-competent virus. This would support further research exploring these compounds against other double-stranded DNA viruses.

#### 72 - RNA-Sequencing Reveals the Mechanistic Role of Gene Expression in Niche Differentiation between *Populus deltoides* Clonal Varieties

Macy Gosselaar

*Mississippi State University, Starkville, MS*

Excessive nitrogen run off from agricultural operations can cause the eutrophication of the Northern Gulf of Mexico. Nitrate (NO<sub>3</sub><sup>-</sup>) contaminated groundwater and surface runoff from the southeast region of the United States flow into the lower Mississippi River Basin, causing hypoxic conditions, and deteriorating water quality. *Populus deltoides* planted in riparian areas could mitigate and intercept this excessive runoff. As an economically viable short rotation woody crop, and a carbon neutral fuel, *Populus deltoides* can quickly produce large quantities of biomass and can easily be converted into biofuels to produce bioenergy. Polyclonal plantings of *Populus* spp. are expected to display an increase in site resource utilization, and growth and productivity through differentiated root system sorption zones (i.e. niche differentiation) and crown/canopy structures. This would increase the efficacy of *Populus* spp. ability to intercept and mitigate excessive nitrogen runoff. However, underlying molecular mechanisms of niche differentiation, including changes in site resource utilization and growth of polyclonal *Populus* plantings in the southeast region of the United States are poorly understood. We hypothesize that our two varieties of *Populus deltoides* in polyclonal plantings will show greater regulation of gene expression compared to our two varieties planted in monoclonal plantings. Early results from a likelihood ratio test identified 102 DEGs (FDR <.05) between polyclonal and monoclonal treatments. 97 were upregulated and 5 were downregulated and annotated through Gene Ontology analysis. Analysis of clones in their respective planting combinations and between endophyte and non-endophyte treatments were also included in this study. A Kyoto Encyclopedia of Genomes and Genomics identified downregulated regulatory networks in monoclonal plantings of pyruvate metabolism, citrate (TCA) cycle, and other types of O-glycan biosynthesis, which efficiently coordinates and controls growth. This project could aid in developing genomic-based tools or approaches to better predict planting combinations providing the greatest productivity for future plantings.

#### 73 - A Floristic Checklist of Shenandoah River State Park, Warren County, Virginia

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A floristic checklist of vascular plants was constructed for Shenandoah River State Park (SHSP) in Bentonville, Virginia during the 2021 and 2022 growing seasons. SHSP encompasses 665 hectares (1619 acres) of the northern Shenandoah Valley between the Massanutten Mountains and Shenandoah National Park. The park lies in Warren County which has been botanically underexplored, consistently ranking last in plant collection metrics among the five northern Shenandoah Valley counties. SHSP contains 11 community types that span 165 – 296 m.a.s.l. from inland wetlands and river scour communities to upland forests, including endemic Central Appalachian Shale Barrens. A total of 797 collections yielded 536 unique taxa from 109 vascular plant families. These collections were secondarily extended with in-field pictures that were aggregated on an iNaturalist

project page. Quick Response (QR) codes were used to link herbarium vouchers to their respective iNaturalist entry. Sixty-four county records of taxa not yet observed in Warren County and one state record, *Equisetum laevigatum* were documented. As a first for SHSP, this floristic checklist provides baseline botanical knowledge of the park and expands the available digital data of vascular plants for Warren County by 20%. This augmented knowledge, in tandem with the accompanying iNaturalist project page, will not only be useful to park managers for future development and conservation plans but will also serve as a tool for community scientists to enhance their knowledge of the local flora.

74 - Taxonomic, conservation, ecological, morphological, and reproductive analysis of 4500 plant species of the interior Southeast US.

Sebyn Brothers, Joey Shaw

*University of Tennessee at Chattanooga, Chattanooga, TN*

Tennessee and Kentucky are centrally positioned in the interior Southeastern US and combined they contain ~3600 vascular plant species and lesser taxa. There are ~900 additional species or lesser taxa across the broader region. Launched in 2020, the Tennessee-Kentucky Plant Atlas (TNKY) has become a major resource for botanists, conservation workers, students, and teachers in the region. Underlying TNKY is a database of >100 taxonomic, conservation, ecological, morphological, and reproductive characters for the ~4500 taxa. While a work in progress, here we report on some characteristics of the flora, which is represented by 215 families, 1024 genera, and ~4500 species and lesser taxa, 95% of which are angiosperms. Conservation characteristics include nativity, region of origin, invasiveness, global and state ranks, and wetland statuses. Ecological characters range from general ecological habitats of terrestrial or aquatic, soil and moisture regimes, to ecological systems of NatureServe. The database has >20 characters related to leaf and stem characters and 30 reproductive characteristics. Eighty percent of the flora is native and over 200 species are listed invasive by at least one southeastern state. Eighty-seven percent are terrestrial, 7% are in mudflats and wet woods, 3% are aquatic, and 1% are strict wetland species. Fifty-six percent are forbs, 19% are graminoids, and 17% are woody. Seventy-six percent are perennials, 19% are annuals, and 4% are biennials. The majority of species reproduce in May, June, or July. Research is ongoing and will continue to fill in more characteristics present within the database.

75 - Moderate windthrow and salvage logging do not alter herbaceous diversity or composition in long-term study of Tennessee coastal plain forest

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The southeastern United States supports high plant biodiversity in forested systems. However, maintaining this biodiversity in the face of climate change will require understanding how natural disturbances facilitate the coexistence of plant species. Few long-term studies have analyzed the effects of varying disturbance severity (from multiple natural and/or anthropogenic disturbances) on herbaceous species diversity and composition. Here, I present a study of a coastal plain forest that weathered a moderate-severity windstorm and subsequent salvage logging in 1999 and analyze how herbaceous diversity and composition have changed between sampling in 2001 and 2021.

In 2001, sixteen 30x30 meter plots were established at Natchez Trace State Park in Tennessee; eight in unsalvaged stands and eight in salvaged stands. Plot-level severity was calculated, based on the proportion of basal area fallen or salvaged. Average herbaceous diversity, composition, canopy openness, and soil moisture did not differ between unsalvaged and salvaged stands (aka severity treatments). In 2021, I collected the same types of data in these plots to make historical comparisons. While the number of subplots sampled differed between years of sampling, average plot-wide comparisons are still valid. I found herbaceous diversity and soil moisture did not vary between severity treatments (2021) or between years (2001 to 2021). Composition did not differ between severity treatments (2021). Canopy openness did not differ between severity treatments (2021) but did decrease with year (2001 to 2021). These data support two conclusions: 1) low-to-moderate severity disturbances (whether from single or multiple disturbances) may not alter herbaceous diversity and composition, and 2) the herbaceous layer, in absence of additional major disturbances, supports similar levels of diversity and composition over long periods of time, as demonstrated by similar values in both 2001 and 2021. This long-term study supports the validity of many short-term (3-5 years) studies' post-disturbance conclusions.

76 - Progress toward the Second Edition of the Guide to the Vascular Plants of Tennessee.

Joey Shaw

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Two comprehensive floristic works for Tennessee have served as compendia of the state's vascular plants, Gattinger's (1901) *The Flora of Tennessee and Philosophy of Botany* and the Tennessee Flora Committee's (2014) *Guide to the Vascular Plants of Tennessee*. The former was an overview of the vegetation and an annotated checklist with representative site locations. It reported 61 pteridophytes, 15 gymnosperms, and 2142 angiosperms (2218 total), although these counts are inflated by today's standards. The latter was built on an additional century of botanical research and included sections on the physical environment, history of botany, overview of vegetation, floristic summary, and keys to families, genera, and species. The *Guide* included 92 pteridophytes, 14 gymnosperms, and 2772 angiosperm species and lesser taxa (2878 total). Following publication of the *Guide*, (1) the SERNEC herbarium specimen digitization increased access to hundreds of thousands of herbarium specimens; (2) workers in the field and across our herbaria were more easily able to recognize and report aspects of the flora not reported before; (3) new tools like iNaturalist allowed more people to document aspects of the state's flora and alerted researchers to findings; (4) scientific progress has resulted in continued clarification of the taxonomy and nomenclature of Tennessee's flora; and (5) with nearly 3000 copies of the first edition sold, many users have

contributed toward refinement of the keys. For these reasons the Second Edition of the *Guide* is expanding to approximately 3000 species and lesser taxa and it will contain more accurate species descriptions, habitat information, and keys. The vascular flora of Tennessee and progress toward the Second Edition will be discussed.

77 - The significance of small parks for preserving rare plant populations and maintaining regional biodiversity

Clara Thiel

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Insular ecosystems, or those that are associated with distinct landscape features, are often maintained by environmental stress and support unique, sub-climax plant communities. Stress-tolerant plants supported by these habitats are often rare or geographically disjunct from their primary ranges and may contribute significantly to regional biodiversity. However, because insular habitats are typically small, fragmented, and fixed upon a landscape, such communities and the rare plants they support may be disproportionately impacted by changes in stress regime or land use. At the confluence of the Potomac and Shenandoah Rivers, a small historical park has been found to encompass an array of insular ecosystems and regionally rare plant species across MD, VA, and WV. A Park-wide resurvey from 2019-2021 aimed to 1) relocate known populations of rare, threatened, and endangered plant species within the Park, and 2) describe changes in dynamics and stability of rare plant communities since the previous rare plant inventory in 1998. A total of 45 rare, threatened, or endangered plant species were observed during the 2019-2021 resurvey. Of the 56 species historically known to occur within the Park, 34 were relocated. Losses and declines in rare plant populations were associated with shifts in stress regime, changes in land use, and increased presence of invasive species. Browsing pressure, increased Park visitation, and implications from invasive pests additionally threaten the integrity and long-term viability of rare plant communities within the Park. Despite these declines over the past two decades, Harpers Ferry National Historical Park continues to support and provide habitat refugia for rare and disjunct plant species. As such, this and other small land preserves remain of high importance for the maintenance and conservation of regional species and ecosystem diversity.

78 - A reevaluation of seed features as a taxonomically valuable feature for lepidote species of *Rhododendron*

Charles Horn

*Newberry College, Newberry, SC*

There are now four described lepidote species of the genus *Rhododendron* (Ericaceae) known in the southeastern United States, *R. carolinianum*, *chapmanii*, *minus*, and *smokianum*, with the last species officially recognized in 2021. Morphological separation has been difficult and sometimes confusing. Features have included corolla tube length, relative corolla lobe lengths, flowering time relative to new leaf emergence, seed size, and seed ornamentation. This investigation was developed to compare seed size and ornamentation as a potential taxonomic feature to separate all four species. Fruit samples from multiple populations of *R. minus* and *carolinianum* were field collected in South Carolina, North Carolina, and Tennessee. Samples of *R. smokianum* and *chapmanii* were limited due to their conservation status and presence of limited herbarium material. Microscopic work was used to compare seed length and width as well as investigate seed surface ornamentation. Data shows that at the extremes, seed length does help separate the species with *R. chapmanii* seeds being the longest, however, much overlap exists. Also observed were variation in seed shape from oval, half moon, and elongate. Seed morphology does not appear to be of taxonomic value.

79 - Improving adult captures of ambrosia beetles using different monitoring tools

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Ambrosia beetles (Curculionidae) are known for their ability to make pin-sized holes in their host trees where they begin to cultivate a fungus as a food source. Because of this, ambrosia beetles can cause significant damage to their host plant through these holes limiting nutrient and water uptake, their damage may lead to death of the host plant as well as affecting the salability of attacked plants. In Virginia, the effects of ambrosia beetle have been well documented, where treatments for this pest involve using pyrethroid insecticides to ensure the tree protection. The use of ethanol lures alongside with killing agents have proven to be effective monitoring methods for ambrosia beetles, but there is a lack of quantitative evidence on how the interaction between killing agents and lures influences trapping ambrosia beetles. In this study, we looked at the effects of soapy water, boat antifreeze, and car antifreeze in combination with ethanol lure or no lure in capturing ambrosia beetles. From this study, it was determined that the presence of an ethanol lure significantly increased the number of ambrosia beetles captured with boat. While captures were lower without the presence of an ethanol lure, it was still documented that the boat antifreeze attracted and captured ambrosia. Therefore, it can be concluded that the combination of boat antifreeze and ethanol lure has a potential interaction that positively attracts beetles which leads to a higher overall capture rate. Having an effective monitoring tool is crucial to time properly a pest control tactic.

80 - Adventures in Beetle Busting: Alternative Methods for the Eradication of Asian Longhorned Beetle in South Carolina

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The Asian longhorned beetle [ALB; *Anoplophora glabripennis* (Motschulsky); Coleoptera: Cerambycidae] is an invasive wood-boring beetle native to China and the Korean peninsula. It is capable of infesting 12 genera of hardwood trees in the U.S., causing structural failure and tree mortality after repeated infestations. The Asian longhorned beetle has been a known invasive in the United States since 1996 when it was discovered in New York, although it may have been present for more than a decade prior to discovery. Throughout its tenure in the United States, six distinct quarantines have been established by the United States Department of Agriculture and cooperative programs have been formed with state authorities to eradicate ALB. Two of these quarantines have been successful in complete eradication with three of remaining four achieving partial eradication in outlying areas. Eradication is primarily accomplished through tree removal and destruction. The newest quarantine zone in Charleston County, South Carolina was established in 2020. As the most southern and most coastal infestation, standard eradication protocol cannot be effectively implemented at all sites within this infested area due to low-country bottomlands, which frequently flood, inhibit survey accessibility, and prohibit tree removal with heavy machinery. In a 66.7-hectare bottomland site in Charleston County, three experimental treatments were assessed to determine their efficacy in eradicating ALB from known infested trees. Fifty-seven *Acer rubrum* between 5.1 and 51 centimeters (2-20 inches) in diameter were selected at random. Experimental groups included 3 baseline harvest, 12 control, 12 May hack-and-squirt, 6 August hack-and-squirt, 12 drop-and-leave, and 12 drop-and-chop. Research was conducted from May 2021-May 2022.

#### 81 - Using eDNA to survey ramp's pollinators

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Ramps (*Allium tricoccum* Ait.) are early spring ephemerals native to North America. They have a pungent onion/garlic smell and are a popular side dish in many regions in the southeast. Despite their popularity, little is known about the life-history traits of ramps, including the pollinators of the flowers. In this study, we collected 14 umbels and 10 individual flowers of ramp in two localities, Bolivar, PA, and Grafton WV, in June and July of 2021 respectively. Using an eDNA approach, we isolated the insect's DNA present in the ramp flowers and amplified the cytochrome c oxidase subunit I (COI) barcodes. Insects observed visiting the flowers were also collected for visual identification. Our eDNA analyses revealed the presence of three major orders of insects: Diptera, Hemiptera, and Lepidoptera, comprising 98.8% of the total reads obtained from the flowers. Within Diptera, snipe flies (Rhagionidae) and midges (Chironomidae) were the most abundant. Spittle bugs (Cercopidae) were the dominant group recovered within Hemiptera, and leafroller moths (Tortricidae) in Lepidoptera. Bumblebees, honeybees, and other bees were observed visiting the flowers; however, no Hymenoptera DNA was recovered in our eDNA analyses.

#### 82 - DNA Barcoding to Determine the Occurrence of Exotic and Native Earthworms in Residential Compost Bins in Nashville, TN

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While earthworms are an essential aspect of indigenous terrestrial ecosystems, they also play a key role in decomposition of organic matter in compost bins. Earthworms and their eggs can enter open compost bins directly or indirectly through organic matter added to bins. This research examines the diversity of earthworm species in residential compost bins and the yard areas surrounding them, specifically noting the distribution of exotic and native species of Oligochaeta. Earthworms were extracted from various compost bins and surrounding sites in a residential backyard in Nashville, Tennessee. After extraction, the worms were placed in ethanol and DNA barcoding was used to identify the species of earthworms. Food waste compost bins were hypothesized to have greater species diversity than yard waste bins, and the yard areas around compost bins would have greater species diversity than either compost bin type. Based on prior studies, it was thought that the yard would contain a prominent native species population. Based on the results of DNA barcoding, there were overwhelmingly more exotic than earthworm species, with most worms being *Eisenia andrei* and *Dendrobaena attenuata*. Overall, sixteen different species were found in and around the compost bins with the highest diversity in the compost bins. More worms were found when the soil pH was closer to neutral, the temperature was warmer, and there was more moisture in the soil. The introduction of exotic species into native habitats creates competition with native species and changes biotic processes, including those in compost bins. Further studies could examine the interaction between native and exotic earthworms in compost bins and how their occurrence might impact decomposition.

#### 83 - Move it Along: Coevolution of Parasite Longevity and Host Morphology

Kate L. Sheehan

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Symbiotic relationships represent combinations of host tolerance and symbiont adaptation. Despite their derived status, the body systems of many parasites are relatively simple, with many vestigial structures lost through evolutionary processes. For example, feeding structures are not developed in any of the life stages of most tapeworm species. While conducting a field study of the avian parasite, *Schistocephalus solidus* (Cestoda: Platyhelminthes) we documented the shedding of parasite stoma that appear to have senesced following a nearly semelparous period of fecundity.

The short-term duration of parasite reproduction coupled with a morphology void of attachment structures (in the adult stage) suggests *S. solidus* is adapted to persist for only days in the gut of the definitive host with limited to no resistance of digestive movement. This is likely the case in Mallards (*Anas platyrhynchos*: Anatidae), that have a relatively lengthy and slow-moving digestive tract. It is in this species that much of the life history of the parasite's adult stage was initially explored. Thus, what we think we know about this parasite is based on limited host taxa. However, the morphologies of other documented definitive hosts like loons (*Gavia* sp.: Gaviidae), gulls (Laridae), and other piscivorous birds are likely to provide habitat and sustenance for the worms for periods too short to allow for successful maturation of the worm. Thus, the phenotype of the parasite is likely to have evolved with these hosts, where they would have to resist peristalsis. Here, we follow the adult stage of *S. solidus* under scenarios intended to mimic those of different groups of avifauna, evaluate the records of infections and gastrointestinal morphologies for several avian hosts, and assess the coevolutionary consequences of other tapeworms that infect these waterbirds.

#### 84 - Intracellular endosymbionts, mitochondria, and temperature in an introduced spider with an expanding northward range

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The brown widow spider, *Latrodectus geometricus*, was initially introduced into south Florida around 1940. Over the last 30 years, the range has expanded northward into Georgia and the Carolinas. This spider is often infected with the maternally transmitted bacterial endosymbiont, *Wolbachia pipiensis*. While *Wolbachia* commonly has significant reproductive and fitness effects on its host, the consequences for brown widows are unknown. Introduced populations of brown widows have a northward trend of decreasing *Wolbachia* infection frequency. This pattern led us to hypothesize that temperature might affect the host-*Wolbachia* interaction. We predicted that infected individuals reared in lower temperatures would have either an increased mortality rate or a decreased *Wolbachia* titer than those at higher temperatures. In addition, because the same cellular components transmit *Wolbachia* and mitochondria during cell division, we hypothesized that the presence of *Wolbachia* would influence the number of mitochondria. To test these predictions, egg sacs of both *Wolbachia*-infected and uninfected spiders were split and reared at either low(15°C) or high(30°C) temperatures. Development time and mortality (% of unhatched eggs) were compared between temperature and infection treatments. Quantitative PCR (qPCR) was used to determine the relative amount of *Wolbachia* and mitochondria on the day of hatching. Development time was 2.5x longer in the cold treatment, but mortality rate did not differ based on temperature or infection status. However, there was a significant decrease in *Wolbachia* density in the low-temperature treatment. In addition, mitochondria copy number was greater in *Wolbachia*-infected spiders raised at the higher temperature than in all other treatments. This result supports the hypothesis that mitochondria and *Wolbachia* have evolved a cooperative relationship, but this relationship is disrupted at lower temperatures. Additionally, the lower *Wolbachia* infection frequency in more northern populations may result from generational dilution due to increased frequency of low temperature exposure.

#### 85 - Prevalence of *Trypanosoma cruzi* in sylvatic populations of Virginia opossum (*Didelphis virginiana*) and raccoons (*Procyon lotor*) in Southwest Georgia and Southeast Alabama, and its implications for human health

Richard A. Matthews<sup>1,2</sup>, Douglas S. Wright<sup>3</sup>

#### 86 - The Effect of Shelter Type, Soil Moisture, and Distance of the Trees to a Water Source on Riparian Restoration Tree Growth

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Riparian zones are the areas between freshwater sources, such as a stream, spring, lake, or river, and the upland. Riparian restoration is important to better protect and preserve sources of freshwater. Passive restoration involves removing the stress on the environment by fencing out livestock, and active restoration involves planting seedlings in tree shelters to promote the seedling's survival to adulthood. In this study, unventilated and ventilated shelters were used to determine if they would make a difference in the growth of the trees. To determine if shelter type influenced soil

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Chagas disease, or American Trypanosomiasis, is caused by the flagellated protozoan parasite *Trypanosoma cruzi* (Colato et al. 2017; Ribeiro et al 2020, DeSouza 2020). The parasite was first identified as the cause of Chagas disease in 1909 (Santos and Falcão 2020). The disease is of major social and public health concern in Latin America and has been regarded as a neglected tropical disease by the World Health Organization (PérezMolina and Molina 2018). It affects many poor and disadvantaged people, and the estimated annual global burden of the disease is between \$62746 million in health care costs. The disease is endemic in 21 Latin American countries, and has usually been confined to poor, rural areas of Central and South America (Pérez-Molina and Molina 2018).

*Trypanosoma cruzi* is spread by kissing bug vectors in the subfamily Triatominae and is maintained in mammal populations in both Latin America and North America. Here, we focus on the presence of *Trypanosoma cruzi* in sylvatic populations of Virginia opossum (*Didelphis virginiana*) and raccoons (*Procyon lotor*) in Southwestern Georgia and Southeastern Alabama in the United States. We collected heart tissue from 22 opossum and 27 raccoons from 2019 to 2022 and sampled it for the presence of DNA from *Trypanosoma cruzi*. Two (9%) opossum and 8 (30%) raccoons tested PCR positive for *T. cruzi* DNA. These data are consistent with other similar studies conducted in rural Georgia and have implications for public health across the area.

moisture content, the distance to both a stream and a natural spring were measured. Four tree species were studied: *Platanus occidentalis*, *Liriodendron tulipifera*, *Amelanchier canadensis*, and *Asimina triloba*. Data were analyzed in ArcGIS and SPSS statistical software. There was no significant effect of shelter type on the first-year growth of the different species of trees. The distance to the groundwater source for all trees proved to be statistically significant showing a positive relationship to growth: the trees grew more when planted farther away from the water source, with a p-value of 0.002 and an R<sup>2</sup> value of 0.064. However, soil moisture was only statistically significant based on the amount of growth for *Asimina triloba* with a negative relationship to growth: the trees grew more when the soil moisture content was lower, with a p-value of 0.005 and an R<sup>2</sup> value of 0.186. Although there was currently no difference in the growth based on shelter type, the growth could become different in the second or third year of growth.

#### 87 - Evaluating existing red spruce data products in West Virginia

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Red spruce forests are a conservation priority within West Virginia because of their large-scale destruction and because they provide critical habitat for species of conservation concern. A number of data layers describing the extent of red spruce forests in WV have been produced; however, these products lack critical information stakeholders require for selecting among the choices when engaging in management activities. Our objectives were to: (1) produce an evidence review to evaluate existing red spruce data products and (2) to quantify the accuracy of the most widely used red spruce data product. For objective 1, we compiled all existing red spruce data products, generated complete metadata, and identified and assigned evaluation criteria to each data product. For objective 2, we sampled 105 red spruce sites and leveraged 189 additional field validation points in WV to assess the accuracy of the most widely used data product. In each site, red spruce cover was quantified as high, medium, low or none. We quantified both user accuracy (how often the map category was present on the ground) and producer accuracy (how often points on the ground were correctly assigned on the map). Only 56% of pixels were correctly assigned to a red spruce cover category based on user accuracy. However, the user accuracy was fairly high for areas where red spruce was absence (84% of pixels classified as none were correctly assigned). Although only 45% of areas with high red spruce cover were classified correctly, 89% of pixels with high red spruce cover were classified as either high or medium cover. Overall, producer accuracy was higher than user accuracy. Our work highlights an opportunity to improve existing WV red spruce data products, particularly for high and medium cover classes, which are critical to delineate for species that depend on red spruce cover. 88 - Using Geospatial Data and Tools to Locate Rivercane

Diane Styers<sup>1</sup>, G. Rebecca Dobbs<sup>2</sup>

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Rivercane is a large native grass (technically a bamboo) located throughout the southeastern US and has been used for centuries by Indigenous peoples to make baskets, mats, blow guns, and more. Because non-Indigenous settlement of the region resulted in destruction of much rivercane, tribal nations such as the Eastern Band of Cherokee Indians (EBCI) have a keen interest in finding and promoting rivercane in the present. In this study, funded by EBCI, we used freely available LiDAR and other geospatial data to identify locations of rivercane within a small study area in Macon County, North Carolina. Using LiDAR, hydrography, and soil data as inputs, we developed a predictive model of rivercane locations, which were then field verified via ocular surveys from adjacent navigable waterways. Within the 30,250-hectare study area, the model identified 28.5 hectares (613,545 five-foot cells) of rivercane along streams and rivers. Field observation revealed an active process of regeneration, with mature canebrakes and smaller, younger areas not yet large enough to qualify as canebrakes. We defined a brake as a rivercane area at least 15 meters long, containing stems ~1 cm in diameter (a suitable size for harvesting by traditional Cherokee artisans). Our results suggest that this geospatial method of predicting rivercane could be used throughout rivercane's natural range and as such would be a valuable tool for use by Southeastern tribal nations in revitalizing access to this key artisan resource and culturally important plant.

#### 89 - Investigation of Glacial Relictual Populations of Chestnut Oak in the Piedmont of North Carolina

Ashlye Dullye, David Vandermast

*Elon University, Elon, NC*

During the Pleistocene Epoch, glaciers covered about 5,000,000 square miles of North America and spread as far South as the latitude 37 degrees North. These glaciers allowed cold adapted flora—otherwise referred to as relictual populations—to flourish. Yet, as the glaciers retreated, these relictual populations did as well. However, small populations continue to persist in higher elevations within southeastern and western North America; this is because higher elevations provide for a colder atmosphere due to the adiabatic lapse rate concept. This study investigates the relictual population *Quercus montana*—commonly referred to as Chestnut Oak—to characterize the populations of Chestnut Oak, to determine which environments relictual populations grow best in, and to discover whether the relictual populations will continue to persist within a warming climate. This past year, data was collected and analyzed over 50 plots on the two mountains in the North Carolina Piedmont, Occoneechee and Cane Creek. While analyses on the older tree populations has been conducted—including the Shannon index, basal area, importance value, and aspect transformation—and it was revealed that the tree populations are healthy going into a changing climate and that the warming climate has not significantly impacted the older populations yet. However, further community analyses are to be conducted on the seedlings and saplings data to determine if the newer generations are being impacted by the warming climate. Additionally, soil analysis is to be conducted on all plots to investigate what environments and with what nutrients relictual populations, particularly Chestnut Oak, grow best with.

90 - Isolated lizard populations face elevated extinction risk during catastrophic hurricanes

Sean Giery

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In The Bahamas, small coastal island ecosystems are frequently impacted by hurricanes and tropical storms. Decades of annual surveys of brown anole lizard (*Anolis sagrei*) and orb-weaver spider (Araneidae) populations on these islands has revealed that extinctions due to catastrophic hurricanes are widespread, but often incomplete. Whether an island population persists through a hurricane is generally determined by island height and area—tall islands provide topographic refugia from storm surge, and large islands harbor larger populations that buffer against the stochasticity of catastrophic demographic loss. In September 2019, Dorian, an historically strong Category-5 hurricane made landfall in the close vicinity of our long-term study system. Post-storm surveys in 2020 revealed that extreme winds (max sustained=295km/h) had devastated these small island ecosystems. However, as usual, some lizard populations persisted. Based on previous work, we expected that Dorian impacts would be driven by island height and area. Curiously, they were not. Using multiple logistic regression to explain post-Dorian persistence for 46 lizard populations, we found no effect of height or area ( $P>0.1$ ). Instead, we found a clear, negative effect of island isolation—that is, lizard extinctions were more probable ( $P<0.01$ ), and the magnitude of declines were greater ( $P<0.05$ ), on geographically isolated islands. The mechanisms underpinning isolation effects remain under investigation. However, a retrospective analysis of hurricanes passing within 30km of our study system indicated that isolation had contributed to lizard extinctions in previous hurricanes as well. Clearly, there is much left to learn about hurricanes. Given the increasing rate and intensity of tropical storms—and their catastrophic effects—understanding the effects of these extreme weather events on ecological communities has never been more important. Only through careful analysis of long-term data and rapid responses to unplanned research opportunities can we better understand and predict the factors influencing demographic and ecological vulnerability to hurricanes and other catastrophes. 91 - Resource Partitioning of Sympatric Carnivores in Western North Carolina

Maya Feller<sup>1</sup>, Aimee Rockhill<sup>1</sup>, Elizabeth Hillard<sup>2</sup>

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Four species of sympatric carnivores occur in western North Carolina, coyote (*Canis latrans*), bobcat (*Lynx rufus*), gray fox (*Urocyon cinereoargenteus*), and red fox (*Vulpes vulpes*). The coyote has been known to suppress the population of smaller carnivores in the area, which can lead to a shift in the overall ecosystem through interspecific competition. Our objective was to better understand how carnivores are partitioning the available resources by using geospatial and diet analyses. To assess habitat use through space and time, foot traps were set in the summer and fall of 2020 and GPS collars were fitted to at least one individual of each species. The results were analyzed in R using resource selection functions with linear mixed models based on distances to habitat and landscape characteristics. To assess diet, scat samples were collected within the study area and the carnivore species identified with a DNA analysis. A subsample was taken from each sample, washed and the contents visually identified. Coyotes stayed in habitats with lower human density that were further away from the other collared individuals. The home range of bobcats and both species of fox were closer to each other and closer to human habitation. Both the red foxes and the bobcats stayed closer to human development though the foxes selected for roads and the bobcats tended to avoid them. The gray fox followed powerline corridors and avoided secondary roads. A total of 31 scat samples were identified with DNA analysis from the samples collected. Most were identified as bobcat and small mammals were the most common dietary item found, particularly in the bobcat samples. Canid scat also included mammals but were more likely to include other food sources such as vegetation, insect exoskeletons, and bird.

92 - Are wet canopies moving lots of carbon? Developing new methods to address the assumption that wet canopies are barriers to the atmosphere.

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Tropical and temperate forests, the major carbon-fixing ecosystems, experience wet canopies from 40-100% of the year. Current Earth System Models that predict global carbon fluxes overlooks the effect of wetness on carbon and canopy conductance largely because there is no method that works under wet conditions. Therefore, conclusions about how canopies move carbon come from an understanding of dry leaves. New evidence suggests these assumptions are incorrect and introduce uncertainty into global CO<sub>2</sub> exchange estimates, which inform approaches to sustain the planet. To solve this urgent problem, we developed a new method for measuring wet-leaf conductance using the LI-6800 portable photosynthesis machine. In this approach we independently measured the flux of evaporation off the leaf surface and transpiration out of the leaf which allowed for us to effectively calculate leaf conductance and leaf carbon fluxes. We measured conductance in two species: *Liquidambar styraciflua* ("sweet gum") and *Viburnum dentatum* ("Viburnum"). Our findings from both species demonstrated that when wetting occurred on the top of the leaf, where there are relatively few stomata, there was little change to leaf conductance, meaning that significant carbon exchange is still occurring. However, when wetting occurred on the bottom of the leaf, where there are more stomata, there was a proportional decrease to about 50% of the originally measured conductance. This suggests that water serves as a barrier only when stomata are obstructed, a scenario that is uncommon under most canopy wetting scenarios in the field. For both wetting scenarios, conductance is still much greater than current Earth System Models would predict. We are now developing new predictive models of carbon cycling of wet canopies that will redefine our predictions of carbon fluxes through forests.

93 - Seedling photosynthetic response of six co-occurring species across gap edge light gradient

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In mixed-oak forest ecosystems, understanding the physiological mechanisms driving competition between co-occurring seedlings can help managers promote oak and hickory regeneration. Canopy gaps increase light to the understory, but also create a gap-edge environment thought to favor oaks. Can light alone explain the success of oak at the edge environment in relation to its two common competitors, tulip-poplar and red maple? Many studies have addressed this question in controlled settings, but none have explored this question in field conditions using co-occurring and naturally regenerated seedlings. This study was conducted four years after canopy gap harvest in the Pisgah National Forest, NC. Across a 30 m transect at three gaps (0.4 ha), locations were selected in the gap, edge, and forest understory. At each location, healthy seedlings of each of six species were studied including white oak, chestnut oak, northern red oak, hickory, tulip-poplar, and red maple. Seedlings were 50% taller in the gap than the edge and forest. Specific Leaf Area was 25-50% higher in the forest than the edge and gap. Light response curves showed higher light saturated photosynthesis (Asat) for seedlings in the gap than the edge and forest, with oak and hickory highest in all three locations. Photosynthetically active radiation data were used to estimate daily carbon assimilation per seedling and evaluate species across light environments. All species had higher daily total assimilation with increasing light except tulip-poplar, which did best at the edge. Light alone did not explain why oak may be most abundant or successful at the edge since tulip-poplar ranked high in daily total assimilation at the edge and forest.

#### 94 - Species-specific thermal stress responses and lethal tolerance limits in freshwater mussels of the Pea River, AL

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Freshwater mussels are highly imperiled and vulnerable to the effects of climate change. Yet, estimates of thermal tolerance exist for only a small fraction of mussels, and comparative estimates for sympatric species are virtually non-existent. Knowledge of species-specific thermal tolerance is essential to unionid conservation efforts, particularly in species-rich areas such as the SE U.S. To address this gap, we quantified interspecific differences in thermal tolerance of 3 sympatric species of the Pea River in Alabama. Measures of thermal tolerance included sublethal indicators of thermal stress, temperature at occurrence of maximum resting metabolic rate (RMRpeak) and behaviors (e.g., foot extension, gaping valves), and the lethal limit of critical thermal maximum (CTmax). Wild-caught individuals of *Elliptio pullata*, *Leaunio lienosus* and *Lampsilis straminea* were compared ex-situ. After acclimation, intermittent-flow respirometry was used to measure resting metabolic rate of eight individuals/species during a 10-12 hour exposure to increasing temperature (2°C/hr). Simultaneous observations of eight additional individuals/species were used to determine the temperature at occurrence of behavioral indicators and CTmax. Mean RMRpeak of *E. pullata* (39.57°C) was significantly different (F2,21= 30.73, p < 0.001) from *L. lienosus* (37.30°C) and *L. straminea* (38.15°C). There were no significant differences among species in the temperature at occurrence of behavioral indicators or CTmax. Mean CTmax occurred at 43.2°C in *E. pullata*, 43.7°C in *L. lienosus* and 44.3 °C in *L. straminea*. Our results indicate that variation in thermal tolerance does not occur among these species at or near their lethal tolerance limits; however, variation does occur among these species at RMRpeak and may occur below RMRpeak. Specifically, *E. pullata* showed evidence of adaptive temperature-insensitive metabolism, a thermal response known to occur in some intertidal mollusks. This suggests that tolerance to acute temperature fluctuations could vary among sympatric species, an important implication for the management of this imperiled group. 95 - Use of Essential Oils to Influence Betta splendens Agonistic Behavior

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Lavender essential oil (LEO) is distilled from the plant *Lavender angustifolia* and is commonly used for aromatherapy in humans. In recent years, it has been used on some fish species to test its efficacy as an anesthetic during medical procedures and as a sedative for stress inducing circumstances, like during transport of fish to new locations. In one study, Rezende et al., (2017), used tea tree, clove eucalyptus, and mint oils to investigate the sedative efficacies and effect on stress. Thus, botanical oil extracts have demonstrable effects on stress, but their effect on agonistic behaviors in fish have not been explored. This study aimed to investigate LEO's ability to alter agonistic behavior in male crown-tail Betta fish (*Betta splendens*). To assess the effect of LEO treatment on agonistic behaviors, a pre/post study design was conducted. Select behaviors of male crown-tail Betta fish were observed, untreated, in response to male Veil Tail Betta fish challengers. Next, fish were treated with LEO and recorded for 3 minutes, during which time they were exposed to challengers to measure differences, if any, of these same agonistic behaviors in the context of the treatment. The data indicate that LEO reduced aggression in male crown-tail Betta fish and that this reduction in aggression was partially reversible. Recovery trials completed in the months following initial exposure demonstrated that a month after LEO exposure, agonistic behaviors by treated fish increased, but did not return to Control (pre-treatment) levels. Four months from last exposure to LEO, the behaviors did not return to Control values, indicating that at the treatment level used, LEO may have a permanent effect on fish agonistic behaviors. This study is being extended to determine the dose-response characteristics of active ingredients in LEO, to determine their influence on *Betta splendens* agonistic behaviors.

#### 96 - The effects of hidden nest ectoparasites on the development of a sexually selected trait in juvenile Eastern Bluebirds.

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Many organisms communicate information about themselves to conspecifics through a myriad of visual and auditory styles. These communication styles become signals when a behavior is elicited from the receiver. Because Eastern bluebirds (*Sialia sialis*) retain their natal flight plumage into their first breeding season, they are a model wild organism to study how natal condition affects the development of ornamental traits. Reliable signals of body condition can mediate parent-offspring interactions—perhaps to offset effects of parasites—as well as play a role in sexual selection, when the ornament is perceived as a measure of quality. Our research goal was to elucidate the effects of ectoparasitism by *Protocalliphora sialia* blowflies on bluebird plumage coloration. We experimentally modified parasite presence in nest boxes using pesticide fumigation or a sham fumigation treatment and incorporated nests with natural variation in parasite load. We then monitored the growth of nestlings in the 2021 and 2022 breeding seasons in Watauga Co. NC. Just prior to fledging, we collected feather samples to measure feather brightness, and after fledging, we collected nests to quantify parasite load. We found an interaction between sex and parasite presence, suggesting that parasitized female nestlings grew duller feathers while males did not. Our results suggest that UV-blue structural plumage in female juvenile Eastern bluebirds is at least partially condition-dependent and may help to explain why plumage color predicts parent-offspring interactions and reproductive success in Eastern bluebirds.

#### 97 - Understanding the Relative Importance of Forest Canopy Development and Leaf Litter Accumulation on Soil Evaporation

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Unlike transpiration, soil evaporation represents water lost from a forest that is not used to support productivity. Management approaches that minimize the relative amount of evapotranspiration resulting from soil evaporation can enhance the water use efficiency of a forest stand but relatively little is known about the fundamental controls of soil evaporation. Here, we explore the influence of leaf litter accumulation and canopy shade on soil evaporation because these two factors should have the strongest impact on soil evaporation and can also be modified through forest management. We measured soil evaporation with lysimeters and modified litter accumulation (0, 200, 400, and 800 g m<sup>-2</sup>) and canopy shade (0, 30, 60, and 90%). Both litter accumulation and canopy shade influenced soil evaporation, but the effect of litter accumulation was stronger than the effect of shade. Compared to soils with no litter accumulation and no canopy shade, 200, 400, and 800 g m<sup>-2</sup> of litter accumulation reduced soil evaporation by 16, 48, and 57%, respectively, whereas 30, 60, and 90% canopy shade reduced soil evaporation by 22, 37, and 39%, respectively. Our results suggest that evaporation is a relatively large water loss in early forest stands, and that transpiration may be maximized by increasing, in order of importance, leaf litter and canopy shade coverage.

#### 98 - Investigating intrasexual variation in allocation strategies in the moss, *Bryum argenteum*

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In plants with separate sexes, the sexes often exhibit sexually dimorphic resource allocation patterns due to sex-specific selection pressures to maximize fitness. However, within-sex groups could differ in their allocation strategies, if those strategies lead to similar fitness. Such intrasexual variation (particularly within males) is well-known within animals but unknown in plants. A previous study on water-holding capacity in the cushion-forming moss, *Bryum argenteum*, found evidence of two groups within males but not females. We hypothesized that these two groups have distinct allocation strategies related to growth and reproduction. Using high and low water-holding capacity genotypes within both males and females (for comparison), we grew plants in a greenhouse common garden to investigate whether reproductive investment (gametangia number and size and proportion of tips with inflorescences), protonema (juvenile filaments) vs. shoot allocation, and growth rate correlated with waterholding phenotype. In partial support of our hypothesis, high water-holding capacity genotypes produced fewer gametangia, but this was the case regardless of sex. Males produced larger gametangia and tended to have proportionally more inflorescences than females, but these differences did not correlate with water-holding capacity. There were strong genotypic differences in early allocation to protonema vs. shoots and growth rate, but these were not correlated with sex or water-holding capacity. These results suggest a tradeoff between water-holding capacity and reproductive investment (gametangia number) across sexes, although this may not be a direct tradeoff because previous work provided evidence that waterholding differences begin before sexual maturity. Given that *B. argenteum* females are generally larger, hold more water, and are more abundant than males, we speculate that the distinct group of low water-holding capacity males may have a more ephemeral strategy, investing more in sexual reproduction, than both females and higher water-holding capacity males.

#### 99 - Below-stump coarse root biomass of mature long leaf and loblolly pine trees grown in Mississippi

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Longleaf pine (*Pinus palustris* Mill.) is a majestic species that once was the dominant species in the southern United States. It ranged from Texas to Florida and northwards to Virginia, as well into northern Georgia. European settlers quickly recognized the potential of the species for its high-quality wood and superior naval stores. Due to heavy exploitation, longleaf pine now occupies 3 to 5 percent of its original extant. Loblolly pine has become important plantation species in the Southeast over the last 100 years. Starting in the early 1960's genetic tree improvement evolved

and has become more and more effective particularly in the last 20 years. Silviculture, including genetic improvement and fertilization, has increased loblolly pine plantation at least four-fold since the 1950's. Common wisdom has assumed that that longleaf pine has deeper and larger taproots than the other southern pines. We felled eight long leaf and loblolly pine trees in one stand at the USDA Forest Service Harrison Experimental Forest in Saucier, Mississippi when they were approximately 50 years since planting. Trees were measured for diameter at breast height (dbh) and tree height. Tap roots were excavated by hand in a one-meter square around the base of the tree down to one meter in depth. The remaining tap roots were removed using a backhoe. Tap roots were then separated into lateral roots and were oven-dried and weighed for biomass. There was no difference in the relationship between dbh or height with below stump biomass between the two species thus refuting the common wisdom stated above. This corroborates a more recent study using stands across much of the species' range. Although in the later study coarse root biomass among trees coarse root excavations had a root: shoot ratio almost twice that have been reported for loblolly pine.

100 - Biomarker bacterial taxa from lead-exposed biofilms provides insight into lead-biofilm interactions in premise plumbing pipes

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Biofilm formation within pipes plays a potentially important role in contamination of drinking water in premise plumbing systems. Heavy metal contaminated water within plumbing systems is a global health concern. The interactions between biofilms and heavy metals are poorly studied. To determine whether there are dose-dependent effects of lead exposure on bacterial biofilm communities within high-density polyethylene (HDPE) and cross-linked polyethylene (PEX-A) pipes, a pipe loop was constructed. Biofilms were established for 30 days using municipal water, followed by lead exposure using synthetic water with three different lead concentrations (0 µg/L, 5 µg/L, and 500 µg/L). Biofilms were sampled at three time points (4-, 8-, and 12-weeks post lead exposure). After 8 weeks, lead was removed from the system to query recovery. To investigate biofilm dynamics, total genomic DNA was extracted from biofilms and metabarcoding was conducted. To test if bacterial communities were impacted by time, lead concentration, pipe material, and all possible interactions, a model based multivariate test (package *mvabund* in R) was conducted and demonstrated all factors structure bacterial communities within biofilms. Further, using the ordination-based analyses Axes Weighted Ordination Distances and Procrustes Association Metric, it was seen that once communities shift with lead exposure, these altered communities persist after lead is removed. Analyses of the responses of individual taxa identified several biomarker taxa that are highly overrepresented in high lead conditions (500 µg/L) and include (OTU30 - *Sphingopyxis* sp., OTU43 - *Rhizobium* sp., OTU69 - *Noviherbspirillum* sp., and OTU86 - *Sphingomonas* sp.) and when lead was removed, these taxa did not change in their relative abundances. This work suggests that it may be possible to detect a previous, but not ongoing, lead exposure event based on microbiome monitoring, which will provide a powerful triage tool for health care workers and water distribution management agencies.

101 - Extracts of plants common in South Carolina exhibit antibacterial effects against ESKAPE pathogens

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Due to the increasing number of antibiotic resistant bacteria, there is a growing need to examine alternate forms of medicines. Six of the most notorious antibiotic-resistant pathogens are *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae/Enterobacter aerogenes*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Escherichia coli*, commonly known as the ESKAPE pathogens. This study examines the antibacterial efficacy of extracts made from plants that are common to South Carolina on the ESKAPE pathogens. Methanol extracts of the leaves of *Gleditsia triacanthos* (honey locust), *Osmanthus fragrans* (tea olive), *Rhododendron* subgenus *tsutsusi* (azalea), and *Camellia japonica* (Japanese camellia) were tested with each of the ESKAPE pathogens to determine their antimicrobial properties. Disc diffusions were performed on each of the extracts; however, no significant inhibitory effects were found for any of these plant extracts. Minimum inhibitory concentrations (MIC) of each extract were also determined. During MIC determination, bacteria were incubated in the presence of plant extracts at various dilutions for 20 hours and the resulting growth curves were analyzed. Azalea and Japanese camellia both showed antimicrobial effects against *A. baumanii* at dilutions of 1:2, 1:4, and 1:8. Azalea also showed antimicrobial effects against *E. coli* at dilutions of 1:1, 1:2, and 1:4. Both honey locust and tea olive showed antimicrobial effects against *P. aeruginosa*, while Japanese camellia showed antimicrobial effects against *S. aureus* at a dilution of 1:2. Interestingly, both honey locust and azalea increased the growth rate of *S. aureus*.

102 - Molecular response of *Microcystis* in response to elevated levels of hydrogen peroxide.

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*Microcystis* spp. produce prolific annual blooms in freshwater systems worldwide. The success of these blooms depends heavily on the bloom population overcoming environmental factors such as oxidative stressors caused by reactive oxygen species like hydrogen peroxide. Hydrogen peroxide is the main source of oxidative stress for *Microcystis*. However, unlike most aquatic microorganisms and cyanobacteria, most *Microcystis* genomes do not contain many genes for responding to oxidative stress, including those that encode enzyme catalase. Other cyanobacteria such as *Prochlorococcus* that also lack hydrogen peroxide degrading enzymes have been shown to benefit from partnerships with heterotrophic bacteria. These heterotrophic bacteria that have these enzymes for degrading hydrogen peroxide show higher expression when exposed to hydrogen peroxide potentially providing a benefit to *Prochlorococcus*. *Microcystis* may also receive similar benefits from heterotrophic bacteria in the phycosphere,

where it exchanges metabolites with bacteria. In this study we examined the effect of bloom stage and how both *Microcystis* and its phycosphere microbiome respond to being treated with an elevated (10 $\mu$ M) concentration of hydrogen peroxide during bloom conditions. We also supplemented catalase positive consortium of bacteria isolated from a *Microcystis* bloom in Lake Taihu, China, to test the impact of these bacteria on the oxidative stress response of *Microcystis*. Metatranscriptomic analysis will be used to evaluate the changes in gene expression and community composition. Analyzing the differential expression of relevant genes will also provide insight into how the community is functioning during stress conditions.

103 - Elucidating synergistic mechanisms of immune evasion performed by a multifunctional lipoprotein of the Lyme disease spirochete

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Many bacterial pathogens have evolved arsenals of outer surface or secreted proteins to interact with their host. For example, the Lyme disease spirochete, *Borrelia burgdorferi*, encodes a multitude of lipoproteins on the surface of its outer membrane that serve as adhesins or function to evade the host immune system. Interestingly, many of these lipoproteins exhibit multifunctionality by interacting with more than one host molecule through distinct protein domains. A prototype for a multifunctional *B. burgdorferi* lipoprotein is BBK32. BBK32 binds to the extracellular matrix protein fibronectin via its intrinsically disordered N-terminal domain leading to bacterial adhesion and escape from the host vasculature *in vivo*. BBK32 is also a potent inhibitor of the classical pathway of complement by preventing C1 activation through the direct inhibition of the complement initiating protease C1r via its alpha helical C-terminal domain. Previous infection studies concluded that BBK32 plays a role in the pathogenesis of *B. burgdorferi* in a model of murine borreliosis. While the relative contribution of complement inhibition to this infectivity phenotype is not known, we hypothesized that BBK32 may work synergistically to simultaneously bind fibronectin and complement C1r. In this study we demonstrate that BBK32 binds to both host molecules simultaneously *in vitro*. Using AlphaFold2 and published structural data, we present a model for the ternary structure of BBK32 bound to the 766 kDa C1 complex and 220 kDa fibronectin proteins. Using surface plasmon resonance and flow cytometry assays we assess the functional implications of the BBK32/C1/fibronectin complex on the recognition of opsonized *B. burgdorferi* cells by immune receptors. This study delineates the simultaneous functions of separate host molecule-binding domains of a borrelial lipoprotein, and contributes to the field of bacterial immune evasion by elucidating synergistic roles of multifunctional bacterial proteins involved in host interactions.

104 - Investigating Protein-Protein Interactions between Bacteriophage Cain and its Host *Mycobacterium smegmatis*

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In the face of the antibiotic resistance crisis, bacteriophages have proven to be useful tools for overcoming bacterial defenses in medicine. However, 65% of phage genes have no predicted function, which limits the ability to utilize phages to their maximum potential. In order to effectively manipulate and apply phages therapeutically, we must close this gap in functional knowledge. After using a phenotypic assay to screen mycobacteriophage Cain gene products for interaction with the host cell, *Mycobacterium smegmatis*, a bacterial 2-hybrid (B2H) assay was used to identify which host proteins were being targeted by phage gene products. To achieve this, a complex array of specifically designed expression vectors, test promoters and cell lines, were used to report potential pairwise interactions between phage gene products and the host's proteome. With no predicted function, and significant homology with well-studied phages in the same K cluster, Cain gp55 was the first phage gene to be investigated. Extensive testing of Cain gp55 revealed interactions with host proteins NusA and GntR. Both these bacterial proteins are transcriptional regulators that are thought to play various roles including transcript elongation/termination (NusA), cell motility, metabolism and even virulence (GntR). Another phage gene of no predicted function, Cain gp2, also interacted with several host cell transcriptional regulators, including TetR, AraC and LysR, that are thought to be involved in the regulation of metabolic genes. Identifying these interactions can give us hints about a phage's gene function, and its role in manipulating its host. While the B2H assay gives us a starting place to speculate about function, it is only the first step into identifying gene function for previously undescribed phage genes. Uncovering these gene functions may lead to unexpected advancements in biotechnology and medicine.

105 - Unintended Antibiotic Target: The (Honey Bee) Reproductive Microbiome

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Historically, honey bee queen lifespan was two to seven years, but now 50% of colonies replace queens within six months, doubling the chance of colony mortality. Colonies that are failing usually have queens with low sperm viability and drone (male) sperm counts are highly variable. Tetracycline is the most used antibiotic in beekeeping to treat common brood diseases and has been shown to decrease reproductive development, sperm viability, and overall success in rodent models. Tetracycline is classified as an endocrine disrupting chemical because of its mitochondrial inhibition in reproductive cells. Microbes have been discovered in reproductive tissues of many animals including insects and have been associated with offspring success. To date, no other studies have investigated if honey bees possess a reproductive microbiome or the impact of antibiotic exposure on bee reproduction. We hypothesized that queens and drones possess a reproductive microbiome and that exposure to antibiotics will perturb the native microbial community, which could lead to reduced fecundity. We have shown that tetracycline is highly toxic to drone sperm *in vitro* and confirmed that ingested tetracycline accumulates in honey bee queen reproductive organs (via mass spec). In addition, we have shown that honey bee drones do have a reproductive microbiome by dissecting reproductive organs under sterile conditions, extracting DNA, and

characterizing the microbial community. Lastly, after treating drones with less than field relative doses of tetracycline we have determined that oral antibiotic exposure perturbs the honey bee reproductive microbiome by reducing species diversity and absolute abundance.

106 - An Investigation of the Microbial Diversity and Antibiotic Production Potential of Commercial, Pasteurized Kefir and Raw, Goat Milk Kefir

Anna Braaten, Stephanie Mathews

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Kefir is a probiotic beverage that is made by fermenting milk with kefir grains or bacterial starter cultures. This investigation focuses on antimicrobial properties of bacteria isolated from kefir and their ability to inhibit bacteria that are resistant to conventional antibiotics. Bacterial colonies were isolated from store bought and raw goat milk kefir and then plated against *Bacillus subtilis*, *Escherichia coli*, *Enterococcus raffinosus*, and *Acinetobacter baylyi*. Bacterial isolates from both store bought and raw goat milk forms of kefir showed that antibiotics were produced as seen by zones of inhibition produced. The bacteria were identified using PCR and Sanger Sequencing. Biochemical tests were performed on the isolates and compared to literature searches to confirm their identity. Chemical extraction was used to isolate antibiotics produced by the kefir bacteria. The antibiotics of some kefir isolates impeded the growth of the relative safe ESKAPES when tested against them. Future testing will be used to identify the antibiotics compounds and confirm their efficacy in impeding safe ESKAPES from growing. The antibiotic compounds could potentially be used to combat infectious disease.

107 - Quantification of Environmental Niche for Testing Hypotheses of Ecological Isolation in *Helianthus* Hybrid Species

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Homoploid hybrid speciation occurs when mating between different species leads to morphologically distinct, reproductively isolated offspring without the occurrence of whole-genome duplication (as is seen in allopolyploid hybrid speciation). The success rate of such an event is low due to high competition against the hybrid offspring and selection against individuals with lower fertility, but there are cases in which the F1 or backcrossed offspring inhabit niches too extreme for either parent. This ecological novelty is hypothesized to be one of the causes of speciation among homoploid hybrid plants. Three *Helianthus* species are hypothesized to be the result of hybridization between *H. annuus* and *H. petiolaris* followed by ecological isolation: *H. paradoxus*, *H. anomalous*, and *H. deserticola*. In this project, ecological niche models were developed for all five species to visualize likely suitable habitats and quantify niche shifts. Extensive literature pertaining to the adaptive traits of these hybrids was used to choose environmental layers for model predictions. Environmental layers used in this project included the 35 Climond layers, elevation, and soil data. Occurrence records were obtained from the Global Biological Information Facility (GBIF), Southeast Regional Network of Expertise and Collections (SERNEC), and iDigBio. Occurrence data were sanitized by removing duplicate records, and limited to the sunflower's native range. Environmental and Occurrence data were used to optimize three models, Maxent, Boosted Regression Trees, and Generalized Additive Models. Each of these three models produced separate predictions. A weighted ensemble prediction was generated to maximize contribution of each model according to its predictive accuracy. Finally, multivariate analysis was used to characterize the preferred environment of the parental and hybrid species.

108 - A Study of Previously Undescribed Morphological, Cytological, and Molecular Variation in the Eastern North American Flowering Plant, *Micranthes virginensis*

Tara Hall, Katherine Mathews

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*Micranthes virginensis* (Saxifragaceae) is an herbaceous, flowering plant native to Eastern North America with a range extending from Arkansas into Maine and Canada. This broad range, known from previous studies to contain individuals with varying chromosome numbers and morphological variation outside the current formal description, indicates the need for reexamination of the taxonomy of this species. Multiple putative *M. virginensis* populations with unresolved phylogenetic placement have been previously identified in the escarpment region of Southeastern Appalachia. These populations display intermediate morphological characteristics between *M. virginensis* and its close relative, *M. careyana*, raising the possibility of hybridization. This study explores hypotheses of hybridization and cryptic species within *M. virginensis*, as currently defined. Morphological, cytological, and molecular data were collected from several populations spanning Eastern North America. Anther squashes indicated tetraploidy ( $2n = 38$ ) in the morphologically intermediate escarpment populations and in central North Carolina populations. Populations in other geographic areas were found to be diploid ( $2n = 20$ ). Chromosome counts of *M. careyana* populations also showed diploidy ( $2n = 20$ ), representing the first reported chromosome count for this species. Tetraploidy in the escarpment region, where the distribution of *M. virginensis* and *M. careyana* overlap, could be indicative of hybrid origin for the populations with intermediate morphological characteristics, whereas tetraploidy in central North Carolina could indicate autoploidy, as *M. careyana* does not extend into that region and the morphology of the *M. virginensis* populations in that region do not exhibit any intermediate traits. Principal Component Analyses indicated no discontinuous morphological variation throughout the range of the species apart from the tetraploid escarpment region populations. Should the escarpment region populations or populations in any other geographic area form discrete lineages based on molecular data, this would indicate the presence of additional species within *M. virginensis* that could be formally described in the future.

109 - A Hyb-Seq phylogeny of *Boechera* and related genera using Angiosperms353 and Brassicaceae764 probe sets

Nikolai Hay<sup>1</sup>, Donovan Bailey<sup>2</sup>, Kasper Hendriks<sup>3</sup>, Terezie Mandáková<sup>4</sup>, Kathleen Pryer<sup>1</sup>, Michael Windham<sup>1</sup>

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*Boechera* is a genus of over 100 recognized species that is most diverse in western North America but also occurs in eastern North America and eastern Asia. Over the past two decades, this group has become a model system for biologists studying the genetics and evolution of apomixis, as well as researchers interested in ecological and evolutionary genomics. Although *Boechera* is estimated to be just 2.5 million years old, it has diversified to form 80+ sexual diploid taxa and hundreds of hybrids, making it one of the most rapid diversifications recorded among plants. This incredible rate of evolution, together with rampant hybridization, has played a significant role in the inability of previous investigators to produce a well-supported phylogeny of *Boechera*. Past attempts at a *Boechera* phylogeny included fewer than ten genes and were plagued by very short branch lengths along the backbone and major discordance between plastid and nuclear phylogenies. To fully realize the research potential of *Boechera* as a model genus, a well-resolved phylogeny is essential. To resolve relationships within the tribe Boechereae, we have utilized a HybSeq approach with targeted sampling of sexual diploids. Our sampling includes 99 sexual collections spanning all recognized species groups in *Boechera* plus at least one representative for each of the other eight genera of Boechereae. We have inferred a maximum likelihood phylogeny combining data from two hybrid capture bait sets that cover 1117 unique low copy loci. The resulting phylogeny strongly supports the recognition of two major clades within Boechereae. One of these encompasses nearly all species of western North American *Boechera*, while the other comprises all other genera of the tribe plus a monophyletic group of eastern North American and eastern Asia (*Borodinia*).

110 - Phylogenetic Relationships in *Seymeria* (Orobanchaceae)

Benjamin Gahagen, Caroline Shaw, Sharon Spiess, John Prater

Abraham Baldwin Agricultural College, Tifton, GA

*Seymeria* is a small genus of root hemiparasites in the Pedicularideae tribe of Orobanchaceae. While the pathogenic associations of *S. cassioides* and *S. pectinata* have been studied in fair detail, relatively little is known about the evolutionary relationships within the genus. In 1925, Pennell described 22 species of *Seymeria* and hypothesized evolutionary relationships based on mostly floral features with emphasis on anther dehiscence. Since then, only two species, *S. laciniata* and *S. pectinata*, have been included in family-level phylogenetic investigations. The aim of this project was to elucidate the phylogenetic relationships within *Seymeria* to achieve a better understanding of the biodiversity in Orobanchaceae. Sequence data from three gene regions (rps2, matK, ITS) were obtained from 22 specimens (16 species) and analyzed in the phylogenetic context for maximum likelihood and Bayesian inference. Distinct clades within *Seymeria* were discovered: *Seymeria pectinata*, *S. deflexa*, and *S. cassioides* form a clade; *S. virgata*, *S. laxa*, and *S. decurva* form another; *S. pennellii* and *S. laciniata* another; and *S. texana*, *S. sinaloana*, *S. coahuilana*, *S. bipinnatisecta*, *S. chihuahuana*, and *S. glandulosa* form the last.

111 - Leaf variation in maples: Two cryptic varieties of *Acer saccharum*

Richard Condit<sup>1</sup>, Lawrence Davenport<sup>2</sup>, Bill Finch<sup>3</sup>, Andrew Hipp<sup>1</sup>, Senna Robeson<sup>1,4</sup>, Madelyn Thompson<sup>2</sup>

Cassandra Fink<sup>1</sup>, Shawn Krosnick<sup>2</sup>

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*Acer saccharum* var. *schneckii* Rehder is a southern to midwestern variety of sugar maple that overlaps in range with northern sugar maple, *A. saccharum* var. *saccharum* Marshall. However, the characteristics used to distinguish between the two varieties are neither well defined nor consistent in the literature. The goal of this research is to determine if there are any distinctions in leaf characteristics between the two varieties that can aid in identification of specimens in the field. Based on historical accounts of var. *schneckii*, it differs from var. *saccharum* by having more pubescent petioles, deeper leaf sinuses, and flatter basal lobes of the leaves. With these characteristics in mind, we tested leaf shape in both an herbarium sample and a field sample using a series of 18 leaf landmarks to measure the basal lobes and sinus depths. The herbarium sample consisted of 120 herbarium specimen images of var. *saccharum* and var. *schneckii* from the herbarium database SEINet, chosen from counties where both taxa occur. The field sample included a total of 65 specimens of both taxa which were collected at Paint Rock Forest Research Center in Jackson County, Alabama. In addition to leaf shape, we quantified pubescence of the petioles, veins, and laminae for the Paint Rock specimens. We found that the taxa are not distinct in leaf shape in either sample, contrary to our expectations, but pubescence can be used to distinguish them in the Alabama population. Learning what distinguishes var. *schneckii* from other taxa is the first step in learning more about this taxon and understanding the diversity within the sugar maple group.

112 - Preliminary assessment of the genetic relationships and self-compatibility of species in *Lilium* L. sect. *Leucolirion* in the southeastern United States

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Lilium L. section Leucolirion Wilson, known as trumpet lilies, consists of nine species native to Central and Southeast Asia. These species are characterized by large white flowers and can be either self-incompatible or self-compatible (the latter is rare in *Lilium*). Four species in this section appear to be naturalizing in the southeastern United States (US): *Lilium formosanum*, *L. longiflorum*, *L. philippinense*, and *L. regale*. The reproductive mechanisms of these species are not fully understood in native or naturalized populations. Previous studies debate whether these species are self-compatible as it varies across sites. These species alone are difficult to distinguish due to morphological similarities. Additionally, hybrids have been shown to form readily among species in this section. With many hybrids entering the horticultural markets, it is hard to discern whether species present in the US represent wild species from Asia or hybrids created for commercial use. Therefore, pollination treatments were conducted at three sites in the southeastern US to test for self-compatibility. Buds were covered with mesh bags to exclude visitors before and throughout the duration of anthesis. Pistils were collected 48 h after pollination and visualized under UV fluorescence. We conducted a preliminary molecular analysis of the populations in the Southeast to explore what species and/or hybrids may be present. Leaf tissues were collected from 23 naturalizing populations between June – August 2022. Total genomic DNA was isolated from leaf tissue and the internal transcribed spacer (ITS) region was used to conduct an initial assessment of phylogenetic diversity among populations. This assessment will be followed up by genotyping-by-sequencing (GBS) to identify genetic differences among the populations. Preliminary results suggest the species present in the US are self-compatible. This study seeks to elucidate the relationships among the populations and be used to better understand the species present in the Southeast US.

#### 113 - Locust Pocus: Molecular Tests of Species Delimitation in *Robinia* of the Southeastern United States

Brandon Wheeler

*Western Carolina University, Cullowhee, NC*

*Robinia* L. (Fabaceae) is a genus of trees and small shrubs native to North America, with several species that are of conservation concern within the Southern Appalachians. The genus has been re-circumscribed utilizing morphologically based taxonomic treatments several times in the last century, though no molecular study has focused solely on the genus. I will be focusing on species delimitation between two narrow endemics of the Southern Appalachians: *Robinia viscosa* and *Robinia hartwigii*, utilizing a robust Restriction Site Associated Sequencing (RAD-Seq) dataset and a wealth of species group discovery (i.e. STRUCTURE, PCA, DAPC) and validation methods (i.e. SNAPPER). In addition, I will examine the commonly held hypothesis that *Robinia hartwigii* is a species of hybrid origin, arising from a hybridization event between *Robinia viscosa* and *Robinia hispida* utilizing phylogenetic networks. Finally, I will utilize the RAD-Seq dataset in order to estimate conservationally relevant population genetic statistics in order to identify potential dangers to these rare species.

#### 114 - Clay Models and eDNA are Useful Tools to Identify Predators of Salamanders

Aidan Shaw, Benjamin Holt

*Baylor School, Chattanooga, TN*

Due to the difficulty of observing predator-prey interactions in the wild without human presence disturbing the natural processes, clay models have been a popular method for tracking predation on small species in their natural habitat. A potential drawback of clay model studies is the ambiguity of identifying predators based on bite marks. Using DNA extraction techniques and PCR it is possible to amplify environmental DNA (eDNA) left behind by attackers and identify the predators responsible for attacking models. Molded clay models that resemble Southern Zig Zag Salamanders (*Plethodon ventralis* Highton) were deployed into the field in habitat-accurate sites and monitored for predator attacks for 8 weeks during the winter of 2021-22. Models displaying signs of predator attack were removed from the field and eDNA was extracted from visible bite marks in the clay. Library preparation and High Throughput Sequencing targeting the 12s rRNA was performed and representative sequences were BLASTed against GenBank to determine the identity of the attacker. DNA was recovered from 14 attacked models and identified as belonging to Raccoons, Virginia Opossums, Chipmunks, and the American Robin. The prevalence of this eDNA in several samples and analysis of the attack marks implicates these species as the most likely predators to the models. Our results show clay models can be used in Southern Appalachia to assess patterns of salamander predation, albeit with a few discussed considerations.

#### 115 - Some Salamanders are more Gracious Hosts than Others: Differential Parasite Infections in Two Cryptic, Sympatric Species of Dusky Salamander (*Desmognathus*)

Carlos D. Camp

*Piedmont University, Demorest, GA*

Aquatic vertebrates are prone to infection by a variety of parasite species. For example, the leech *Placobdella biannulata* commonly parasitizes aquatic and semi-aquatic salamanders in the Appalachian Mountains and possibly serves as a vector for blood-parasitic trypanosomes. This leech species is known to infect two cryptic, semi-aquatic salamanders, *Desmognathus amphileucus* (formerly *quadramaculatus*) and *D. folkertsi*, which live sympatrically across the Blue Ridge of northern Georgia. In a comparison of parasite infection, we found that leeches commonly parasitize *D. amphileucus* but rarely *D. folkertsi* from the same streams. In a separate study of metacercarial trematode infection, we found that the trematode *Metagonimoides oregonensis* infects *D. amphileucus* with prevalence rates as high as 100% in some streams. However, we found zero cases of

infection by this trematode in *D. folkerti*, even when from the same streams. Although cryptic with *D. amphileucus*, *D. folkerti* is phylogenetically sister to southern populations of the aquatic *D. marmoratus*, which can also have a high prevalence of leech infection. Moreover, we found metacercarial infections of *M. oregonensis* at the same rate of prevalence in *D. marmoratus* as in sympatric *D. amphileucus*. Our results indicate that *D. folkerti* is unique in its ability to avoid infection by these two abundant parasites, raising the question of why. Because both parasites attach to or burrow through the skin of their hosts, we hypothesize that the skin of *D. folkerti* contains unique proteins that these parasites fail to recognize or actively avoid. Another possible hypothesis is that *D. folkerti* has developed a unique immune system that enables this salamander species to resist infection.

116 - Seeing the Forest for the Trees: Impacts of Woody Understory Removal on Rock Outcrops Used by Crevice-Dwelling Salamanders

Walter Smith

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Concerns related to impacts from vegetation removal around rock outcrops on crevice-dwelling salamanders, particularly those in the Green Salamander (*Aneides aeneus*) complex, have been growing in recent years. Researchers have long advocated for the preservation of forested buffers around rock outcrop habitat as a best management practice for these species, although scant empirical data exists in the literature to support these recommendations. I performed a season-long survey of microclimatic characteristics within crevice refugia at a rock outcrop complex in the Cumberland Mountains experiencing recent Green Salamander declines coincident with woody understory removal for rock climbing development. Crevices in outcrops experiencing adjacent understory vegetation removal were significantly drier and warmer throughout the study period than those at nearby outcrops not impacted by the development of climbing routes, despite an intact forest canopy providing heavy shade at both sites. Impacted crevices were up to three times warmer and six times drier than unimpacted crevices at the same site, suggesting the potential for negative impacts on resident salamanders. These results confirm that understory vegetation removal immediately adjacent to rock outcrops significantly alters the microclimatic profiles of habitats used by crevice specialists, including the interiors of crevice refugia. I will discuss the relevance of these findings to the design of appropriate management guidelines for crevice-dwelling salamander habitat and will outline novel stakeholder partnerships developed using these results and aimed at mitigating impacts to rock outcrop habitat at the aforementioned study site.

117 - Weather Driven Movement Patterns of Semi-Aquatic Water Snakes, *Regina septemvittata* and *Nerodia sipedon sipedon*

Timothy Moore, Nicoleena Storer, Cassidy Gebhardt, Mary Ranson, Zachary Loughman

*Department of Organismal Biology, Ecology, and Zoo Science, West Liberty University, West Liberty, WV*

The secretive manner of snake species causes inherent difficulties in observing and understanding their behavioral choices in relation to their environment. This study hopes to expand upon the understanding of environmentally driven behavioral choices of two semi-aquatic snakes, *Nerodia sipedon sipedon* and *Regina septemvittata*. *Nerodia sipedon sipedon* is considered a dietary generalist, whereas *Regina septemvittata* is a post-molt crayfish obligate. *R. septemvittata* is a federally listed endangered species in Canada and considered imperiled in several US states, and *N. s. sipedon* is considered invasive in some regions of the Western US. Both study species occur in the 4.2 km study site, the North Fork of Short Creek, West Liberty, West Virginia. In 2022, six *N. s. sipedon* and four *R. septemvittata* were tracked via active acoustic radio telemetry. Additionally, traps were utilized for passive collection of specimens. Observations of individuals responses to inclement weather were analyzed to determine the extent of movement before and after a rainfall event. Understanding the behavioral responses to short term environmental changes, such as rain events, of both species can aid in the development of more effective population management within their respective distributions.

118 - New insights into the pollination ecology of the federally endangered mustard *Physaria globosa*.

Christopher Waters<sup>1</sup>, Shawn Krosnick<sup>2</sup>

119 - Response of invasive Japanese stiltgrass (*Microstegium vimineum*) and its associated plant community to management treatments in a residential wetland

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*Physaria globosa* (Desv.) O'Kane & Al-Shahbaz (Brassicaceae) is a federally listed species found in 33 extant populations across middle Tennessee, central Kentucky, and southern Indiana. Only one previous study investigated the pollination ecology of *P. globosa* which found six effective pollinator species. Contrary to other species of *Physaria* in western North America, the previous study also suggested *P. globosa* is primarily pollinated by syrphid flies. Pollinator communities are likely to be more diverse across the range of *P. globosa* and will likely be primarily pollinated by solitary ground-nesting bees. This research is part of ongoing recovery and conservation efforts to document effective pollinator species across the range of *P. globosa*. Floral visitors were collected from five populations of *P. globosa* in spring of 2021 and 2022 representing each EPA level IV ecoregion where it is known to exist. Pollen was removed from floral visitors using an ethyl acetate wash and stained with fuchsin in glycerol to classify and identify effective pollinator species. *Physaria* pollen has a pentacolpate shape making it morphologically unique when compared to other cohabiting angiosperms. Preliminary results indicate the primary pollinators of *P. globosa* are polylectic ground nesting bees representing all

Lissa Legee, Dahja Wright, Josie Richards

*Georgia Southern University, Statesboro, GA*

The annual grass, *Microstegium vimineum* (Japanese stiltgrass) is a problematic invader in wetland ecosystems, spreading rapidly and smothering competing vegetation. It rapidly invaded a residential forested wetland in southeastern Georgia, USA, following removal of five acres of invasive Chinese privet (*Ligustrum sinense*). We measured the response of the invader and the community to several management treatments over the course of a year, comparing key growth and diversity indicators at the end of the first growing season, and at the start and end of the second growing season. We used a 4 x 2 factorial design to compare the impact of stiltgrass removal treatments (mechanical removal with a weed eater, herbicide with RoundupTM, hand pulling, and untreated control) in combination with pine needle litter (added or not added) on stiltgrass growth and recruitment and on plant community characteristics. Treatments were applied in September 2021 during the growing season before the grass set seed. By the end of the first season, all treatment combinations were equally effective at reducing stiltgrass cover relative to controls, signaling successful stiltgrass removal. Additionally, removal treatment had no effect on species richness, suggesting that non-target vegetation was not harmed by any treatment method. In May 2022, the treatment effects held: stiltgrass seedling emergence was 24 times higher in control than in treatment plots. By the end of Fall 2022, only the herbicide treatment effect persisted, supporting half as much stiltgrass cover as in control plots. Additionally mechanical removal plots supported twice as many species as control plots, and species richness was negatively correlated with stiltgrass cover. These results suggest that herbicide treatment is most effective in controlling stiltgrass over the long term, but that species richness is best supported by mechanical removal. Removal over >1 growing season will likely be required to control stiltgrass invasion. 120 - An ecological study of a South Carolina salt marsh

Richard Stalter<sup>1</sup>, John Baden<sup>2</sup>, Joseph Rachlin<sup>3</sup>

<sup>1</sup>*St. John's University, Jamaica, NY*, <sup>2</sup>*US Core of Engineers, Retired, Wilmington, NC*, <sup>3</sup>*Lehman College, CUNY, Bronx, NY*

The present study was conducted on a 7,500-acre salt marsh at Georgetown, South Carolina. Salt marsh vegetation was sampled at 75 sites using the line intercept method. Distribution of vegetation was mapped along an elevation gradient using a surveyor's transit and stadia pole. The vegetation was placed in three arbitrarily determined zones: High Marsh, subdivided into the High High Marsh and Low High Marsh, Middle Marsh, and Low Marsh, divided into High Low Marsh and Low Low Marsh. Soil samples were taken from each vegetation zone and analyzed for chlorinity, salinity, conductivity, and pH. pH is not a limiting factor. Soil salinity, chlorinity, conductivity, and duration and depth of flooding effect salt marsh vascular plant species distribution. The most flood tolerant taxon and most widely distributed taxon was *Sporobolus alterniflorus*. We continue to monitor the effect of wrack on species composition at this salt marsh as well as climate change as it effects sea level rise and the replacement of less flood tolerant taxa by more flood tolerant taxa.

121 - Growing nestlings of an aerial insectivore show differential developmental sensitivity to environmental variation

Sophie Vazquez, Lynn Siefferman

*Appalachian State University, Boone, NC*

Aerial insectivores are a paraphyletic group of specialized predators of airborne insects experiencing dramatic declines in recent years, likely due to cascading effects of declining insect populations. Information on how this group is affected by weather patterns is essential to understanding the outlook for aerial insectivores under future climatic changes. We use insect samples, weather records, and a short-term tree swallow (*Tachycineta bicolor*) reproduction dataset to assess how environmental variation interacts with nesting dynamics to influence offspring growth and success in one breeding season. The spring and summer of 2020 were exceptionally cool and wet for the Southern Appalachian region. Temperature drops can restrict the availability of flying insects, the only food resource for aerial insectivores like tree swallows and their nestlings. We investigated (1) how the local abundance of flying insects was influenced by daily weather conditions and (2) how the availability of food affects fledging success and nestling body mass at different developmental ages. We concluded that insect biomass fluctuates daily with temperature and the effect of food supply is significant for nestling growth, changes as they grow, and positively predicts the number of offspring fledged. Nestlings are most sensitive to changes in food supply at intermediate ages, likely because at this stage they are expending the energy associated with physiological development with the limitations of a partially developed thermoregulation system. Here, we propose that the nesting dynamics of tree swallows in the Southern Appalachians are partially driven by the consequences of changing environmental contexts. 122 - Changing spring phenology and freezing risk for native trees under changing climates

Evan Rehm, Joe Endris

*Austin Peay State University, Clarksville, TN*

Exiting dormancy in the spring represents a tradeoff for temperate trees; maximizing carbon gains while minimizing potential freezing damage to newly formed, cold-temperature sensitive tissues. As winters become milder, trees have advanced their phenology earlier in the year. However, late

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five major bee families in North America. Hymenopterans constituted approximately 65% of all captured floral visitors and 70% of all potentially effective pollinators. Bees of the genus *Hylaeus* were among the most frequently caught pollinators in 2021, but their pollen loads were substantially smaller than bees of the genera *Andrena* and *Ceratina* and the tribe Augochlorini; which together are likely the primary pollinators of *P. globosa*. Future work will utilize environmental DNA (eDNA) from openly pollinated *P. globosa* flowers to detect and monitor the pollinator communities of *P. globosa* across its range. Preliminary experiments have yielded successful extraction, amplification, and identification of pollinator eDNA.

spring freezing events are still occurring at regular intervals, potentially increasing freezing risk to trees. We compare temporal and spatial variation in freezing tolerances of three foundational forest species (*Liriodendron tulipifera*, *Acer saccharum*, *Fagus grandifolia*) and assess short and long-term freezing damage risk. To determine how freezing tolerance varies during spring, we sampled species every 2 weeks from February to May 2022 in Tennessee. To determine how freezing tolerance varies across a species' range, we sampled along a latitudinal gradient (Alabama, Tennessee and Indiana) roughly two weeks before and after the mean last spring frost date.

We found no interspecific differences in freezing tolerance during repeated sampling in spring 2022 in Tennessee. On just a single occasion in mid-April, *A. saccharum* and *F. grandifolia* freezing tolerances were within 2°C to air temperatures. Across all other species and sampling dates, freezing resistances were normally >5°C cooler than air temperature. When compared to long-term data (1900-2022), spring 2022 freezing tolerances were warmer than extreme low temperatures on a given sampling date.

When sampled along a latitudinal gradient, freezing tolerance varied across species with *F. grandifolia* and *L. tulipifera* able to withstand colder temperatures than *A. saccharum*. In just one location (Indiana), freezing tolerance varied before and after the last freeze date. However, contrary to our predictions, species were able to withstand colder temperatures further into spring. These findings suggest important forest species currently have some thermal safety margin against freezing temperatures, but that margin may decrease under future climates in some parts of species' ranges.

#### 123 - Climatic Influences on *Phlox buckleyi* Populations in Shenandoah National Park

Courtney Rosenstadt

*Mary Baldwin University, Staunton, VA*

*Phlox buckleyi*, Polemoniaceae, or the sword-leaf phlox, is a small globally rare wildflower endemic to parts of West Virginia and Virginia. This study evaluates the possible influences of climatic trends and variables on the population change over time of *P. buckleyi*, such as solar radiation, precipitation levels, degree heating days, and changes to wind and humidity levels. This data was gathered from the Remote Automatic Weather Station's digital network from the Sawmill Ridge location, which is in close proximity to *P. buckleyi* populations within Shenandoah National Park. The data has been analyzed independently for outliers before overlaying it with population history of *P. buckleyi*; provided by Shenandoah National Park's 'RarePlant Visit History' account of the species. This is a bank of annual population surveys from two locations within the park, completed from 1993 through 2021. The population history, when evaluated independently, displays two major decreases in population, one in the early 2000s and another in the late 2010s. Multiple regression analyses were used to assess each climatic variable's influence on the *P. buckleyi* population trends over time to elucidate correlations with the two major dips in population. Precipitation, solar radiation, and/or degree heating days are expected to have the greatest correlation while humidity and wind are predicted to have minimal correlation with population trends. 124 - You're Stressing Me Out: Native Tree Response to Extreme Heat

Joseph Endris, Evan Rehm

*Austin Peay State University, Clarksville, TN*

Climate change will force trees to face new temperature regimes they are ill adapted for. One of the many effects of climate change is an increase in number, duration, frequency, and severity of heat waves. Trees have limited capacity to cool themselves and as such, canopy leaf temperatures can often exceed ambient air temperatures. This dichotomy is especially prominent during the period of the day surrounding solar noon. At increased temperatures, photosynthesis slows or stops, decreasing the amount of Carbon uptake and storage. As the upper thermal limits of temperate tree leaves is not well understood, we sought to determine heat thermal thresholds for common forest species to increase our predictive ability about how species will perform under future heat waves.

We chose 11 species of hardwood trees present at a forest lot in Clarksville, TN. We selected species that are common and abundant throughout forests of the eastern United States. Additionally, these species were selected across a range of varying successional traits to examine differences based on shade tolerance. Leaf samples were subjected to heat treatments of 38, 40, 42, 44, 46, 48, 50, 52, 54, 60°C. We then measured chlorophyll fluorescence, a commonly used technique to measure thermal stress, to determine the temperature where heat stress begins but remains reversible (LT<sub>15</sub>), and the temperature at which irreversible damage begins (LT<sub>50</sub>).

We found that three of the species exhibit LT<sub>50</sub> temperatures of ~44°C, which is in line with current observed high air temperatures for our region. Conversely, three other species show no stress until 48°C providing these species with a significant buffer against current and future extreme heat events. However, as climate change intensifies, other factors such as drought may decrease the ability of plants to evapotranspire and cool themselves, exacerbating thermal stressors.

#### 125 - Opportunities to conserve insect biomass and diversity in a region of urban sprawl: A systems approach

Sarah Parsons<sup>1,2</sup>, Elsa Youngsteadt<sup>2</sup>, Peter Willadsen<sup>2</sup>

<sup>1</sup> Duke University, Durham, NC, <sup>2</sup>North Carolina State University, Raleigh, NC

Recent reports of insect biomass decline have raised concern about the future of insect dependent food webs and ecosystem services, such as pollination and nutrient cycling. The causes of insect decline are difficult to pinpoint, but urbanization is a clear threat. Cities, however, are mosaics of land uses, which likely have different effects on different insect groups. Moreover, patterns in non-urban systems predict that urban forests will support the greatest net primary productivity (NPP) and insect biomass. Here we examine links between land use, NPP, and insect biomass and diversity across land uses in Raleigh, NC. First, we developed land use–insect biomass relationships for five urban land use types based on level of

insect biomass. In conclusion, land use categorization and zoning alone may not be good in helping identify areas for insect biomass conservation in cities. Furthermore, NPP-insect biomass relationship in cities may not exist in the way they do in non-urban systems. Our findings are important for understanding how city planners may be able to play a role in helping conserve insect biomass and wildlife both within cities and beyond.

126 - Helping Conservationists Easily Identify *Sarracenia purpurea* var. *montana*, *S. jonesii*, and Their Hybrids in the Field.

Todd Brasseur, Dr. Rebecca Hale, PhD, Jennifer Rhode Ward, Caroline Kennedy

*University of North Carolina Asheville, Asheville, NC*

Hybridization of rare species poses complicated issues for species managers. For example, hybridization and introgression blur the boundaries between species and poses challenges for maintaining species identity. However, gene flow into populations of rare or endangered species can increase genetic diversity, may facilitate adaptation to changing environments, and may facilitate the persistence of threatened populations. Regardless of whether managers want to prevent or facilitate hybridization, it is important for them to recognize the hybrids of the pitcher plants observed in this study. The goal of this project was to identify a quick method that managers could use to identify hybrid pitcher plants in the field. We compared morphology of *Sarracenia purpurea* var. *montana* (Mountain Purple Pitcherplant), *S. jonesii* (Jones' Pitcherplant), and their putative hybrids at two sites in western North Carolina. We conducted principal components analysis on eight pitcher measurements. The plants formed two major clusters that differed significantly in PC1. One cluster was distinguished by tall, narrow pitchers with hoods angled tightly over pitcher apertures, whereas the other cluster was distinguished by shorter, broader pitchers with hoods angled away from the pitcher aperture. These clusters mostly, but not entirely, corresponded to our a priori identification of individual *S. purpurea* var. *montana* and *S. jonesii* plants. Two plants we identified as putative hybrids were intermediate in PC1 score. Although distinguishing genetic identity is the most accurate way to recognize these plants in a lab setting, our morphological measurements provide a promising and pragmatic method of identifying hybrids in the field.

127 - Recent shelterwood harvests promote breeding bird diversity and shrubland birds for less than 10 years in eastern hardwood forests

Cathryn Greenberg<sup>1</sup>, Maria Whitehead<sup>2</sup>, J. Drew Lanham<sup>3</sup>, Joseph Tomcho<sup>4</sup>

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Some bird species are associated with mature closed canopy forest; others require open, young forests created after substantial overstory reduction by natural or silvicultural disturbances. Yet, few are sufficiently long-term to assess changes in breeding bird communities and species abundance over time as young forests mature. We surveyed breeding bird communities most years over a 17-year period (2000-2016) along 200 x 50-m strip transects in 16 mature forest stands (M) and 15 young 2-age stands created by shelterwood-with-reserves regeneration cuts (SW) (1998-1999). Total species richness was greater in SW than M. Total bird abundance was also greater overall in SW than M but did not differ from M by 2011. Abundance of most tested species was greater in SW or did not differ between treatments. Ovenbirds were an exception, with fewer in SW than M; a trend of increasing abundance in SW was evident by 2009. Abundance of indigo buntings, chestnut-sided warblers, Carolina wrens, and eastern towhees was greater in SW than M overall but decreased in SW over time as young trees grew taller. These species differed in their timing of decreased abundance over the 17-year period, suggesting that subtle differences in forest development and structure affected them differently. Our results indicate that early successional forests promote greater abundance and diversity of birds, and suitable habitat for disturbance-dependent species for <10 years post-harvest. Continual disturbances are needed to maintain a forest landscape with a mosaic of age classes and structural heterogeneity that promotes breeding bird diversity at multiple scales.

128 - Establishing Least Cost Corridors for Wildlife Safe Passage On Interstate 40 Near the Wilkins Creek Tract.

Aurora Wynne

*Warren Wilson College, Swannanoa, NC*

Fragmentation of landscapes by roads is a serious threat to wildlife. Roads limit species' dispersal, reducing genetic diversity and resources. These structures are also one of the primary sources of mortality in wild animals. Wildlife-vehicle collisions are a threat to both animal and human life. In Haywood County, North Carolina, the focal area of this study, the danger to wildlife and humans is particularly high within a 28 mile stretch of Interstate 40 (I-40) that runs through the Pigeon River Gorge (PRG), a narrow canyon surrounded on both sides by large tracts of public land. Previous research has shown a strong need for improvement of wildlife corridors and passage points across this section of I-40. A species of particular concern is the American black bear (*Ursus americanus*), which has growing populations in the region and is consequently suffering increased vehicle mortality along I-40. This study assessed potential corridor locations for black bears using path-level movement and least cost

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development impact (greenways, parks, residential, commercial, and industrial areas) by sampling insects with malaise traps and pitfall traps at 30 sites in Raleigh, NC. Second, we measured within-city NPP variation, canopy cover, and temperature within 200m and 1km of sites using geospatial data and correlated this data with insect biomass and diversity data. We hypothesized that land uses with greater canopy cover would have greater NPP and more insect biomass and diversity city-wide. We found that land use was not predictive of canopy cover and was a weak predictor of NPP. Contrary to prediction, hotter sites and sites with less canopy cover had more insect biomass in late summer. Land use was also not predictive of

corridor analyses in a one mile stretch of the PRG. This geospatial analysis was paired with field research and ground truthing to determine best locations for the placement and establishment of new wildlife passage structures, such as an overpass. I-40 divides one of the largest patchworks of protected lands on the east coast of the U.S.. Facilitating connectivity in this area is of high priority for state and national conservation groups. This study furthers the research of Safe Passage: The I-40 Pigeon River Gorge Crossing Project, and develops new novel methods for estimating scientifically informed fine-scale locations for wildlife overpasses within the region.

129 - Partners for Agricultural Innovation and Sustainability: Fostering Collaboration in West Alabama and East Mississippi through Education and Outreach

Caleb Mullins, Aubree Plymale, John McCall

*University of West Alabama, Livingston, AL*

Since 2018, the University of West Alabama (UWA) and the Sumter County (Alabama) Soil and Water Conservation District (SCSWCD) have coordinated a USDA-NIFA funded project with the goal of providing assistance to farmers, ranchers, foresters and other stakeholders in the West Alabama and East Mississippi region. The project employs outreach activities coupled with hands-on assistance provided by trained student interns. From 2018-2021, Project PAIS (Partners Against Invasive Species) focused primarily on invasive species management, with an emphasis on cogongrass (*Imperata cylindrica*) and feral swine (*Sus scrofa*). Through the success of educational and outreach activities, PAIS eradicated approximately five million square feet of cogongrass and eliminated over thirteen hundred feral swine from the service area. In addition, the program fueled growth of student enrollment in conservation and agricultural sciences at UWA. In 2021, the success of PAIS enabled UWA and SCSWCD to continue these efforts through Partners for Agricultural Innovation and Sustainability (PAIS 2.0). PAIS 2.0 increased the geographical area by establishing partnerships with Coastal Alabama Community College and East Mississippi Community College to provide area stakeholders across West Alabama and East Mississippi with the knowledge, tools, and manpower to combat environmental and economic threats. PAIS 2.0 is continuing to provide education and assistance in the battle against invasive species; as of January 2023 the project had eradicated approximately six million square feet of cogongrass and over twenty four hundred feral swine from West Alabama and East Mississippi. In addition, PAIS 2.0 has expanded the scope of the project thematically by aiding stakeholders with issues related to forest management, pollinator support, and soil quality assessment. These efforts have resulted in significant positive impacts for, not only through the eradication of cogongrass and invasive swine but by also raising stakeholder awareness regarding the impacts and management of regional resources.

130 - Effects of Planting Rate on *Pinus palustris* Plantations: Tradeoffs Between Timber and Habitat Quality

Jacks Hausle<sup>1</sup>, Jodi Forrester<sup>1</sup>, Christopher Moorman<sup>1</sup>, Melissa Martin<sup>2</sup>

<sup>1</sup>*North Carolina State University, Raleigh, NC*, <sup>2</sup>*NRCS, Washington DC, DC*

Government cost-share programs are a conservation tool that incentivize the establishment of longleaf pine (*Pinus palustris*) plantations on private land place. These programs place restrictions on planting rates to promote wildlife habitat, as greater tree planting density may reduce canopy openness and herbaceous plant cover that are critical components of habitat for priority species, including gopher tortoise (*Gopherus polyphemus*) and Bachman's sparrow (*Peucaea aestivalis*). While lower planting rates are believed to benefit wildlife use, there is expressed concern among some forest managers that more open grown trees in the plantations will be of inferior timber quality with more and larger horizontal branches and associated knots. We examined the dynamics among understory vegetation structure and composition, longleaf pine stem form (branch density and straightness), and longleaf pine survival by sampling 73 plantations of various ages (5-25 years) and planting rates (653-2445 trees per hectare (TPH)) throughout the southeastern United States. We documented a strong relationship between planting rate and longleaf pine density at time of sampling ( $r=0.69$ ,  $p=0.0001$ ) and strong relationships between stand density and habitat and timber quality metrics. Planting rates greater than 1483 lead to less desirable conditions for wildlife, and smaller individual tree diameters, without improving treeform characteristics. Higher stand densities resulted in lower average tree diameters but greater stand basal area than lower stand density. These higher planting rates led to lower branch density and lower straightness grades than lower planting rates. Canopy openness, bare ground cover, and herbaceous cover all decreased with higher planting rate and stand density. Based on our results, we suggest that lower maximum planting rates (less than 1483 TPH) are appropriate when wildlife habitat is a program objective, and leading to fewer tradeoffs than those that occur from higher maximum planting rates that have only mixed benefits on timber quality.

131 - Insights into genome evolution and host-symbiont relationships from long-read sequencing of a single feather louse

Andrew Sweet<sup>1</sup>, Daniel Brown<sup>2</sup>, Alvaro Hernandez<sup>3</sup>, Kevin Johnson<sup>4</sup>, Stephen Cameron<sup>5</sup>

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Advances in long-read sequencing technology have made it possible to obtain high-quality genomes from non-model organisms. This is a crucial development for understanding genome evolution across many different taxa. However, long-read approaches are typically not feasible for individual small-bodied organisms due to limitations in extracting sufficient genetic material. Recent library construction approaches have reported sufficient material for long-read sequencing, but it is not known if these methods are widely applicable. Here, we sequence and assemble the genome of an individual feather louse – *Brueelia nebulosa* from a European starling (*Sturnus vulgaris*) – using PacBio HiFi and TELL-Seq sequencing. Lice are very small insects, and all previous reference-quality genomes from lice came from pools of hundreds of individuals. We used long reads

to assemble and annotate a draft genome that is comparable in completeness to previous louse genomes from pooled individuals. We also used our data to better understand the demographic history of *B. nebulosa*. Our assembled genome had relatively high levels of heterozygosity (~1%), suggesting consistent outbreeding among populations. This is a surprising result for a highly host-specific parasite like *B. nebulosa*. We also reconstructed the ancestral effective population size, which revealed a recent bottleneck in the species. This could be consistent with the introduction of its host (Starling) into North America in the 19<sup>th</sup> century. Finally, because we used the entire louse body to extract DNA, we could assemble and annotate the genomes of both the primary endosymbiont and mitochondria. We found strong evidence for the presence of a *Sodalis*-like endosymbiont and a single circular mitochondrial genome, both of which are consistent with other species of *Brueelia*. Overall, our study is a valuable step for increasing the availability of genomics for small-bodied, non-model species.

#### 132 - Therapeutic Treatments for Epilepsy in *Drosophila Melanogaster*

Francois Desautels, Andrew Bellemer

*Appalachian State University, Boone, NC*

Epilepsy affects approximately 50 million people worldwide. Epilepsy is characterized by seizures, which is the result of abnormal electrical activity within neuronal networks causing convulsions, muscle spasms, and unconsciousness. *Drosophila melanogaster* is an excellent model organism for studying epilepsy because 60% of their genes have a homologous human gene, they have short generation periods, and they are powerful genetic tools that allow us to investigate the molecular mechanisms of nervous system function. In this study we examined possible therapeutic treatments for epilepsy using a “bang-sensitive” seizure-prone mutant fly line (Parabss1) which contains a gain-of-function mutation within the alpha subunit of its sodium voltage-gated channels. Previous literature has shown that anandamide (AEA) and its metabolite arachidonic acid (AA) reduce the proportion of parabss1 flies that seize following mechanical stimulation, but no one has asked what their effects on seizure severity may be. We define seizure severity as the time it takes the flies to recover from a seizure. We demonstrate that AEA, its metabolite AA, and fish oil reduce seizure severity in parabss1 flies showing promise as possible therapeutic treatments for epilepsy. The exact mechanisms by which these manipulations reduce seizure severity is not fully understood and are still under investigation within the lab. Additionally, we are currently testing AA metabolites and drugs that are inhibitors of enzymes that metabolize AA to see how they may affect seizure severity.

#### 133 - Unraveling protein interactions involved in chloroplast group IIA intron self-excision in rice.

Michelle Barthet

*Coastal Carolina University, Conway, SC*

An essential step of gene expression is the removal of introns from precursor RNAs in order to generate the correct mature RNA template for RNA function or protein translation. Lack of proper intron excision may result in aberrant RNA and protein products leading to subsequent loss of organelle or cell function. In the nucleus of eukaryotic cells, introns are excised by a large protein and RNA complex known as a spliceosome. Prokaryotes, however, utilize maturases, enzymes that target single introns (e.g. maturase A binds to intron A, maturase B binds to intron B), typically group II introns, and aid in folding the introns into the catalytically active structure for self-excision. The chloroplast of plant cells is the result of an endosymbiotic event and, as such, has characteristics of both the prokaryotic ancestor and the eukaryotic host. Only a single maturase, MatK, is known to be encoded in the plastid of plant cells. MatK, unlike its prokaryotic relatives, binds to seven group IIA intron targets. Several of these intron targets reside within precursor RNAs for essential components of plastid function. Several nuclear-encoded proteins also are implicated as intron excision factors for the same group IIA introns as MatK. These data suggest a divergence in chloroplast maturase function from its prokaryotic relatives to a role more similar to the nuclear spliceosome. We demonstrated using *in vitro* activity assays that MatK alone was not sufficient to promote self-excision of some chloroplast group IIA introns. Further, findings from co-immunoprecipitation assays supported MatK-protein interactions with nuclear-encoded proteins known to play a role in chloroplast group IIA intron self-excision. These data support our hypothesis that multiple protein interactions are required to promote efficient chloroplast group IIA intron self-excision and suggest the formation of a splicing complex in the chloroplast of plant cells.

#### 134 - Drought stress response revealed by differential gene expression for two African acacia species

Ellen Weinheimer, James Pease

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How species respond to stress can reveal the selective pressures that shaped their evolution as well as unique interspecific vulnerabilities and adaptations. Genetic sequence variation is examined to identify the relevant markers that correspond to an organism's ability to respond to stress, but transcriptional variation elucidates the gene pathways and molecular mechanisms associated with the response. We investigated differential gene expression in response to *in vitro* drought stress within two African acacia species, *Vachellia tortilis* and *Vachellia robusta*, to explore how this specific stress factor has shaped adaptive strategies. African acacias are an ecologically important group in savanna ecosystems, and these two species are distributed on opposite ends on an aridity gradient, providing a promising system for investigation. Given acacias' ability to survive under harsh environmental conditions and the broad phenotypic diversity observable within the African acacia clade, we predict that molecular mechanisms across a drought time course may be divergent between species. Identifying the interspecific response of African acacia seedlings to drought will inform our understanding of how trees respond to this stress at the level of transcription as well as the adaptive mechanisms evolved under a variable environment.

135 - Stilbene compounds reduce cell motility of triple-negative breast cancer cells through regulation of epithelial-to-mesenchymal transition related genes

Dr. Lyndsay V. Rhodes, PhD

*Florida Gulf Coast University, Fort Myers, FL*

1 in 8 women in the United States will develop invasive breast cancer in their lifetime. Triple Negative Breast Cancer (TNBC) is categorized as an aggressive subtype of breast cancer that lacks expression of estrogen receptor, progesterone receptor, and non-HER2 amplified. This subtype also disproportionately affects minority and younger women, with a high rate of distant metastasis and overall poor prognosis. As TNBC lacks targetable receptors, patients must rely on general chemotherapies, surgery, and radiation. TNBC also has a high rate of de novo and acquired drug resistance, highlighting the need for identification of alternative therapies.

Stilbenes are defense compounds produced by plants and have been popularized in recent years based on observed health benefits. Resveratrol, the most widely studied stilbene, has been revealed to possess significant antioxidant and anti-cancer effects. We have previously published data demonstrating the anti-cancer activity of resveratrol stilbene analogues in reducing TNBC viability through the up-regulation of apoptosis. In this study, we screened a library of resveratrol stilbene analogues at sublethal doses and identified several compounds that altered the morphology of TNBC cell lines (MDA-MB-157, MDA-MB-231, BT-549) to a more epithelial-like phenotype. Treatment of TNBC cells with select stilbene compounds significantly reduced cell migration and invasion capacity using *in vitro* transwell assays. Realtime PCR revealed stilbene regulation of epithelial-to-mesenchymal transition (EMT) gene expression, suggesting the change in morphology and cell motility is due, at least in part, to regulation of EMT. Taken together this data indicates that stilbene compounds have anti-metastatic properties to reduce cell migration and invasion through regulation of EMT associated genes. Although further study is needed to identify the mechanism of action, our data indicate that select stilbene compounds hold promise as novel therapeutics to target metastatic spread of aggressive TNBC.

136 - Triple Negative Breast Cancer: Investigating the role of the Hippo Pathway in Cellular Proliferation

Victoria Peterson

*Mary Baldwin University, Staunton, VA*

Triple Negative Breast Cancer (TNBC) is a type of breast cancer that does not express estrogen, progesterone, or HER2 receptors. TNBC is the most aggressive form of breast cancer with a low survival rate and with treatments that cause patients with this type of cancer to suffer. Estrogen, progesterone, and HER2 receptors help control cell growth and are also the most common targets of breast cancer therapy. The lack of these receptors makes it hard to control cell growth in TNBC and adds to the difficulties of manufacturing TNBC therapies. The Hippo pathway is a pathway that controls organ size through the regulation of cellular proliferation and apoptosis. YAP is a transcriptional coactivator that works to induce the expression of genes that promote cell proliferation, survival, and migration. Population-based genomic studies have shown that activating Hippo signaling correlates with TNBC breast cancer risk, whereas high YAP expression is associated with poor survival in breast cancer patients, suggesting Hippo/YAP plays a critical role in breast cancer progression. Staurosporine is a kinase inhibitor that affects the activation of the Hippo Pathway in breast cancer cell lines. This research aims to evaluate an opening for control of cellular proliferation in TNBC cells through the Hippo pathway. Can Staurosporine identify a target for therapy that reduces cellular proliferation? Here we examine the activation of the Hippo pathway in MDA-MB231, a TNBC cell line, with Staurosporine administration. RT-PCR (real-time) will be used to evaluate if the expression of CCN1 and CCN2, genes regulated by the Hippo Pathway, are affected by the Staurosporine treatment and correlate with changes in cell proliferation. Because CCN1 and CCN2 are cell cycle regulatory genes, these experiments would provide information on the control of the Hippo pathway and potentially provide another chemotherapeutic option that directly influences cell proliferation in TNBC cells.

137 - Pilot Data from a Coordinated Five-Course Experiential Learning Career Preparation ePortfolio Project

Becky Fiorillo, Karen Perell-Gerson, Victoria Bali, Caroline Hanson, Julia Shearer, Wendy Dustman, Sharon Keller, Rebecca Cooper, Ramata Cisse, Whitney Smith, Charmita Burch, Neville Forlemu, Rebecca Kalman, Xiaoping Li, Thomas Lilly, Gillian Rudd

*Georgia Gwinnett College, Lawrenceville, GA*

This pilot is an ePortfolio project for pre-nursing students to develop artifacts reflecting experiences obtained across five pillars (critical thinking, career exploration, communication, leadership, connection) for success in healthcare professions. These experiences occur through the five prenursing science courses (CHEM 1151K/1152K, BIOL 2451K/2452K, BIOL 2516K). The objectives are 1) enhance retention & progression toward graduation through realistic career goals; 2) facilitate students' use of artifacts to evaluate their competencies; and 3) coordinate students' application of skills learned in courses to chosen career paths.

138 - Promoting Diversity in STEM through Biodiversity

Deirdre Gonsalves-Jackson<sup>1</sup>, Victor Townsend,Jr.<sup>2</sup>

Carrie Koenigstein

*Anderson University, Anderson, SC*

University science departments working with citizen science programs provide key opportunities for partnerships, lab class experiences, community service and undergraduate research opportunities. Anderson University has a long history of working with the South Carolina Adopt-a-Stream program which has benefitted both organizations. Adopt-a-stream protocols have been the basis of lab activities with both science major and general education courses. The protocols have also been used as the basis for several undergraduate research projects. Some projects have solely focused on the published Adopt-a-Stream protocols while others have utilized additional techniques beyond those represented in the citizen science program. The work at the university has contributed to data collection by Adopt-A-Stream as well as provided additional awareness of the citizen science organization. Science professors and citizen science organizations also tend to share overlapping interests and areas of expertise that can lead to grant possibilities, community outreach opportunities for the faculty and students, and beneficial networking between the organizations. 140 - iNaturalist: The Indefinitive Dummies Guide to a Smartphone App

Shem Unger<sup>1</sup>, Mark Rollins<sup>2</sup>

<sup>1</sup>*Wingate University, Wingate, NC*, <sup>2</sup>*Wingate University, Wingate, NC*

When conducting biological surveys or engaging students in field biology, proper identification of taxa can present challenges to even well-seasoned naturalists. An emerging smartphone application, iNaturalist, continues to provide an avenue for identifying and documenting biodiversity while concomitantly allowing users to contribute their observations on floral and faunal species as citizen scientists. Surprisingly, few studies have incorporated this application in their research or surveys. To this end, we evaluated the use of iNaturalist to identify species, engage undergraduates in outdoor laboratories, document animal behavior, and as a research database for exploring biological trends. Overall, we find iNaturalist works well to identify most taxa, is highly accessible to first-time users (like students), and can teach important biological concepts. We recommend incorporating the iNaturalist application for outdoor laboratories, biological surveys that use citizen science, and even outreach programs. Furthermore, iNaturalist can serve as an informative tool to both collect and analyze readily available data on local species biodiversity.

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At Virginia Wesleyan University (VWU), there is an initiative underway to establish outreach programs with local schools to attract students to STEM. One initiative is a dual enrollment program designed to recruit and provide talented students with early exposure to STEM and opportunities for study at the VWU campus. This study presents the results of a three-year dual enrollment partnership with local secondary schools designed to promote diversity in STEM. The program consisted of a three-week summer course, The Diversity of Life, taught on the VWU campus by biology faculty. The course involved intensive, rigorous, hands-on learning in the laboratory utilizing the scanning electron microscope, as well as field experiences on the VWU research vessel to the Chesapeake Bay and Atlantic Ocean and nearby beaches such as First Landing State Park. The program was successful in recruiting program participants from diverse racial and ethnic backgrounds, with 55% of participants from underrepresented groups and 81% of participants being female. This level of matriculation of minorities and women is higher than that of the University at 44.8% and 58%, respectively. Moreover, academic achievement of participants was high with 89% earning a transferable grade to colleges and universities. The results of the VWU dual enrollment program therefore serve as a model for how secondary schools can collaborate with area institutions of higher learning to create similar partnerships that increase participation and retention of underrepresented groups to STEM.

139 - How can undergraduate science programs benefit by working with state Adopt-A-Stream Organizations?

## ASB Lightning Talks

### LT 1 - Climate Change Effects on Small Mammal Communities of the Southern Appalachian Mountains

Jenifer Mallinoff<sup>1</sup>, Marketa Zimova<sup>1</sup>, Corinne Diggins<sup>2</sup>

<sup>1</sup>Appalachian State University, Boone, NC, <sup>2</sup>Virginia Tech, Blacksburg, VA

The Southern Appalachian Mountains host a rich diversity of small mammals. Many of these species are rare or endemic, including the federally listed Carolina northern flying squirrel. Despite the ecological significance of the Southern Appalachians, we do not know much about the small mammal communities within them nor do we know how these wildlife species might respond to the changing climate. My thesis aims to document the diversity of small mammal communities across an elevational gradient in the Southern Appalachian Mountains and determine the sensitivity of each species to climate change. I am using a combination of live-trapping and various non-invasive survey methods, including acoustic monitoring and remote camera trapping.

### LT 2 - Exploring genetic diversity in yaupon holly (*Ilex vomitoria*), an American native beverage plant

Ben Long, Jeffery Bennetzen, James Leebens-Mack

*University of Georgia, Athens, GA*

Despite their unique physiology and ubiquity in human culture, beverage plants have received little attention from the genetics community. Yaupon holly (*Ilex vomitoria*) is a southeastern American native plant that has been used by local Indigenous peoples to brew a caffeine-containing infusion for thousands of years. Its unique phytochemistry, wide natural range, and observed variation in secondary metabolite profiles would make it an excellent system to study how the production of beverage-relevant chemicals is shaped by evolution, but the genetic architecture of these traits remains unknown. To address this gap, I am genotyping 300 individuals sampled throughout the species' native range and propagating cuttings from each to construct the first duplicated diversity panel for yaupon holly. A forthcoming chromosomal genome assembly for *I. vomitoria*, together with metabolomics data, will enable genome-wide association studies and quantitative trait locus mapping for caffeine production. Ultimately I will characterize diversity in caffeine biosynthesis pathway genes, as well as genetic and genomic components of variation in caffeine production and other important horticultural traits. Completion of these objectives will bring us a step closer to unraveling the complex relationship between ecology, genetics, and secondary metabolite diversity.

### LT 3 - MAGA: Make Antibiotics Great Again!

David Giles

*University of Tennessee Chattanooga, Chattanooga, TN*

Antibiotic resistance is now a given, an inevitable adaptation by bacteria that creates roadblocks for treatment options. The World Health Organization and National Institutes of Health have encouraged the exploration of new strategies for targeting pathogens. Most new treatments for bacterial infection are originating from chemical adjustments to existing antibiotics and surveying small molecule libraries for efficacy. The Giles laboratory has demonstrated that polyunsaturated fatty acids (PUFAs) can modify membrane phospholipid composition in Gram-negative bacteria. A novel strategy is emerging, wherein the 'Trojan Horse' effect of PUFAs renders bacteria more susceptible to antibiotic treatment. Intriguingly, several Gram-negative bacteria of medical importance display altered susceptibility to antibiotics when PUFAs are administered concurrently. Thus, the therapeutic combinations, with low expected toxicity and almost limitless chemical variation, could represent viable synergistic treatment options for a variety of bacterial infections. We may have a way to polish those silver bullets!

### LT 4 - NEON: Open data for examining the impact of wildfires and beyond!

Margaret Cumberland

*National Ecological Observatory Network (NEON), Oak Ridge, TN*

Data from the National Ecological Observatory Network (NEON) is bursting at the seams! This massive effort to produce long-term, open access ecological data is in full stride, with hundreds of data products to choose from! NEON data cover a wide range of biological subject areas and are collected from 81 field sites, 17 of which are located in the Southeast. This lightning talk will provide a brief look at herbaceous and woody data from Great Smoky Mountains National Park and how it reveals a clear trend in vegetation recovering from the 2016 Chimney Top's Fire. This is a small fraction of the amazing analysis already taking place with more to come!

### LT 5 - Creating Custom Webpages and Apps to Enhance Learning

Joni Criswell

*Anderson University, Anderson, SC*

As higher education continues to adapt to the seismic impact of COVID-19, the technology ecosystem used for instruction and learning continues to be examined and reevaluated. With the preference of both students and faculty to maintain the flexibility of hybrid and blended learning, quality digital resources are needed to help support this method of teaching and learning. Although the market has been flooded with new digital and virtual resources many of them are not cost-effective, lack rigor or provide an overwhelming amount of unnecessary material. This has been especially true for virtual resources focused on general biology courses. In the past few years, the science education field has increasingly become digital in nature with some schools only offering online general biology courses. This has led to the development of several virtual lab based apps and web pages that are currently available to the public, however, many of these are focused on medical students or require payment to access. There seems to be a need for digital learning resource that specifically focus on what the instructor covers. This has lead to Biology department at Anderson University to focus on projects that develop a digital learning resources for students either in the form of an app or cloud-based web page.

LT 6 - Tracking the source of sewage contamination in a mountain stream

Kayla Deel, Isabella Maggard, Teresa Brown, Bruce Cahoon

*University of Virginia's College at Wise, Wise, VA*

Karst springs are biodiverse hydrologic features that are both under-studied and under-sampled. Due to the direct connections between sinkholes, sinking streams, and underlying cave systems, these springs are often impacted by human land use activities on the surface. A preliminary biodiversity survey of the microbiome inhabiting Mill Creek Springs (Southwestern Virginia) was conducted in 2021 using eDNA metabarcoding, and bacteria associated with sewage were the predominant microbes detected. The goal of this 2022 project was to determine if any specific sources of fecal contamination could be detected in groundwater upstream of Mill Creek Springs. We hypothesized that the source of contamination may be septic systems serving residences and schools and/or the infiltration of agricultural runoff. Five sites upstream from Mill Creek Springs were sampled using two techniques – water filtration and glass beads suspended in the water to collect periphyton. The highest concentration of *Escherichia coli* was detected from Mill Creek Springs using IDEXX Colilert tests, however, each of the upstream samples were elevated above levels deemed safe for recreational contact. Environmental DNA metabarcoding indicated the microbiomes of all the sites were similar, with no significant differences. While these results did not allow us to determine the source(s) of the fecal contamination, we cannot rule out the additive contributions of multiple sources of surface water and groundwater that enter the cave system already contaminated with feces, most likely due to shallow soils and poorly-maintained septic systems.

LT 7 - Assessing Bacterial Diversity in Local Aquatic Environments: A Metabarcoding Study of Polluted and Unpolluted Surface Freshwater Bodies and Adjacent Sediments

Jacqueline Valiente<sup>1</sup>, Analuz Nieves<sup>1</sup>, Tammy Laberge<sup>2</sup>

<sup>1</sup>*Miami-Dade College, Miami, FL*, <sup>2</sup>*Miami Dade College, Miami, FL*

In the environment, a wide range of organisms, including plants, animals, and microorganisms, leave behind genetic material termed environmental DNA (eDNA). Environmental DNA can be collected in local habitats, for example in air, water, sediment, snow, soil, etc. In our study, aquatic samples were obtained from a diversity of freshwater bodies that were collected from previously polluted and traditionally clean sampling sites (estuaries, tributaries, man-made waterways, lacustrine environments, and lentic environments). A multifaceted approach was employed for the collection and filtration of water samples, which involved the use of two different filters with different pore sizes (0.22 µm and 0.45 µm). Further, in an exploratory effort, sediment samples were collected from the surrounding areas of the previously obtained water samples. Following collection, samples were processed for DNA extraction and quantification analyses. Recently, the V4 hypervariable region of the prokaryotic 16S ribosomal RNA (rRNA) gene was amplified by polymerase chain reaction (PCR) using a universal primer set (515f and 806r). PCR and gel electrophoresis provided information about the amplicon size and quantity of the amplified DNA fragments. As a result of our analyses, our current research focuses on sequencing approximately thirty candidate PCR amplicons using next-generation sequencing and using bioinformatics tools to perform DNA metabarcoding. Our goal is to determine the local bacterial diversity and abundance present in each sample. Studying the diversity and abundance of freshwater local microbiomes is crucial as they play a vital role in ecosystem functioning, impact larger organisms' health and survival, and aid in identifying environmental stressors and conservation efforts.

LT 8 - *Packera dubia* (Spreng.) Trock & Mabb., a Species Complex of a Dubious Nature

J. Brandon Fuller

*University of North Carolina, Chapel Hill, NC*

*Packera* (Asteraceae) is one of the most species-rich genera in North America, with 73 named species and numerous subordinate taxa. *Packera*, along with other members of the Senecioneae, is undergoing rapid and recent diversification, which has led to taxonomic difficulties as many of the species have a wide degree of overlapping morphologies. Members of *Packera* have eclectic distributions with many narrow endemics, 1000+km disjunctions, or contiguous across the continent. The center of diversity of *Packera* is the southwestern US and co and is where the bulk of named taxa reside. Historically, there were many named species, varieties, and forms of *Packera* east of the Mississippi River, but lumping based on scant

evidence and justification in the 20th century left eastern North America with only eight named species and no subordinate taxa. Since the 1990s, the use of genetic information and renewed interest in the plant diversity of Eastern North America has seen the naming of new species and varieties, including a single site endemic of serpentine barren of western North Carolina, *P. serpenticola* Boufford, Kartesz, S.H. Shi & R. Zhou. Botanists across eastern North America have also discovered several entities that do not fit within the concepts of known species and await taxonomic description. One such species complex that warrants attention is *P. dubia* (Spreng.) Trock & Mabb. *Packera dubia* has a disjunct and fragmented range with four main areas of distribution: Atlantic coast, Piedmont granite flat rocks, Gulf Coastal Plain, and Texas-Arkansas prairies. Preliminary evidence based on whole genome skimming, morphology, and ecology supports four possible taxa with a shared ancestry, two species, each with two varieties. Further population sampling for genetic and morphological data is needed to justify splitting the species complex, and this holistic approach could help clarify other species complexes in eastern North America.

#### LT 9 - Syrphidae of the Southern Appalachians: Diversity, Distribution, and Foraging Behavior

PJ Coleman<sup>1</sup>, Paul Super<sup>2</sup>, Jennifer Geib<sup>1</sup>

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Flies of the family Syrphidae are a widespread and diverse group of dipterans consisting of over 6000 species worldwide. Although most of the buzz surrounds bees, syrphid flies are thought to be equally important pollinators of native plants. Additionally, the larval stages of many species are predatory towards common plant pests such as aphids, making them promising biological control agents in both natural and agricultural settings. Despite the dual ecosystem services offered by their holometabolous lifestyle, studies exploring their diversity, distribution, and efficacy as pollinators are lacking. My study seeks to describe the diversity and distribution of Syrphid Flies in the Southern Appalachians along the Blue Ridge Parkway, determine best inventory practices, and explore aspects of their foraging behavior relevant to their efficacy as pollinators. A citizen scientist inventory collected Syrphid Flies through passive trapping (2019, 2020) as well as active netting efforts (2021) at 64 sites alongside the Blue Ridge Parkway in North Carolina and Virginia. I will use this data to map the distribution of various genera and compare the efficacy of inventory methods. Foraging choices such as floral preferences and constancy through behavioral observations both in the field and using artificial arrays in mesh tents. Gaining a baseline understanding of hoverflies and their potential as pollinators may prove beneficial in supporting pollination services throughout natural, recreational, and agricultural assets, particularly as many other native pollinator species begin to experience declines in abundance.

#### LT 10 - Repurposing Citizen Science Data To Understand Behavior and Ecological Interactions

Linnea Lyons, Matt Heard

*Belmont University, Nashville, TN*

Studying how organisms interact with each other and their environment can pose many challenges. Some of the major challenges include laborintensive field work, working with cryptic or secretive species, and navigating private property. In this lightning talk, we examine how we can use repurposed citizen science data from large-scale datasets like iNaturalist and the Global Biodiversity Information Facility (GBIF) to quantify and better understand behavior and ecological interactions at local, regional, and continental scales.

#### LT 11 - New Framework for Communities to Detect Pharmaceuticals in Wastewater

Rachel Barkley

*North Carolina State University, Raleigh, NC; Sequence Inc, Raleigh, NC*

Pharmaceuticals are emerging as contaminants of concern. In particular, antibiotics have a quantifiable, negative impact on the environment which ripples out to everything from human pathogens to ecosystem degradation. In addition, the local communities responsible for the public health of their residents do not have the resources to monitor and regulate every pharmaceutical. This lightning talk will provide a new framework that communities can use for starting a monitoring program. The framework proposes that communities focus on pharmaceutical inactive ingredients—that is, the other ingredients in any drug to make it safe and effective for people. Many inactive ingredients are not contaminants of concern but studying their fate and transport through the environment can give insight into where harmful drugs could be going. Inactive ingredients are helpful proxies for assessing what is in wastewater today and what could appear in wastewater tomorrow. This talk will include resources that audience members can take back to their home institutions.

#### LT 12 - Awards in Science are evil.

Patrick Cain

*Georgia Gwinnett College, Lawrenceville, GA*

Science is a noble pursuit, in and of itself. On the forefront, basic science guides the path for focused, high-quality inquiry. This approach to science often goes unrecognized by the awards we commonly see given at venues ranging from local conferences to the illustrious Nobel Prize. Here, I

argue that awards in science are counter to the spirit of academic inquiry, and I provide examples of alternatives to such exclusionary forms of recognition.

LT 13 - Putative history of the boreal-arctic peatmoss *Sphagnum warnstorffii* Russow in the Southern Appalachians

Karn Imwattana

*Duke University, Durham, NC*

*Sphagnum warnstorffii* Russow is a peatmoss that is commonly found in boreal and arctic peatlands. Its occurrence in the Southern Appalachians is very rare. In this study, *S. warnstorffii* is reported from one site in Watauga county, North Carolina, possibly one of the southernmost sites for the species. It is possible that the Southern Appalachians might act as a glacial refugia for the species during the ice age. However, phylogenomic analysis has shown that the plants were nested within the clade of eastern North American *S. warnstorffii*. Furthermore, plants from two wetlands 500 meters apart within the site were genetically identical. This result suggests that the population of *S. warnstorffii* within the Southern Appalachians is of recent origin, refuting the hypothesis that the Southern Appalachians acts as glacial refugia for *S. warnstorffii*.

LT 14 - Revisiting the vascular flora of the Francis Beidler Forest in Four Holes Swamp, Berkeley and Dorchester Counties, South Carolina

Daniel Koenemann

*Claylin University, Orangeburg, SC*

The Francis Beidler Forest in the South Carolina low country represents one of the largest primary black water cypress-tupelo swamps in world and has been managed by the Audubon Society since the late 1920s. The reserve has grown over the years, and now encompasses an area of over 18,000 acres. This large, protected tract of land is home to valuable ecosystems that are in need of monitoring. A survey of the plants in the reserve was conducted shortly after the reserve became open to the public in the late 1970's. This initial flora of the reserve focused exclusively on the 1800-acre undisturbed "core" of the forest and swamp. We are approaching 50 years since this initial survey of the reserve. Enough time has passed to use this initial survey as a baseline to track floristic changes in the reserve, should they have occurred. Therefore, I am conducting a second floristic survey of the Francis Beidler Forest. This second survey looks to accomplish two things. The first goal of the second survey is to repeat the first flora, by sampling intensely in all of the same areas. I will then compare the list of plants from the 1970s and the list of plants from the 2020s. I will look particularly at the identities of floristic additions and losses in an attempt to understand changes in the flora of the forest and swamp. The second goal of the second survey is the expansion of the sampling area to include a larger percentage of the currently 18,000-acre reserve. While this larger land area has been substantially more disturbed than the core of the reserve, understanding the floristic composition of this area is nevertheless important. I will discuss preliminary results of the collecting to date.

LT 15 - Genetic Analysis of Distinct Chestnut Weevil Populations in the Northeastern United States

Cassius Guthrie, Graham Reynolds, Camila Filgueiras

*University of North Carolina at Asheville, Asheville, NC*

The American chestnut was a tree of vital importance to the forests of the eastern United States before the accidental introduction of the pathogenic chestnut blight which decimated the population. Through efforts to reestablish the historic American chestnut tree, an unwanted pest has also regained its prevalence. *Curculio sayi*, the lesser chestnut weevil, is a specialized seed predator that not only damages the nuts through the oviposition of eggs but also through its ability to introduce toxic fungi to the plant. As the population of its host plant has been increasing, *C. sayi* populations have re-emerged at an unnerving rate. Developing a strategy of population control of *C. sayi* stands to directly support the chestnut industry and the successful reintroduction of this historical tree. Trapping and monitoring efforts have revealed divergences in reported phenologies in Northeastern *C. sayi* populations as well as distinct morphological differences that indicate a possibility of at least two diverging species. Currently, the origin of weevil reintroduction and human impact on dispersal are unknown. Therefore, characterization of *C. sayi* phenology and the Northeastern population structure would directly improve the efficacy of pest management efforts. The goal of this project is to examine population dynamics of *C. sayi* through DNA barcoding analysis to establish sources of origin, rates of population dispersal, and human impact on population spread. Combined with previously collected data on chestnut weevil phenology, the DNA analysis I am performing will provide insight into population variation as well as potential species divergence of *C. sayi* in the Northeastern United States.

LT 16 - Soil Microbiomes of Healthy vs Blighted Cultivated Ginseng (*Panaxquinquefolius*) Roots

Isabella Maggard<sup>1</sup>, Kayla Deel<sup>1</sup>, Ying Gao<sup>2</sup>, Bruce Cahoon<sup>1</sup>

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<sup>1</sup> *University of Virginia's College at Wise, Wise, VA*, <sup>2</sup> *Middle Tennessee State University, Murfreesboro, TN*

was unidentifiable. However, a bacterium in the genus *Rheinheimera* was exclusively associated with healthy plants. In Wisconsin, soil samples were collected from two cultivation methods - field and woods. There was a statistically significant difference in their microbiomes. LT 18 - Soundscapes as key indicators for biodiversity markets

Dr. John Quinn

*Furman University, Greenville, SC*

Biodiversity markets are rapidly emerging as a tool for conservation. To support these markets, data are needed to track and report biodiversity patterns to funders, supporters, and researchers. However, there is no consensus on what the "best" indicator of biodiversity might be for these markets. In this lightning talk, I will present what soundscape ecology is and the tools used to collect and analyze the data, how it represents biodiversity at a fine resolution across space and time, and why it is an import and valuable tool to support biodiversity markets.

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American Ginseng (AG, *Panaxquinquefolius*, L.) is one of the highest unit value cash crops and one of the most well-known medicinal plants originating from North America. After 300 years of foraging, wild populations have become scarce. Nearly all ginseng is now commercially cultivated. One of the greatest challenges faced by ginseng producers is control of fungal pathogens such as blight, damping off, root rotting, and anthracnose. The goal of this project is to survey the bacterial soil microbiome of healthy versus diseased ginseng to search for differences. Our hope is to identify soil bacteria with anti-fungal properties associated with healthy plants that could be developed into an organic fungicide. Soil associated with healthy and diseased plants were collected from commercial farms in Garrett County, Maryland and Marathon County, Wisconsin. Environmental DNA metabarcoding was used to compare the microbiomes of healthy versus diseased plants from both locations. Beta diversity comparisons identified statistically significant differences between the states. Analysis of Maryland samples identified statistically significant differences between soil samples taken from healthy and diseased plants. *Mycoplasma* sp. was the predominant bacterium in soil associated with healthy plants from Maryland. In diseased plants, an unknown bacteria predominated. In samples from Wisconsin, there was no statistically significant difference between healthy and diseased samples. The most common bacteria in soils associated with both healthy and diseased plants

## ASB Posters

### P0 - ASB's Global Change Committee: Efforts to highlight members' work in global change biology

Prof. Christopher Paradise<sup>1</sup>, Nisse Goldberg<sup>2</sup>, Jennifer Hancock<sup>3</sup>

<sup>1</sup>*Davidson College, Davidson, NC*, <sup>2</sup>*Jacksonville University, Jacksonville, FL*, <sup>3</sup>*Mary Baldwin University, Staunton, VA*

ASB's Global Change Committee (formerly the Conservation Committee) is interested in learning about your research, teaching and service in global change biology which we define as the study of the effects of global climate change, land use change, invasive species, urbanization, wildfire, and pollution on biological systems. To submit your stories, please complete the survey that can be accessed using the QR code from this poster (or here: <https://forms.gle/v1JHuaqMmoHHkfDd6>). We will share the stories via ASB publications and communications, and at future annual meetings. We also plan to develop workshops, symposia, and field trips that promote and celebrate ASB efforts in global change biology, and to propose resolutions and circulate petitions to ASB members in support of reducing impact of global change, and so much more!

### SERP1 - Development and Implementation of a Bachelor of Science in Medical Biology at Lander University

Elyse Donaubauer, Melissa Hayes

*Lander University, Greenwood, SC*

More than 150,000 people in South Carolina are employed in hundreds of different Health Science occupations. According to the Bureau of Labor Statistics, healthcare jobs are projected to grow 14% between 2018 and 2028, nearly three times the average growth rate for all other occupations. In Fall 2022, the Lander University Biology Department started a Bachelor of Science in Medical Biology to address the growing demand of students entering careers in the health sciences. The Medical Biology degree program provides graduates with a robust foundation in biology and creates an opportunity to add specialized courses designed to prepare pre-healthcare students for professional programs. The curriculum is based on recent guidelines of the Association of American Medical Colleges (AAMC) and the Howard Hughes Medical Institute (HHMI). It focuses on competencies in the natural and social sciences relevant to medical education and to the practice of medicine. This program emphasizes the development of critical thinking and communication skills, as well as competency in experimental design, data analysis and methodology for research. Specialized career-focused courses, such as Emergency Medical Technician (EMT) certification, are embedded into their curriculum allowing students to gain necessary patient contact hours while staying on track to graduate with their academic class. When students graduate from the Medical Biology degree program, they will bring experiences with them that go beyond memorization and mastery of material. They will have spent time working toward goals in the program that put their skills and talents to the test, pushed them to leave their comfort zones, and shaped empathetic professionalism, setting them up for better success in matters outside the classroom. This presentation details the process of evaluating the curriculum and designing the new major, describes some of the difficulties in its implementation, and reviews outcomes from the new major to date.

### SERP5 - What determines student success in a freshman biology course: Preparation, effort, or identity?

Emily Prince, Kerry Hansknecht, Andrew Schwendemann, Lisa McDonald

*Lander University, Greenwood, SC*

Despite the critical need for scientists and engineers in the United States, fewer than half of the students enrolling in college as STEM majors graduate with a STEM degree within six years. Introductory biology courses often act as gatekeepers, limiting the number of students who advance to upper-level science courses. The biology faculty at Lander University recently redesigned the first-year biology course, biology 111, to improve student success by focusing on active learning. Although success significantly increased in the new biology 111, DFW rates remain as high as 40% in some years. We used multiple linear regression and multiple logistic regression to identify factors associated with student performance and student success, respectively, among students enrolled in biology 111. The factors we considered fell into one of three categories: factors indicating student preparedness for the course, including high school GPA and standardized test scores; factors indicating student effort in the course, including completion of pre-class reading assignments, performance on formative practice assignments, and submission of extra credit work; and a factor indicating marginalized student identity, calculated as a risk assessment (RA) score, a sum of student risk factors for failure (i.e., underrepresented minority, first-generation, low socio-economic status, and female). We found that student performance on formative practice assignments, standardized test scores, high school GPAs, and submission of extra credit assignments were all predictors of both student performance and success in biology 111. Interestingly, RA scores were excluded from both models, suggesting that a marginalized identity does not predict failure in itself. Our results suggest that student performance and success in our introductory biology class are predicted both by how prepared students are for the course as well as the amount of effort they place in the class.

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### SERP3 - PULSE: Empowering Departments Transforming Education

Judy Awong-Taylor<sup>1</sup>, Loretta Brancaccio-Taras<sup>2</sup>, Christopher Finelli<sup>3</sup>, Mark Lee<sup>4</sup>, Sara Lindsay<sup>5</sup>, Jennifer Rhode Ward<sup>6</sup>

Visit our poster to learn about the Partnership for Undergraduate Life Sciences Education (PULSE) and our key programs. PULSE is a national network of university faculty and administrators launched in 2012 as a collaboration between the National Science Foundation, the National Institutes of Health/National Institute of General Medical Sciences, and the Howard Hughes Medical Institute to stimulate department-level implementation of the changes recommended in the 2011 publication *Vision and Change in Undergraduate Biology Education: A Call to Action*. PULSE helps life sciences departments at all higher education institutions align with national education initiatives to develop inclusive, student-centered, evidence-based teaching and learning to cultivate the development of scientists who reflect the diversity of American society. PULSE focuses on the department as the unit of change because this is the level at which many decisions are made that affect curriculum as well as teaching and learning practices. Importantly, to effect change, departments need to engage in a cycle of continuous, data-driven self-assessment and improvement. As such, PULSE programs provide tools and strategies for self-assessment and teach collaborative strategies that foster cycles of improvement. We will introduce you to PULSE's key programs and showcase some of our recent efforts in our Recognition Program, Ambassadors Program, and Regional Institutes/Workshops. We encourage ASB attendees to stop by to learn how PULSE can inspire their own departments to implement the recommendations of Vision & Change.

### SERP7 - An Institutional Model for Increasing Faculty & Student Engagement through Research-Based Curricula

Clay Runck, Judy Awong-Taylor, Allison D'Costa, David Pursell, Tirza Leader, Cindy Achat-Mendes, Chantelle Anfuso *Georgia Gwinnett College, Lawrenceville, GA*

Our project addresses the challenges of reforming STEM education using a comprehensive, interdisciplinary, systems-approach model that focuses on the entire institution rather than a single course, a few courses, or a single department. Inspired by our highly diverse student population, our model was designed to meet the need to graduate *more diverse students with STEM competencies, including research and creative skills, content knowledge, and who contribute to the STEM community and workforce*. Our systems-approach model includes three interconnected levels: (1) a macro-level focusing on the **institution's** objective to increase enrollment and retention rates of under-represented, under-prepared students in STEM fields; (2) a meso-level addressing **curriculum** reform to provide rigorous research-based curricula that build STEM skills through multiple research and creative experiences for all students during their years of matriculation (via Course-embedded Undergraduate Research Experiences (CURES)); and (3) a micro-level focused on **faculty** development and **student** engagement and learning. Each level requires important processes and resources to enable institutional reform. Additionally, ongoing formative assessments between these levels inform our processes and decisions to drive institutional change. Although our project utilizes the extensive research base of 'best' practices (HIPs), our application of these practices is innovative since it promotes systems thinking among and between the different levels, thus simultaneously impacting student learning and engagement, faculty development, curricula reform, and decision making. Preliminary assessment indicates that our model may lead to improved teaching and student engagement, as evidenced by increased: (1) STEM skills in students, (2) student GPA; (3) STEM retention rates, especially for minority and under-represented groups (pre-COVID-19); and (4) student attitudes about STEM. The model also promotes collaboration and scholarship opportunities for faculty to hone scientific teaching skills; redesign curricula to include important STEM skills; collaborate with faculty outside their discipline; engage in SoTL research, and publish their scholarly work.

### SERP4 - Using variables to structure how we teach the process of science

Ann Showalter, Barbara Musolf, Michael Sitvarin  
*Clayton State University, Morrow, GA*

Teaching the process of science is ultimately about teaching students to problem-solve in a precise and methodical way. Effective instruction to develop scientific thinking and problem-solving can be quite challenging. To meet our learning goals, we have spent several years crafting curricula for our introductory biology laboratory courses using a course-embedded research experience (CURE) approach. Through numerous iterations, we have developed a simple framework centered around variables to effectively scaffold instruction on the process of science. How we define and classify the variables in a system shapes our hypotheses and predictions, our study design, the graphs we produce to visualize our results, and how we use statistics to analyze the data. Our scaffolded framework breaks down the process of science into its core components to help students build their problem solving and data analysis skills. We apply this framework to both our cellular/molecular introductory CURE lab and our organismal CURE lab, and the approach can be easily expanded and extended for more advanced courses.

### SERP2 - The effect of augmented reality on student learning and technostress in a biological sciences course at an HBCU

Elizabeth Deimeke, Rayne Thompson  
*Clark Atlanta University, Atlanta, GA*

Augmented reality (AR) applications are promising tools that engage learners with science course content (Hamilton et al., 2021). However, use of unfamiliar digital tools in a STEM course may stimulate technostress. Technostress in the workplace is associated with declines in productivity and job satisfaction (Brod, 1984; Tarafdar et al., 2007). The influence of technostress on student learning is less clearly understood. While some report

a positive association between general technology use and student performance (Dunn & Kennedy, 2019; Qi, 2019), others have shown technology induces technostress, resulting in lower levels of student productivity and quality work (Upadhyaya & Vrinda, 2021). The relationship between specific software applications and technostress as it relates to student learning in a STEM course is unknown. Set at an HBCU in southeastern U.S., this study explores the relationship between AR and technostress in a biological sciences course to better understand how this technology influences student learning. EON-XR software was used to generate AR lessons on water molecules, cell membranes, eukaryotic cells, and DNA. Student learning gains were measured using pre- and posttests, while surveys were used to capture student perceptions of learning and technostress (Kay, 2011; Tarafdar et al., 2007). Data will be gathered in February and April 2023. It is expected that students reporting high levels of technostress will demonstrate lower learning gains. However, as students gain experience with the technology, technostress levels may decrease and learning outcomes improve. This study has the potential to reveal temporal patterns in student experiences of technostress and learning outcome achievement as the result of new course technology use. Faculty awareness of student perceptions of technostress can inform implementation strategies to minimize its effect on student learning.

#### SERP8 - Implementing DNA Barcoding CURE for Community Colleges in BIO 112: General Biology II

Brandie L. Littlefield, Scott Johnson, Kimberly Fishback, Kimberly Fishback

*Wake Technical Community College, Raleigh, NC*

The standard practice of undergraduate laboratory exercises requires students to complete an ordered progression of steps to arrive at known and replicated results. While effective for developing lab skills, these exercises fail to engage students fully in the scientific process. Studies have shown that providing students with an undergraduate research experience significantly increases lifelong participation in STEM fields, particularly for historically excluded or marginalized groups. However, due to the costs and logistics of research, these experiences are often only available to students that are able to participate in out of class projects or internships at four year universities. Our goal was to develop a research project based on DNA barcoding technology for implementation across all sections of BIO 112 (second semester biology for majors) at the community college level. After several years of training, development, and laboratory experimentation, we implemented this CURE across all sections of BIO 112 in the fall of 2022. In order to establish support from administration and faculty, we ensured that this project aligns with college and department goals, as well as specifically addressing BIO 112 student learning objectives and core competencies. To facilitate the transition, we developed a comprehensive lab manual and led a training workshop for faculty and lab technicians. After one semester of implementation, we are continuing to troubleshoot the protocol and collect quantitative data on student learning and success. Preliminary data on success rates (earning a C or above) show a slight increase after the implementation of the CURE (FA2022: 72.7% (N=172) compared to FA2019: 70.2% (N=181)), but these results are impacted by the pandemic. More data are needed utilizing pre and post lab surveys that target specific SLOs. Student feedback has been positive, with particular emphasis on learning skills that prepare them for transfer to a 4-year institution.

#### SERP6 - STEMMing the TIDE: Incorporating environmental science activities into middle school Mobile County Training School science curricula.

Mark Meade<sup>1</sup>, Megan Meade<sup>2</sup>

<sup>1</sup>*Birmingham Southern College, Birmingham, AL*, <sup>2</sup>*Jacksonville State University, Jacksonville, AL*

Mobile County Training School (MCTS) is among the oldest county schools in Alabama with its beginnings dating to the 1880s. The School is in the Mobile Africatown district, a historically significant region known for environmental issues. As part of a National Academies of Science sponsored program, Birmingham Southern College (BSC), and partner institution Jacksonville State University, are working collaboratively with MCTS educators and administration to develop and implement activities to encourage and enhance middle school student participation in environmental sciences. Using improved lecture and laboratory activities and equipment, students will become agents of change as they participate in citizen science and monitor air and water quality in the local region. Further, students will propagate consumable plants and fishes, monitor water quality, and explore sustainable practices using an aquaponics system. Students will demonstrate career driven skills involved in the economics of aquaponics and will explore the use of their products in the school cafeteria and, eventually, a community garden. Modules to be implemented meet many Alabama Course of Study standards in Earth, Life, and Physical sciences and are designed to increase academic performance by engaging students in hands-on learning. The lasting goal of this project is to empower students at an early age to learn about the environment and continue to foster these practices in their homes and throughout the community.

#### P1 - The Diversity of Vitamin D3: Calcitriol and the Immune Response

Christopher Brandon, Bagie George

*Georgia Gwinnett College, Lawrenceville, GA*

Active vitamin D3 (D3, calcitriol) is best known for its role in calcium and phosphate homeostasis, however it has also been shown to exert a number of other beneficial physiological effects. For example, D3 has been shown to play an inhibitory role on the innate and adaptive immune response, and further, D3 deficiencies have also emerged as potential risk factors for the contraction of Covid-19. Specifically, it has been reported that hospitalized patients with COVID-19 had significantly low D3 levels (Brandon, B, personal communication, July, 2020). In view of the potential correlation with D3 deficiencies and COVID-19, we will investigate possible correlations between D3 and the immune response. Briefly, RAW cells will be cultured in MEM + 10% FCS, 37° C, and 5% CO<sub>2</sub> and grown to confluence. Following this growth period, RAW cells will be cultured in 24-well plates at a density of 30 x 10<sup>3</sup> cells/well and allowed to recover. Following this period, cells will then be incubated in MEM +

10% FCS alone, or MEM, 10% FCS, and increasing concentrations of D3: 25 nM (severe deficiency D3), 50 nM (deficient D3), 75 nM (insufficient D3), 125 nM (normal D3), and compared to controls (growth media alone). Following a 24-hour period, cells will then be challenged with 200 ng LPS, incubated for an additional 24 hrs., and the immune response assayed by the expression of the three inflammatory mediators, namely TNF<sub>α</sub>, IL-6, and NO. Our hypothesis is that as D3 concentrations increase from severely deficient (25 nM) through the normal D3 concentration (125 nM), there will be a concomitant decrease in the expression of the three inflammatory mediators. These results will indicate some of the antiinflammatory properties of vitamin D3, which will warrant further study into the precise mechanisms involved in this relationship.

#### P2 - A novel look into associative learning and long-term memory retention in corn snakes

Adam Green, Cliff Zeyl

*Wake Forest University, Winston Salem, NC*

Although learning and cognition have been extensively studied in mammals and birds, little is known about the cognitive abilities of non-avian reptiles. Previous studies have shown that lizards and tortoises are capable of several higher cognitive functions, however, few studies have used snakes as a model organism. Snakes offer a unique glimpse into cognitive processing because they rely heavily on chemosensory stimuli to navigate their environment as opposed to visual and tactile stimuli. Because of this, snakes have performed poorly in previous cognitive tests designed for other model organisms and have gained a reputation of being reflex machines with inferior learning capabilities. The goal of this study was to challenge this notion by looking at the cognitive abilities of 8 sub-adult corn snakes (*Pantherophis guttatus*) using operant conditioning and reversal learning. To conduct this study, the snakes were conditioned to associate small food rewards with both a scented and visual stimulus. After initial habituation, the snakes were able to reliably choose the correct stimuli during both the acquisition and reversal phases of the experiment, showing that they are capable of both associative learning and inhibition of learned responses. Upon completion of the scented trials, the snakes were subsequently tested for long-term memory capacity every 4 sessions after meeting the criteria for the last scented reversal. The data, thus far, suggests the snakes are capable of remembering a stimulus several months after exposure.

#### P3 - HSP90 and Triglyceride Levels in Partially Migratory Canada Geese (*Branta canadensis*) Populations

Audra Cote<sup>1</sup>, Andrew Dolby<sup>2</sup>, Parrish Waters<sup>1</sup>

<sup>1</sup>*University Of Mary Washington, Fredericksburg, VA*, <sup>2</sup>*University of Mary Washington, Fredericksburg, VA*

The Canada Goose (*Branta canadensis*) is a partial migrant species, which means that some individuals migrate between breeding and nonbreeding grounds, while others remain in one location throughout the year. Eastern Virginia contains both permanent residents and overwintering migrants which breed in Canada's James Bay region. The metabolic demands of migration predict physiological differences between migrants and more sedentary residents, but they have been little studied in Canada Goose. Understanding such differences may help predict the resiliency of migratory birds to rapid climate change. We compared triglyceride and HSP90 (heat shock protein) in liver tissue extracted from resident and migrant goose carcasses donated by licensed hunters. HSP90 is an oxidative stress indicator, while triglycerides indicate differences in energetic status based on differences in activity and diet. Our prediction was that HSP90, and triglyceride levels would differ between resident and migrant Canada Goose populations, with high HSP90 associated with migratory and physiological stress and high triglyceride levels associated with a more sedentary activity level. Most of the resident geese were collected in September, and migrant geese were collected in January. Body length measurements were also used to assign individuals to migrant and resident categories. Neither triglyceride nor HSP90 levels significantly differed between residents and migrant geese. They also did not differ between sexes. However, HSP90 levels marginally differed by location in Virginia, perhaps relating to differences in land use and resources available to geese. We plan to increase HSP90 sample sizes and additionally measure HSP70 and enzymes associated with triglyceride metabolism.

#### P4 - Evaluating the nonnative Joro Spider as bait for sampling fish populations

Kelly Simpson, Lisa Bachtel, Christopher Brown, Peter Sakaris

*Georgia Gwinnett College, Lawrenceville, GA*

Joro spiders, *Trichonephila clavata*, have invaded the wooded areas of Georgia. They have not been officially declared invasive yet, though several teams from institutions and agencies throughout the state are investigating their impacts on native Georgia species and the environment. While their invasive status is still being assessed, it can certainly be argued that they are a nuisance species at this time. Our team studied whether or not the arachnids make an effective fish bait. If they attract significant numbers of aquatic species, Joro spiders can potentially be harvested when they appear in large numbers and of significant size in late summer, for this purpose. In this study, Joro spiders were processed into bait balls and placed into mesh bags, and then inserted into two types of traps (minnow traps and eel traps). Joro-baited minnow traps were deployed at eight randomly selected locations in the Bromolow Creek Watershed, with minnow traps being set in both stagnant and flowing water. At each location, positive (with hot dog) and negative control (empty) minnow traps were set approximately 1-2 feet apart from the Joro-baited trap. The same method was utilized for deploying three sets of eel traps. A second trial was conducted in Tree Creek near the Georgia Gwinnett College campus. Mean CPUE was not significantly different between the three treatments; however, more fish were typically caught in Joro-baited traps. Mean total length of fish caught was not significantly different between the three treatments. Due to an unexpected weather event during trial 1 and degraded conditions during trial 2, sampling conditions were less than ideal during these trials. We plan to conduct an additional trial this spring to further evaluate the efficacy of using the Joro Spider as fish bait.

P5 - Validating point of care blood meters for assessing biomarkers of stress in sand tiger sharks, *Carcharius taurus*

Chestina Craig, Christine Bedore, Johanne Lewis

*Georgia Southern University, Statesboro, GA*

Sand tiger sharks (*C. taurus*) are large migratory sharks, with populations worldwide, including the eastern USA. They are a species of concern due to their slow reproductive output. *C. taurus* experiences regular interactions with humans as they are a popular aquarium species, targeted by recreational anglers, and are frequently studied by shark biologists. Often these human interactions involve some form of capture and handling which elicits a physiological stress response. This is typically quantified by changes in blood chemistry associated with mobilization of fuel stores (increased glucose and ketones) and changes in metabolic state (increased lactate). Stress response can be highly variable amongst species, sexes, and sizes. Researchers and aquarium professionals need to be able to analyze blood rapidly on site to ensure accurate results and inform animal care decisions. Small, battery powered, point of care (POC) blood meters make this possible. Most devices were developed for mammal physiology, are costly, and have not been thoroughly validated for use in elasmobranchs. The objectives of my study are: 1) validate use of three different POC meters (The iPet Pro glucometer, Ketosense ketone meter, and the Lactate Plus lactate meter) for use in *C. taurus* and, 2) compare glucose, ketone, and lactate levels across sex and fork length of the southeastern population of *C. taurus*. In April 2021 and 2022, a total of 31 sand tiger sharks were captured via longline within St. Helena sound, South Carolina. Sharks were sexed, measured, blood sampled, and released. Blood glucose, lactate, and ketone levels were tested on the POC meters in the field and with laboratory biochemical assays. Correlation analysis was used to assess whether POC meters produce representative data comparable to the laboratory assays. Ability to assess blood chemistry rapidly and reliably in the field will lead to greater shark stress physiology research accessibility.

P6 - Nematodes on the Move: Understanding Species Specific Nematode Extraction Efficiencies

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Entomopathogenic nematodes are typically recovered from soil media using the Baermann extraction method. This method, while effective at recovering nematodes, can have variable extraction efficiencies. These extraction efficiencies are often reported collectively for all nematode species in a sample, however. Nematodes vary in size and, in the case of entomopathogenic nematodes, display behavioral phenotypes (ambusher and cruiser) that may alter Baermann extraction efficiencies. We hypothesize that large, slower moving (ambusher) entomopathogenic nematode species will have lower Baermann extraction efficiencies than smaller, faster moving species which will more easily fit through filter holes. Knowing these efficiencies, we can better understand field sampling methods to back-calculate more accurate representations of nematode populations.

P7 - The Dietary Preferences of Five Freshwater Mussels

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Mussels play a crucial role in freshwater river environment as they are long-lived benthic ecosystem engineers. Freshwater mussels are bivalve filter feeders that remove small particles from the water, also known as suspension feeding. Studies of marine species have shown that the diet selection of an individual bivalve species depends upon a complex set of factors, including physical characteristics such as particle sizes and chemical factors such as nutrient content. Freshwater mussels tend to live in multi-species aggregates and it is unknown if they are competing for the same nutrient rich particles or if each has a preferred food source. The goal of this project was to determine if five species of mussels found in the Clinch River, Virginia would have dietary preferences or if they all feed upon the same set of microbes. Mussels grown in captivity at Virginia's Aquatic Wildlife Conservation Center were introduced into tanks of water collected from the Clinch River. After a 24hr incubation period the microbes remaining in the water were assessed using quantitative PCR and eDNA metabarcoding. We found that all five species preferentially consumed protists over bacteria. Most protist taxa, including numerous Stramenopiles, Alveolates and Ciliates, were consumed by all five mussel species but some were preferentially filtered from the water by one to four species, suggesting they may have dietary preferences. P8 - Lessons learned from an campus-based banding program

Benjamin Haywood, Dr. John Quinn

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Avian species are increasingly faced with a host of threats and pressures from natural and human forces. These forces are often unique in urban and suburban ecosystems. Bird banding can provide unique insight into these drivers and higher education campuses often supply favorable avian habitats, though such projects do come with challenges and limitations. In 2022, we tested multiple banding protocols across the Furman University campus to evaluate the impact of location, time of day, and audio lures on capture rates and success. We found that birds on campus were more difficult to capture than expected but audio lures improved capture rates of target species. Campus-based banding efforts should consider including lures to increase success in banding, though caution should be taken depending on project focus and research questions.

P9 - Oral CBD administration adversely affects mouse weight and performance in learning and memory tasks.

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*College, Macon, GA*

*Cannabis sativa* is an herbal well known for its pharmaceutical and psychotropic use, from which  $\Delta 9$ -tetrahydrocannabinol (THC) and Cannabidiol (CBD) are derived. THC is the most psychoactive chemical whereas CBD is the inactive form that has become popular for treating a variety of conditions such as epilepsy, Parkinson's disease, Alzheimer's disease, anorexia, and many others. The most common form of administration of CBD is in oil. CBD oil is a concentrated extract of cannabis flowers or leaves that are dissolved in oils such as sunflower, hemp, coconut, or olive oil. The physiological benefits of CBD have been widely studied, but less is known about the influence of CBD oil on learning and memory. To study the effects of CBD oil on learning and memory in mice, an oral form of administration was chosen. Twenty male and female C3H mice were used in the experiment. Ten animals from each sex were treated with CBD oil and the other 10 with vehicle control (Coconut oil). For each animal, 10mg/kg of CBD or Coconut oil was administrated every day for a period of six weeks, using a 0.4g vanilla wafer cookie. In the context of assessing effects of treatment on learning and memory, subjects are water-deprived for 12 hours prior to testing, a protocol that has been previously published and resulted in no adverse effects for animals. However, in the context of the CBD oil treatment regimen, requiring a period of reduction in access to both water and food, animals lost weight and failed to complete any task. The data indicate that there were slight differences in the average weight of animals that were treated with CBD oil and that under the combined stress of treatment and longer food deprivation, completion of the single alternation task was not possible without risking animal health.

P10 - Caffeine & Taurine: Potential Rescuers of Neurodegenerative Dysfunction?

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<sup>1</sup>*Philadelphia College of Osteopathic Medicine - South Georgia, Moultrie, GA*, <sup>2</sup>*Abraham Baldwin Agricultural College, Tifton, GA*

Alzheimer's Disease (AD) is characterized by an over-accumulation of  $\beta$ -amyloid plaques and neurofibrillary Tau tangles present within the brain, effectively contributing to associative neurodegeneration. Taurine is an essential amino acid found within the nervous system; however, it is one of a few amino acids not required in routine protein synthesis. Caffeine is commonly found in coffee, while taurine is found in fish, eggs, and organ meat, however, both are identified as active ingredients in energy drinks with the benefit of improving nervous system function and supporting neural recovery and synaptogenesis. In previous studies, taurine has been shown to increase memory retention in animal models, and consumption of caffeine reflects improvement in locomotion, memory, and lifespan. This project aims to determine the impact of caffeine and taurine on a mutant *C. elegans* strain, *ynls-79*, which over-expresses the amyloid precursor protein ortholog, APL-1 protein within its nervous system. We expect that supplementing the mutant *ynls-79* nematode's diet with caffeine and taurine will positively influence neurodegenerative dysfunction by reducing the number of taps required to induce habituation. We assessed anterior and posterior habituation following exposure to caffeine and taurine by tapping the worms with a sterile eyelash until the nematodes recognized the taps as a non-threatening stimulus. We hypothesize that treating the nematodes with caffeine and taurine will result in a statistically significant decrease in the number of taps required to induce a habitual response and for the mutant nematodes to associate it as a non-harmful stimulus, suggesting an improvement in memory and neurodegenerative dysfunction. Control and mutant *C. elegans* were treated with 0.250mL of caffeine and taurine with the following drug concentrations: 0mcg/mL(control), 1mcg/mL, 10mcg/mL, 250mcg/mL, and 500mcg/mL. Current preliminary results indicate a significant decrease in the number of taps required for habituation in the mutant *ynls-79* *C. elegans* strain, comparatively.

P11 - Comparing the diversity and acoustic behaviors of bats in wetlands versus forest edge in the Piedmont region of North Carolina

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Bats are one of the most economically important vertebrates. Anthropogenic disturbances such as deforestation, pesticide use, and urbanization have decimated bat populations around the world. Previous studies demonstrated that wetlands and riparian zones are heavily used habitats by bats. Many insects depend on water for part of their life cycles, making riparian and wetland habitats a potentially important foraging ground for bats. No study has specifically investigated how bats use wetlands. This study is one of the first to examine fine-scale wetland use by bats as foraging habitats. To address this research question, 10 wetlands and corresponding forest edges were acoustically monitored in the Piedmont region of NC. Using unidirectional microphones at each site, bat echolocation calls were recorded and classified into bat species and acoustic commuting or foraging calls. This project had two aims: Aim 1 is to determine the species richness of bats at the sites. The independent variable is the site location, and the dependent variable is the number of species found at each site. Aim 2 is to determine if bats use open wetlands for foraging more than forest edges. Acoustic calls will be used to monitor bat calls to quantify foraging calls versus commuting calls aimed over wetlands and along nearby forest edges. Results for Aim 1 will be presented from this summer's fieldwork. 9,451 auto-identified calls from Kaleidoscope Pro showed all seven species of bats found in the Piedmont were present at the 10 sites. The 7 bat species that are in the Piedmont of NC include: big brown bat (*Eptesicus fuscus*), eastern red bat (*Lasiurus borealis*), hoary bat (*Lasiurus cinereus*), silver haired bat (*Lasionycteris noctivagans*), evening bat (*Nycticeius humeralis*), tricolored bat (*Perimyotis subflavus*), and Mexican free-tailed bat (*Tadarida brasiliensis*).

P12 - Moth Biodiversity at the Tater Hill Plant Conservation Preserve (Watauga County, NC)

Adrianna Nelson, Matt C. Estep

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Moths (Order Lepidoptera) are a large, diverse group. Compared to butterflies, moths are far less researched and understood. Recording moth biodiversity can be a valuable step to understanding more about the health and functioning of an ecosystem. To better understand moth diversity in the mountains of western North Carolina, species richness and distribution were documented on the Tater Hill Plant Conservation Preserve in Watauga County, North Carolina. From June to August of 2022, six locations were surveyed once every other week. Bucket traps fitted with UV blacklights were deployed before sunset and moth diversity was documented via photography the following morning. Traps were placed across elevation and habitat gradients. Twenty-four sampling events resulted in 2,633 individual moth observations and preliminary identification of 198 species. In addition to common eastern moth species, northern species made a frequent occurrence, some of which were at the southernmost reaches of their ranges. Of particular note were several county and state first records, high counts, and species that had not been recorded in Watauga County in 10+ years. Documenting moth diversity contributes to our knowledge of ecosystems, which has the potential to inform future conservation management.

P13 - A ten-day snapshot of the movement ecology of Hoffmann's Two-Fingered Sloth (*Choloepus hoffmanni*) in a semi-wild environment

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Activity budgets provide a framework to understand how animals spend their time engaging in behaviors such as eating, resting, moving, and sleeping. However, in understudied or elusive taxa, such as sloths, this information is difficult to obtain in wild populations. Therefore, studies of the activity budget of elusive animals in semi-wild natural enclosures provide an opportunity to learn about their biology and behavior. Here, we measured the activity budget of two-fingered sloths (*Choloepus hoffmanni*), an animal that spends most of its time in trees where they slowly move which makes them difficult to locate and observe. Fifteen sloths were studied at Natuwa Wildlife Sanctuary, Puntarenas Costa Rica in a 65m x 65m semi-wild natural environment. We took recordings of sloth positions and behaviors across the enclosure for a continuous 10-day period during December 2022. Mapping and behavioral data were used to elucidate interesting movement patterns and behaviors that historically are difficult to record in the wild, as well as providing information that may be helpful for institutions that keep sloths.

P14 - Understanding the Effect of Controlled Burns on Soil Biodiversity

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Soil biodiversity is imperative to the overall health of an ecosystem. Many of the organisms that live there complete essential tasks and provide important structure to the soil. Moisture levels, nutrient retention, and the decomposition of organic matter rely on the species that live in the soil. The biodiversity of these soil organisms can be impacted by natural disasters that occur in their environment. In this particular case, the effect of fire is being observed. Burning forests can contribute to loss of air quality, property, crops, and animals. However, burning also contributes to forest health. Controlled burns are prescribed to supply the benefits of burning while avoiding much of the collateral damage associated with wildfires. The goal is to assess soil biodiversity in areas that have received different treatments and prescribed burns, and analyze the difference and potential effects these treatments have on species richness. To assess this, soil samples will be taken from sites at the Dupont State Forest that received 5 different treatments plus a control and placed into 18 corresponding Berlese funnels. The organisms within the soil samples will be collected in a Falcon tube, identified, and counted to quantify the species richness of each soil sample. The species richness is expected to differ between soil samples. We hypothesize that burns that occurred longer ago will be more similar to unburned soils, and closer in richness to the control samples. The more recently burned samples may have a lower species richness than the older burned samples, since they have had less time to recover. We also hypothesize that the environment type (e.g. coniferous vs deciduous), may influence the response to controlled burns. Understanding the impacts of controlled burns on soil diversity will aid conservation managers in planning for and managing the effects of burns on forested lands.

**P15 - Genetic Analysis of Distinct Chestnut Weevil Populations in the Northeastern United States**

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The American chestnut was a tree of vital importance to the forests of the eastern United States before the accidental introduction of the pathogenic chestnut blight which decimated the population. Through efforts to reestablish the historic American chestnut tree, an unwanted pest has also regained its prevalence. Curculio sayi, the lesser chestnut weevil, is a specialized seed predator that not only damages the nuts through the oviposition of eggs but also through its ability to introduce toxic fungi to the plant. As the population of its host plant has been increasing, *C. sayi* populations have re-emerged at an unnerving rate. Developing a strategy of population control of *C. sayi* stands to directly support the chestnut industry and the successful reintroduction of this historical tree. Trapping and monitoring efforts have revealed divergences in reported phenologies in Northeastern *C. sayi* populations as well as distinct morphological differences that indicate a possibility of at least two diverging species.

Currently, the origin of weevil reintroduction and human impact on dispersal are unknown. Therefore, characterization of *C. sayi* phenology and the Northeastern population structure would directly improve the efficacy of pest management efforts. The goal of this project is to examine population dynamics of *C. sayi* through DNA barcoding analysis to establish sources of origin, rates of population dispersal, and human impact on population spread. Combined with previously collected data on chestnut weevil phenology, the DNA analysis I am performing will provide insight into population variation as well as potential species divergence of *C. sayi* in the Northeastern United States.

**P16 - A SEM-based Survey of the Impact of Infestation by Parasitic Freshwater Mussel Glochidia upon the Gill Morphology of Two Species of Salmonid Fish**

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Freshwater bivalves in the order Unionida are among the most endangered taxa with over 40 species presumed to be extinct in the U.S. These mussels are unique because they possess a parasitic life history stage known as a glochidium that generally infests the gills of fish and amphibians. Upon contacting the host tissue, the glochidium ‘bites’ down causing minor surface trauma but leaving most structural tissue unharmed. The tissue of the host immediately reacts and encompasses the larval mussel in a cyst where, if able to survive, the glochidia will develop and ultimately excise as free-living mussels, living in the sediment for up to 300+ years. The infestation period can last from several days to almost a full year and is generally considered to have a minimal impact on the host. As most species live in rivers, it has been proposed that the primary purpose of this parasitic stage is dispersal, not growth. While many histological studies have detailed both the encystment process and larval development with a fair degree of detail, to our knowledge none have utilized SEM to add further prospective. Here, we investigate three stages of Margaritifera margaritifera (European Freshwater Pearl Mussel) larval development on two host salmonid species (*Salmo trutta* and *S. salar*). Our survey supports previously published results and provide an important incite as to the optimal methodology for future sample preparation. Specifically, we recommend the use of both a detergent wash and a sonication step for cleaning samples before dehydration.

**P17 - The effects of dissolved organic matter (DOM) on mosquito oviposition and developmental survivability**

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Dissolved organic matter (DOM), a mixture of organic molecules derived from the decomposition of plants and algae, is increasing in freshwater systems both regionally in the southeastern United States and globally. This upward “browning” trend raises important questions about the effects on certain species in the ecosystem, particularly those that are disease vectors like mosquitoes. DOM has been shown to protect mosquito larvae from harmful ultraviolet (UV) radiation, but little is known about the effect on oviposition. Our research explores whether female mosquitoes have a preference for DOM during oviposition. We hypothesized that because female mosquitoes use both visual and chemical stimuli during oviposition, there would be a preference to lay eggs in high DOM water versus low DOM water. In the first part of our research, 500 mL containers were filled with DOM water derived from oak leaf leachate or tap water as a control and placed in a location receiving partial sunlight on the Greensboro College campus. Oviposition within each replicate was observed over a four-week period. The experiment revealed a preference for female mosquitoes (all belonging to the species *Aedes albopictus*) to lay their eggs in DOM water. We also attempted to investigate whether the presence or absence of DOM and UV exposure affects mosquito larvae survivability. Replicates representing four distinct conditions were set up in an area with constant direct exposure to sunlight: +UV/+DOM; -UV/+DOM; +UV/-DOM; and -UV/-DOM. The experiment was inconclusive because of high larval mortality across all treatments, which was likely due to cold weather; however, it was beneficial in drawing attention to certain executional challenges and providing a direction for worthwhile future research. Overall, understanding mosquito oviposition and development is important with the combined increase in invasive mosquitos and the browning trend in the eastern United States.

**P18 - How varying nutrition levels and population density affects cannibalism rates of red flour beetles**

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Red flour beetles (*Tribolium castaneum*) are observed to be an ideal model for behavioral research due to their ability to spend the entirety of their life cycle buried in flour, often used for nutritional and microbial purposes as well as habitat. But when placed in nutritionally low flour or in

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The southern Appalachians are home to a high diversity of flora and fauna. One group, leaf litter invertebrates in the forest, can have high diversity and populations can vary based on abiotic factors (e.g., temperature, precipitation, elevation, etc.). As such, previous studies suggest that population abundances change throughout the year and by site. One interesting factor not investigated has been the impact of underlying geology. Thus, our study investigated the seasonal abundance of invertebrates in the leaf litter communities throughout North Carolina and northeastern Tennessee across a range of elevations and geologic substrates. We collected wet leaf litter samples three times from the late spring to fall from 41 plots. All samples were preserved in ethanol and enumerated by taxa and site. We detected a negative trend in invertebrate abundance from spring to fall. We

locations with high population density, known markers of stress in *Tribolium*, adult-egg cannibalism would occur. Using this information, we based 2 experiments with the intention of observing the positive correlation between stress levels and cannibalism rates. In our first experiment, the role of various diets was tested by adding 20 beetles to jars containing (1) whole wheat flour (highest nutritional value), (2) high-gluten flour, (3) teff flour, (4) oat flour (lowest nutritional value). The population density was the focus of our second experiment, accomplished by adding 20, 40, 80, and 100 beetles to individual jars containing purely whole wheat flour. The rate of cannibalism was tracked by measuring the number of eggs in each sample every 2 days. The results of the first experiment exploring the role of flour nutrition levels displayed that *Tribolium* reared in the least nutritious flour type had the highest rates of cannibalism. And in the second experiment testing population density, it was shown that as the population density grew, the number of cannibalised eggs also grew. The results of both experiments supported our hypothesis that there was indeed a significant, positive correlation between raised stress levels and rates of cannibalism. With a reliable marker of stress confirmed, further experiments are able to use the occurrence of cannibalism in red flour beetles to know when beetles are under stress.

P20 - Seasonal Fluctuations of Invertebrate Abundance in Forest Floor Communities Across the Southern Appalachians

Gwendolyn Leggett<sup>1</sup>, Alan Richardson<sup>1</sup>, Patrick Valente<sup>1</sup>, Jon Davenport<sup>2</sup>

also detected higher abundances of invertebrates from Amphibolite geological substrates compared to other geologic substrates. Collectively, our data results suggest that the seasonal abundance is greater during the warmer months and that some geological substrates may support greater numbers of invertebrates. Future research will continue this sampling to fully understand what selective forces are controlling the abundance of leaf litter invertebrates across the southern Appalachians.

P22 - Habitat Preferences of the Big Sandy Crayfish (*Cambarus callainus*) and the Role of Substrate Compositions on Available Interstitial Space

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The Big Sandy crayfish, *Cambarus callainus* is a federally threatened species endemic to the coal fields of West Virginia, Kentucky, and Virginia. *Cambarus callainus* imperilment has been directly linked to habitat degradation from extractive industries, including mountaintop-removal coal mining and logging practices that presently and historically occur across its native range. *Cambarus callainus* prefers streams with abundant slab boulders and fast flowing water. *Cambarus callainus* occupies the interstitial spaces beneath and between slab boulders and benthic substrates. To determine ideal habitat for *C. callainus* an assessment of substrate type and water velocity is required. Assessing the presence of substrate types can help determine ideal locations for *C. callainus* habitat. An acoustic active telemetry project was conducted in the Tug River in Big Sandy, WV to track movements of *C. callainus* and identify micro- and macro-habitat associations and habitat selection preferences of *C. callainus*. An assessment of substrate type in relation to interstitial space showed an increase of interstitial space with an increase in the presence of slab boulders, and a decrease with the presence of sand/silt, as well as a decrease with cobble and gravel. The combination of large slab boulders, low sedimentation, and heterogeneous substrates provides the ideal conditions for ample spaces that crayfish and other benthic species can occupy. Our data supports previous findings that *C. callainus* prefers large slab boulders with interstitial space free of excess sedimentation. Previous findings recorded that *C. callainus* had a preference of riffles over runs, but our data shows a preference for runs over all other macro-habitat types.

P23 - The Characterization of Two Novel Calcium Signaling Proteins in *Toxoplasma gondii*

Abby Delapenha

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*Toxoplasma gondii* is an opportunistic apicomplexan parasite infecting humans and livestock. Infection in immunocompromised individuals can cause neurological damage and infection during pregnancy can lead to fetal death. These parasites engage in a complex life cycle, involving repeated invasion of the host cell and egress from the host cell. Calcium signaling is an important regulatory mechanism for many essential processes in the parasite, including gliding motility (actomyosin-dependent mode of motion), invasion, and egress. Our work focused on two previously uncharacterized calcium regulator proteins (TGGT1\_253640 and TGGT1\_222060). To characterize the role of these proteins in parasite viability and calcium regulation, we fused an auxin degron system using CRISPR-cas9 modification technique. We have localized the TGGT1\_253640 to endoplasmic reticulum and TGGT1\_222060 to the nucleus of the parasite. The proteins are downregulated upon the addition of auxin as early as four hours. Future work aims to determine the role of these proteins in parasite growth and fitness using plaque assays and replication assays.

P24 - Thermal Preference of the Costa Rican Zebra Knee Tarantula (*Aphonopelma seemani*)

Zach Kaminski

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Tarantulas are ectothermic invertebrates that rely on their environmental temperatures to depict their body temperatures. Current data suggest keeping tarantulas in human care around room temperature (65-85°F). To determine if warmer or cooler temperatures are preferred by the Costa

Rican Zebra Knee Tarantula (*Aphonopelma seemani*), a choice based study is performed with 6 wild-caught subadult specimens. Each tarantula is placed in their own enclosures made up of 2 uniform tubs. Both tubs are temperature controlled for a warm and cool side. Through data collection there is clear depiction of individual preference of choice in temperature and burrowing opportunities. Tarantulas in the study prefer burrowing on the warmer side when in premolt and during ecdysis.

P25 - The Conservation Status of *Cambarus jezerinaci*

Jaden Rusnak<sup>1</sup>, Kaleb Norris<sup>2</sup>, Zachary J. Loughman<sup>1</sup>

hand excavation, dip netting, and sein hauls at each of the sites. Out of the over 40 sites that were surveyed in the summer of 2022, 12 had occurrences of *C. jezerinaci*. Our recent efforts to survey the Upper Cumberland River and Kentucky River basins have allowed us to gather genetic information from *C. jezerinaci* which will be used to assess the conservation status. Further surveys will need to be done to help solidify the distribution range of the species. Understanding the population size, distribution, and density will help determine the conservation status of *C. jezerinaci*. To date, *C. jezerinaci* has only been documented in one county in Virginia so it is important to appropriately assess the populations to find out whether they are a conservation concern in the state.

P26 - Effect of Density of Previous Host on Metacercarial Density in the Trematode *Metagonimoides oregonensis*

Cooper Kework, Jenna Ash, Carlos D. Camp

*Piedmont University, Demorest, GA*

Host-parasite relationships are important and often under-considered aspects of community ecology. Previous research has shown patterns of density of parasites with complex life cycles being related to the density of the current host but not the previous host. We investigated a system involving the trematode *Metagonimoides oregonensis* to test the hypothesis that the relative density of the previous host (snail) is not related to the relative density of parasites (metacercariae) in the current host (salamander). We chose 13 streams that are all part of the watershed of the Middle Fork in the Broad River in Habersham and Stephens counties in Georgia. In each stream, we measured the relative density of snails (*Elimia proxima*) and the relative density of larval salamanders (*Desmognathus amphileucus*, formerly *D. quadramaculatus*). We then counted the relative intensity of parasite infection in each salamander and multiplied results by the relative salamander density, generating the relative density of metacercariae. Regression showed a significant relationship between relative density of metacercariae and relative density of snails. Our results show that previous host density does influence parasite density in this system. These findings refute our tested hypothesis, indicating that relationships between parasite density and previous host density may be system specific.

P27 - Mesh exclusion bags as an effective non-insecticidal tool to manage citrus greening

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*University of Florida Citrus Research and Education Center, Lake Alfred, FL*

The citrus industry in Florida has been devastated by the advent of citrus greening, a fatal citrus disease vectored by the Asian citrus psyllid (ACP). Current management relies on protecting trees from psyllids with frequent insecticide applications, leading to keen interest in developing noninsecticidal tools to reduce pesticide reliance and costs. Citrus growers are exploring options such as mesh exclusion bags over trees, reflective mulch ground cover, and red kaolin clay particle film sprays, although the effects of these treatments have never been fully evaluated. We tested the efficacy of these three treatments in reducing ACP densities on young citrus trees against a grower standard control as well as their effects on other citrus pests and plant pathogen incidence. Among these treatments, the mesh bags successfully excluded ACP, with no trees developing citrus greening over a two-year period. Trees in this treatment also showed the lowest densities of citrus leafminer infestation and citrus canker incidence. However, trees with mesh bags were susceptible to spider mites, sooty mold, and greasy spot, suggesting that trade-offs exist with this tool. While mesh exclusion bags appear to be a promising tool for protecting young citrus trees from citrus greening, citrus growers need to monitor for other pests and pathogens that can thrive within the closed environment created by the netting.

P28 - Preliminary assessment of sodium fluoride and sodium selenate on the developmental toxicity to *Xenopus laevis* and *Ambystoma maculatum* embryos

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*Cambarus jezerinaci*, also known as the Spiny Scale crayfish, is a stream-dwelling crayfish that lives in small, rocky streams located in the Powell River and Upper Cumberland River basins in Tennessee, Kentucky, and Virginia. Little is known about their distribution, biology, and conservation status. Although there has been some disagreement with the taxonomy of the species, they were previously grouped with another similar-looking stream crayfish, *Cambarus parvoculus*. Recent genetic analysis has shown that *C. parvoculus* and *C. jezerinaci* are separate species and that there are subtle genetic differences between the Virginia and Kentucky populations of *C. jezerinaci*. The in-situ data collection methods consisted of

Shelby Wolfram<sup>1</sup>, James Rayburn<sup>2</sup>

Hannah Holbert, James A. Hartley, Zachary J. Loughman, Zackary A. Graham

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*Cambarus robustus* is a tertiary burrowing crayfish native to the northeastern United States. As *C. robustus* is a large bodied *Cambarus* species that prefers to inhabit streams dominated by slab boulders, this species may serve as a surrogate for species of conservation concern like *C. callainus* and *C. veteranus*. Currently, there is no standard for general housing and propagation of *Cambarus* species in captive care. In order to create a protocol for basic substrate requirements, we will use *C. robustus* as a surrogate species for other federally listed *Cambarus* species. *Cambarus robustus* will be monitored in three types of substrate: bare-bottom, pea gravel, and natural substrate collected in the field over a duration of two weeks. Once the first two week period is over, the *C. robustus* will be transferred to another substrate to be observed for the same duration again for the third type of substrate. Levels of activity and type of activity will be recorded based on if the subject is exhibiting natural or relaxed behaviors in comparison to stressed behavior. Ultimately, we seek to set a standard for captive care of endangered *Cambarus* crayfishes.

P30 - Establishing a before-after control-impact (BACI) framework for the assessment of Eastern Hellbender (*Cryptobranchus alleganiensis*) habitat restoration efforts in the Upper Tennessee River Valley of Southwest Virginia

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Eastern Hellbenders (*Cryptobranchus alleganiensis*) are experiencing declines in streams and rivers throughout southwest Virginia in large part due to habitat degradation. The United States Department of Agriculture (USDA) and several public and private partners are currently undertaking stream restoration efforts with private landowners across the Upper Tennessee River Valley of southwest Virginia, although few habitat data exist from portions of this watershed to guide the design of restoration efforts. To address these knowledge gaps, we examined various locations suspected of providing hellbender habitat throughout the Middle Fork Holston River, a portion of which has been targeted for restoration efforts. We took a series of samples from six locations (three degraded reaches and three reference reaches) from 2020-2022 to quantify physical characteristics of each reach that are critical components of hellbender habitat. We specifically quantified fine sediment intrusion within riverbed substrate, assessed channel morphology and estimated particle size distributions within available facets (riffle, run, pool, glide), and measured the size, orientation, and cavity dimensions of potential cover and nesting rocks at each location. Our sampling approach allowed us to establish a before-after

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Amphibians are often the main vertebrate group at risk of exposure to contaminants in ephemeral systems (Mann 2000). Sodium fluoride is an environmental pollutant source widely existing in nature. Sodium selenate can be found in nature as organic and inorganic forms and is used in fertilizer, insecticide, and fungicide. *Xenopus laevis* is a standard test model due to its transparent embryos and low maintenance. *Ambystoma maculatum* is a native species that is native to Alabama and has shown successful promise as a test model for toxicity development. *Xenopus* make an acceptable model for estimating the developmental effects of chemicals on native amphibians, specifically *Ambystoma* species. *Xenopus* and *Ambystoma* were used to determine the toxicity of sodium fluoride and sodium selenate. The assay uses multiple serial dilutions to determine how toxic the chemicals could be to the amphibians. *Xenopus* assay lasts 96 hours holding 20 embryos in small petri dishes. *Ambystoma* assay lasts 12 days holding 10 embryos in large deep petri dishes. Throughout the assay, mortalities were counted for each day. Mortalities and Malformations were counted on the last day of the assay and embryo length were measured. Means, Standard error, probit analysis (for LC50 and EC50(malformation)), ANOVA and Bonferroni's post hoc test were calculated using Systat. The teratogenic potential was calculated using the formula 96-hr LC50/96-hr EC50(malformation). Selenate was more toxic than fluoride. However, high concentrations of fluoride exposed embryos die earlier than selenate exposed embryos. Malformations such as stunted growth, edemas, and loose gut was seen in both assays. These assays will help prove the toxicity levels of sodium fluoride and sodium selenate towards amphibians.

P29 - Substrate Preference and Behavioral Analysis of *Cambarus robustus*, a Surrogate Species for the Federally Threatened *Cambarus callainus*.

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The eastern newt, *Notophthalmus viridescens*, is a semiaquatic salamander ranging throughout most of the eastern United States. Four different eastern newt subspecies are recognized: *N. v. dorsalis* (broken-striped newt), *N. v. louisianensis* (central newt), *N. v. piaropicola* (peninsula newt), and *N. v. viridescens* (red-spotted newt). From 2009 - 2019, we sampled eastern newts in eight states to collect tissue for a genetics study exploring hybridization in *N. v. dorsalis*, *N. v. louisianensis*, and *N. v. viridescens*. Sampling began in South Carolina, the state where the three focal subspecies come into contact. Over time, sampling gradually progressed west to Mississippi and north to Pennsylvania. We captured eastern newts at 102 localities from 86 counties across the eight-state study region. Seventy of the localities represent new, undocumented sites. *N. v. viridescens* composed 69% of detections, followed by *N. v. louisianensis* (21%), *N. v. dorsalis* (9%), and *N. v. viridescens* x *dorsalis* (3%). Adults (n = 675) and larvae (n = 676) composed 98% of the captures with efts (n = 28) representing the remaining portion. Knowledge of documented localities, drought severity, season, and land use history influenced capture success, particularly in the Sandhills and Coastal Plain where detection rates were low despite intensive sample effort. We captured newts in the Coastal Plain most frequently in short hydroperiod wetlands whereas Piedmont and Blue Ridge newts were captured in wetlands spanning the hydroperiod spectrum. The localities we report for *N. v. louisianensis* and *N. v. dorsalis* are significant and should further conservation attention for eastern newt populations in the Atlantic and Gulf Coastal Plain.

controlimpact (BACI) design that can be used to assess the success of planned restoration efforts in our degraded reaches and provide a model for assessing future efforts across other portions of the species' range. We will present the results of preliminary comparisons of field data across two years of pre-restoration sampling from this study and discuss how project engineers are currently using these data to implement habitat restoration efforts on a working farm along the Middle Fork Holston River.

**P31 - Weather Patterns and Weary Travelers: Uncovering Weather Preferences in Snakes and Salamanders Under Cover Boards**

Ninknee Kongmanichanh, Christopher Brown, Hannah Talbert, Ryan Davenport, Kelly Simpson, Riccardo Fiorillo, Patrick Cain *Georgia Gwinnett College, Lawrenceville, GA*

Coverboards provide a long-term, but low maintenance way to assess animal diversity in a landscape. Here, we present ongoing results from coverboards that were placed on campus in May 2019 and have been checked once a month since. Two transects of 10 coverboard arrays were placed in the wooded area north of Georgia Gwinnett College in a suburb of Atlanta, GA with each array being approximately 20m away from others. Each array consists of three plywood boards of different sizes: small (30.5cm x 30.5cm), medium (61cm x 61 cm), and large (61cm x 122cm). Each transect runs from a powerline cut downhill through a hardwood forest to the floodplain of the Yellow River. Here, we present a preliminary analysis of the relationship of generalized weather patterns and the presence of snakes and salamanders as well as other taxa.

**P32 - Eastern Newt (*Notophthalmus viridescens*) Localities and Notes From 10 Years of Sampling Across the Southeastern United States**

Eran Kilpatrick<sup>1</sup>, Gavin Lawson<sup>2</sup>

**P33 - Phylogeographic Patterns Among Eastern Newts (*Notophthalmus viridescens*) in the Southeastern United States**

Gavin Lawson<sup>1</sup>, Eran Kilpatrick<sup>2</sup>

<sup>1</sup>*Bridgewater College, Bridgewater, VA*, <sup>2</sup>*University of South Carolina Salkehatchie, Allendale, SC*

Prior molecular phylogenetic analyses of eastern newt populations in South Carolina found evidence of introgressive hybridization among the three subspecies occurring in the state (*Notophthalmus v. viridescens* – red-spotted newt, *N. v. louisianensis* – central newt, *N. v. dorsalis* – brokenstriped newt). Observed patterns of genetic variation indicate that population groups likely represent one or more refugial populations established during the last glacial maximum (Lawson and Kilpatrick 2014). To evaluate the regional extent of these groups, we sequenced 1,204 bp of the NADH dehydrogenase (ND2) and flanking tRNA genes in 610 specimens from a total of 83 populations in North and South Carolina, Virginia, Georgia, Maryland, Pennsylvania, Alabama, and Mississippi. Even with the extended sampling, haplotypes did not group by taxonomic designation in phylogenetic analyses supporting the hypothesis that introgressive hybridization has occurred. Statistical parsimony analysis recovered two population groups corresponding to those recovered in South Carolina, with one possibly the result of refugial populations isolated along exposed Atlantic Coast continental shelf during the last glacial maximum. Lastly, SAMOVA analysis recovered smaller population groups within this “refugial” group that may represent new lineages established during post-glacial range expansion and isolation by newly developed drainage patterns.

**P34 - Identifying the Role of Eukaryotic Translation Initiation Factor 3G1 in Nociception**

James Auwn, Andrew Bellemer

*Appalachian State University, Boone, NC*

The Bellemer lab seeks to investigate the cellular and molecular factors that influence nociceptor sensitivity. Nociceptors, or the neurons involved with the perception of pain, can be manipulated using tissue specific knockdown in *Drosophila melanogaster*. The *D. melanogaster* model is a powerful tool for investigating novel genetic factors involved with nociception. The roles of these novel genetic factors in nociception can be identified via RNAi knockdown; behavioral, molecular, and microscopic assays are then used to assess the effect produced. Eukaryotic translation initiation factors are an important mechanism for post-transcriptional regulation of gene expression. The knockdown of eukaryotic translation initiation factors have previously produced defects in nociceptor morphogenesis and behavioral response. Eukaryotic translation initiation factor 3 subunit g1, or eIF3g1 when knocked down results in high order branch loss and a decrease in total dendrite length, along with significant latency in larval nociceptive response time. As an ongoing project to investigate the roles of all seven eIF3 subunits in *D. melanogaster* nociception, thermal

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Chronic pain affects over 50 million people in the United States annually. Chronic pain arises from neuroplastic changes that alter the sensitivity of pain sensing neurons called nociceptors. Elucidating the molecular regulation mechanisms that lead to greater pain sensitivity will shed light on how to treat chronic pain. An important layer of regulation in neurons is the post-transcriptional regulation of mRNA. The control of which transcripts are translated into proteins, when and where they are translated, and in what quantity can all affect the sensitivity of neurons through

nociceptive assays were conducted to report the role of eIF3g1 in the behavioral facet of nociception. Based on the results, several models of eIF3g1 function in the pathway for nociceptor development are proposed, and plans for investigating the behavioral, molecular, and morphological effects of this subunit—and future subunits—on nociception are discussed.

#### P35 - Life history correlates of alternative reproductive tactics in Blue Ridge Two-lined Salamanders

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Male Blue Ridge Two-lined Salamanders (*Eurycea* cf. *wilderae*) are characterized by alternative reproductive tactics, resulting in morphological and behavioral differences in reproductive characters. One morph, “searching males,” can be identified by elongate cirri, a mental hedonic gland, and protruding premaxillary teeth. The other morph, “guarding males,” can be identified by enlarged jaw musculature and agonistic behavior. Larval *E. cf. wilderae* can exhibit either a one- or two-year larval period before metamorphosing, and this larval period is correlated with stream order. While there has been no indication that male reproductive tactics are plastic over the course of their life, it has not been formally studied. Our study had two goals: 1) evaluate whether male *E. cf. wilderae* can change reproductive tactics across breeding seasons; and 2) evaluate for relationships between larval period and reproductive tactic. We performed terrestrial and stream surveys for adult and larval salamanders. We documented adults for photographic mark-recapture using Wild ID, measured SVL of larvae, categorized microhabitat (pool, riffle, or run), and collected DNA samples from larvae for genetic assays with PCR primers. We only identified one match using the Wild ID program, which may be indicative of limitations of the program or a need for additional standardization in our photographic techniques. We found no significant difference in larval period or microhabitat preference among sexes or reproductive tactics. These results support the hypothesis that the development of alternative reproductive tactics in *Eurycea* cf. *wilderae* is not linked to environmental factors.

#### P36 - The Role of the RNA-binding protein, Pumilio, in the Regulation of Nociception

Sara Palega<sup>1</sup>, Rebeccah Stewart<sup>2</sup>, Andrew Bellemer<sup>1</sup>

affecting their signaling. RNA-binding proteins are a crucial part in this process due to their ability to bind mRNA transcripts and affect their expression. The RNA-binding protein, Pumilio, is hypothesized to be a post-transcriptional regulator of nociceptor sensitivity. *Pumilio* RNAi knockdowns in *D. melanogaster* larval nociceptors leads to mechanical and thermal hypersensitivity, suggesting *pum* could be limiting baseline nociceptor sensitivity. Pumilio has been shown to repress the translation of the voltage-gated sodium channel, Para, in motor neurons. This sodium ion channel is responsible for depolarization during action potentials, suggesting that the increased sensitivity observed in *pum* knockdown larvae could be a result of decreased repression of Para translation. This interaction will be researched further by tagging Para transcripts with Green Fluorescent Protein and observing its fluorescence in *Drosophila* nociceptors. By doing this, the effect of *pumilio* RNAi knockdown on the expression of Para can be visualized. If the knockdown of Pumilio results in hypersensitivity through the regulation of the ion-channel Para, it is expected that an increase in the expression of Para will be seen when Pumilio is knocked down.

#### P37 - Early life Corn Snake (*Pantherophis guttatus*) life history attributes under human care: determination of growth, mortality, and reproductive success for Corn Snake culture

Brianne Light, Andrew Delph, Natalie Burke, Kaleb Mill, Tyler Schwisow, Ciera Sullivan, Zachary Loughman *West Liberty University, West Liberty, WV*

Biology has historically utilized model organisms to represent a broad range of biological responses in related taxa or taxa that are not easily cultured. Until the past few decades, snakes did not have species that fulfilled this role. With the advent of conservation initiatives, understanding snake life history in captive settings was important to initiate captive breeding and head-starting programs for endangered taxa. Corn Snakes (*Pantherophis guttatus*) have long been a mainstay in herpetoculture circles, and have been bred en-mass for over five decades. Because of their ease of care as neonates, juveniles, and adults, they currently fulfill the role of model snakes for conservation-based herpetoculture. To date, a published description of feeding and growth rates, morphological response, and dedicated care needs in a conservation culture setting has not been published. To remedy this, 7 pairs of corn snakes were brumated and bred in 2022 resulting in 8 clutches and 111 eggs. One hundred and ten hatchlings were produced from these efforts. Hatchlings were measured and weighed monthly for the first 6 months of life. Growth was tracked for this time, as well as feeding rates, deaths, and sheds for each individual hatchling to produce a model of the first 6 months of life under captive conditions. Pitfalls of culture and husbandry improvements were identified. Parent's response to breeding and egg deposition was also monitored over this time. Ultimately, this work can be used by conservation managers for Corn Snake adjacent species to understand husbandry needs and expenses over time.

#### P38 - The Investigation of Endothelial Protein C Receptor Expression in Prostate Cancer

Sydney Hunter<sup>1</sup>, Jessica Bonner<sup>2</sup>

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Cancer is the second leading cause of deaths in the United States and the leading type of new case diagnoses per year (Siegel et al. 2023). In 2023, an estimated 288,300 of these new cases will involve the prostate, resulting in 34,700 deaths. Early detection and diagnosis are imperative for successfully treating these prostate cancer (CaP) patients. The most widely used diagnostic marker for prostate cancer has been prostate specific antigen (PSA). Although PSA concentration has been a valuable clinical tool for determining treatment for men with CaP, elevated serum PSA alone is not an absolute indicator of CaP since other prostate disorders can also result in elevated PSA levels. It is also important for development of treatment strategies to determine the cause of prostate cancer progression at the molecular level. Previous research has shown that an endothelial receptor, endothelial protein C receptor (EPCR) is expressed in breast cancer and ovarian cancer cells. In an effort to identify a potential new diagnostic marker for CaP, we investigated the expression of EPCR in men with CaP and in an *in vitro* in a CaP cell line. Using ELISAs, we determined that men with CaP have elevated concentrations of serum EPCR compared to healthy controls and that men with higher grade CaP (Gleason scores greater than 5) have the highest concentrations of serum EPCR. We also determined by Western blotting that EPCR is expressed *in vitro* by a CaP cell line. We conclude that EPCR is expressed by CaP cells and that patients with CaP have statistically elevated concentrations of serum EPCR. These data indicate that EPCR could be used as a serum marker for CaP. Establishing that EPCR is expressed *in vitro* by a CaP cell line lays the groundwork for future studies into the role of EPCR in CaP progression.

P39 - Rabbit Tobacco improves short-term memory in *C. elegans*

Limari Vasquez<sup>1</sup>, Courtney Alexander<sup>2</sup>

P40 - Modeling emergent patterns of synthetic Notch signal transduction *in vivo*

Abby Bryant, Amber Ajamu-Johnson, Paul Langridge

*Augusta University, Augusta, GA*

Notch is a transmembrane receptor found in all multicellular animals. The receptor has key roles in development, and mutations in the Notch system have been linked to diseases, including cancer. Notch receptors are activated by mechanical force, where ligand-receptor binding causes endocytosis of the complex by the ligand cell, resulting in conformational change of the regulatory region that allows for a series of cleavages that result in the release of an intracellular transcription factor that alters gene expression.

Recently this activation mechanism has been co-opted to produce synthetic Notch (synNotch) that can transduce a unique signal from a neighboring cell and produce a customized, and potentially therapeutic, response. SynNotch replaces ligand/receptor binding domains, regulatory regions, and transcription factors to alter receptor sensitivity and create unique signals and cellular responses.

SynNotch has the potential to be used in future therapeutics. For example, classical tissue engineering approaches aim to correct structural craniofacial defects using scaffolds or bioprinting to organize the patient's cells into desired shapes for repair. A future approach is to use cells that are capable of self-organizing into the required 3D tissue organization using developmental programs triggered by synNotch activation at precise times and locations. However, little is known about the capacity of synNotch to control cell behavior within a growing tissue. Here we aim to address this by producing a computational model of synNotch activation within a tissue.

Previous research has established synNotch ligand and receptor fly lines that are known to recapitulate Notch signal transduction. Our overall aim is to produce multiple fluorescent microscopy images of synNotch signaling interfaces within this tissue at specific developmental stages. These images will then be quantitatively assessed and used to help build a computational model of the signaling events. Overall, this research aims to further the understanding of controlling synNotch *in vivo*.

P41 - Analysis of Flt-1 sgRNA for CRISPR Cas9 mediated gene ablation in *Danio rerio*

Maraline Argueta, Holly Boettger-Tong

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As we age, most people experience memory decline, affecting one's activities of daily living. *C. elegans* are microscopic organisms used across different laboratories in many biological fields. In nematodes, chemosensation is often used to test memory. Nematodes have several neurons devoted to chemosensing and can be conditioned easily with food. I used a standard short-term memory protocol to test the effect of plant extracts on memory. For my experiment, I wanted to measure these organisms' short-term memory and chemosensation abilities. To do such, I utilized a chemotaxis assay set-up to condition my *C. elegans* to associate the smell of butanone with food. Half of my *C. elegans* would be preconditioned with the smell of butanone, the other half preconditioned with butanone and rabbit tobacco, an important medicinal plant for the Lumbee Indian Tribe of North Carolina. Over a series of trials, I found that I was able to effectively train the worms to associate butanone with food, as evidenced by the positive chemotaxis index collected for the control groups. In addition, worms that were preconditioned with rabbit tobacco performed better than those with butanone alone. Utilizing the assay helped to produce consistent results across trials. For my next experiment, I will be testing the *C. elegans* spatial and short-term memory in a T-maze.

Wesleyan College, Macon, GA

In 2012, Jennifer Doudna and Emmanuelle Charpentier revolutionized gene editing when they discovered that the CRISPR-Cas9 system could be used to ablate specific DNA sequences. Clustered regularly interspaced short palindromic repeats (CRISPR) were initially found in *Escherichia coli* in 1987; it was later discovered that other bacteria and archaea used these repeats to fight off infections from viruses by incorporating a portion of viral DNA in their own genetic material, through which the bacteria could recognize foreigners and immunize itself. Doudna and Charpentier won the Nobel Prize in 2020 for developing the ability to manipulate CRISPR-Cas9 to alter specific DNA sequences. This technique can selectively target genes for disruption or repair and is an incredibly powerful tool that can be used to rewrite the genome. Our lab is developing this technique to ablate specific genes expressed during development in zebrafish, *Danio rerio*. Previous DNA microarray studies in our lab have shown that treatment of teleost fish embryos with all-trans retinoic acid, a potent teratogen, resulted in altered expression of both VEGF and Flt-1 mRNA. Vascular endothelial growth factor (VEGF), a highly conserved protein, plays an important role during embryonic development regulating angiogenesis and vasculogenesis via two receptor tyrosine kinases, Flt-1 and Flk-1. To better understand the direct relationship between Flt-1 mediated VEGF signaling and regulation of early development, CRISPR-Cas9 mediated ablation of Flt-1 will be attempted. Candidate sgRNAs were identified using CRISPRscan and selected sgRNA DNA templates synthesized via fill-in PCR. Successful sgRNA DNA template generation was verified through gel electrophoresis and PCR products were purified using three different methods to compare yields for sequencing and to obtain clean products for subsequent in vitro transcription reactions. Co-injection of these in vitro transcription reaction products with Cas-9 will allow us to examine the role Flt-1 plays in early *Danio rerio* development.

#### P42 - Effect of Domain structure on BURP-domain protein Expression in Arabidopsis

Sara Traver, Breanna Wimbush, Eric Johnson

*Virginia Wesleyan University, Virginia Beach, VA*

BURP domain proteins (BDPs) are prevalent in the cell wall of higher plants, but their molecular activity has been poorly characterized. These proteins are known to play key roles in stress responses as well as in facilitating certain developmental pathways. The BDP lineage underwent extensive diversification as seed plants emerged and recent studies have begun to define BDPs as peptide cyclases, modifying their own repeat domain as an early step in the biosynthesis of peptide-based specialized metabolites. Though this activity was shown among diverse lineages of vascular plants, including a lycophyte, it is unclear if it is common among all BDPs. We recently identified the elevated expression of PpBURP4 and PpBURP5 during spermatogenesis in the model moss, *Physcomitrium patens*. These are members of a unique BDP lineage only found in seedless plants which lack the repeat domain shown to be acted on by the BURP domains of other BDPs. We ectopically expressed and fluorescently tagged these genes under a constitutive *ACTIN* promoter in *Arabidopsis* to characterize their subcellular localization. Surprisingly, expression was isolated to the apoplast of mature trichomes suggesting that they are subject to post-transcriptional regulation, preventing expression in non-trichome tissue. We hypothesize that this results from the activity or interactions of the BURP domain itself. To test this, two domain-deletion lines for each BDP are being produced, removing either the BURP domain or the variable N-terminal domain, and changes in expression or localization will be analyzed by confocal microscopy. Furthermore, the proteins' size & expression are being further assessed through western blots. Results will be discussed in an effort to better understand the role BURP domains play in plants.

#### P43 - The Role of RNA Binding Proteins and CRISPR/Cas9 as a Gene Editing Tool in *Drosophila* Nociception

Erik Rangel Silva, Andrew Bellemer

*Appalachian State University, Boone, NC*

Chronic pain is a major issue in the United States and across the globe: current opioid treatments are inadequate due to its addictiveness and inconsistent symptom management. To elucidate the underlying mechanisms of the pain response, we study the role that RNA-binding proteins (RBPs) play in regulating the function of pain-sensing neurons. *Drosophila* is a powerful model to study this due to its genetic tractability, simple maintenance needs, and high proportion of genes that have human homologs. In addition, larval and adult stages can be used to study how changes in neuronal plasticity affect nociception. This provides a wealth of experimental paradigms that can be used. The neurons of interest in this study are nociceptors which detect harmful stimuli. To determine the role of candidate genes in nociception, genetic manipulations are restricted to nociceptors with the GAL4/UAS system. RNAi then targets and degrades the mRNA of these genes, silencing them at the translational level. Although this is a powerful method, there are drawbacks, including inconsistent phenotypes and off-target effects. A novel method uses CRISPR/Cas9 to silence expression and provides a new way to determine the function of candidate genes in nociception. My research produced a transgenic fly line incorporating both Cas9 and GAL4/UAS technologies to silence nociception-related genes exclusively in nociceptors. This was confirmed with Cas9-mediated knockout of a fluorescent reporter gene. My results show that knocking out expression of the mechanosensitive ion channel subunit Pickpocket significantly reduces nociceptive function, which is consistent with previous findings. Additionally, Cas9-mediated knockout of a developmental kinase Lk6 resulted in no change in nociceptive function, as expected. This suggests Cas9-associated cytotoxicity is minimal. These findings support the utility of Cas9 gene editing as a supplemental method to gain a deeper understanding of normal nociceptor function that may be used in larval or adult models.

#### P44 - The antimalarial Tafenoquine alters the expression of genes that protect against oxidative stress

Amanda Long, Chris Barton

*Beloit College, Beloit, WI*

Cancer is a major health concern and there is a huge need for future research into new disease therapeutics. One feasible option is drug repurposing, or the use of currently FDA-approved drugs for additional diseases. Recently, a number of reports have surfaced showing that antimalarial compounds are viable options for cancer therapy. Our group has previously shown that one antimalarial compound, Tafenoquine, is extremely useful in decreasing the viability of multiple cancer types grown in culture, though the mechanism of how this drug is affecting cellular viability is currently unknown. Here, we show that Tafenoquine alters the expression of multiple genes that provide protection from oxidative stress. These findings may suggest that Tafenoquine is negatively affecting cellular growth through the induction of oxidative stress.

**P45 - Phenotypic Effects of RING1- and YY1-Binding Protein in Glioblastoma**

Ronald Bucher, Lauren Patterson, Brayden Fults, Daniel Stovall *Winthrop*

*University, Rock Hill, SC*

Glioblastoma multiforme (GBM) is an aggressive form of brain cancer that has horrendous survival outcomes with the use of current therapies. Further study into its molecular mechanisms will inform development of new, more effective treatments. The Polycomb protein RING1- and YY1-Binding Protein (RYBP) has emerged as an important gene in multiple cancers. In complex with other Polycomb proteins, RYBP acts to repress regions of chromatin, though it also performs other functions independent of these complexes. RYBP has a tumor suppressive role in various cancers, but may act as an oncogene in others, demonstrating its context-specific effects. The role of RYBP in GBM has not yet been elucidated. In GBM, RYBP expression is frequently downregulated compared to normal brain tissue, suggesting it may act as a tumor suppressor in GBM. Thus, we hypothesized that forced expression of RYBP in GBM cell lines would activate apoptosis while decreasing cell invasion, migration, and proliferation. We transduced U-118 or T98 GBM cell lines with lentivirus expressing RYBP or a GFP control and established stable cell lines. RYBP-expressing U-118 and T98 cells showed decreased migration in wound-healing assays and invasion in Matrigel-coated Boyden chamber assays when compared to control cells. SDS-PAGE and Western blots were performed to measure changes in epithelial-to-mesenchymal (EMT) and apoptotic protein markers among transduced cells, and WST-1 assays were conducted to study the changes in proliferation. Overall, our findings suggest RYBP exerts anti-tumor effects in GBM and acts as a tumor suppressor gene. Future work should investigate the mechanism of RYBP's phenotypic effects in GBM.

**P46 - Biofilm Inhibition in *Staphylococcus aureus* using Small Molecules as Competitive Inhibitors**

Shelby Blanchard<sup>1</sup>, Tori Glazier<sup>1</sup>, Evan Floyd<sup>1</sup>, Anjali Patel<sup>2</sup>, Matthew Simmerman<sup>1</sup>

to clinical settings to help stave off the worsening issue of antibiotic resistance by making bacteria more susceptible to host immune responses and to existing antibiotic treatment.

**P47 - Anti-Proliferative Effects of Mefloquine on Lung Cancer Grown in Culture**

Diego Melendez, Chris Barton

*Belmont University, Nashville, TN*

Cancer is a major health concern and there is a huge need for future research into new disease therapeutics. One feasible option is drug repurposing, or the use of currently FDA-approved drugs for additional diseases. Recently, a number of reports have surfaced showing that antimalarial compounds are viable options for cancer therapy. Here, we show that one antimalarial drug, Mefloquine, is effective at decreasing the viability of lung cancer cells grown in culture. Furthermore, we show that treatment with mefloquine results in activated caspases, suggesting that cells are dying due to apoptotic signaling pathways.

**P48 - Treatment with benzimidazole derivatives suppresses migration of cervical and breast cancer cells on ibidi plates**

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In the world of modern medicine, the issue of antibiotic resistance has become more of a problem than ever before. Because of biofilm's role in assisting the incidence of antibiotic resistance, and its association with various health conditions, there arises a need for studying the inhibition of biofilm formation as an alternative to traditional antibiotic treatment. Recent studies have found that bacteria utilize small auto-inducers to communicate between planktonic cells in a process known as quorum sensing. These signals trigger metabolic cascades that culminate in the production of biofilm. By mimicking the structure of these auto-inducers, our team was able to synthesize and develop a library of 220+ compounds that were used to screen for biofilm inhibiting activity in *Staphylococcus aureus*. Through triplicate confirmation in Crystal Violet testing and various secondary assays, multiple drugs were found to successfully inhibit biofilm formation in these species without affecting the growth rates of the bacteria. Most of these drugs include a functional group of a long carbon chain with a neutral amino acid. These compounds could be applied

Micah Lamour, Daniel Sapozhnikov, Kevin Suh

*High Point University, High Point, NC*

In all multicellular organisms, cell migration is essential in multiple processes such as tissue development, wound repair, homeostasis, and immune response. Cell migration also plays a pivotal role in cancer as it is closely associated with tumor development, invasion, epithelial to mesenchymal transition, and metastasis. Drugs such as fenbendazole and albendazole are benzimidazole derivatives and are used as anthelmintics to treat a variety of parasitic worm infestations by targeting helminth tubulin proteins. However, these drugs can also target mammalian tubulins and therefore act as moderate microtubule destabilizing agents leading to cancer cell death. Previously we have shown that these drugs can cause apoptotic cell death in prostate and cervical cancer cells. In this study, we tested whether these drugs could inhibit migration of cancer cells at sublethal dose. A typical migration assay involves manual scraping of confluent cell layers which generates inconsistent gap width. To avoid this, we used ibidi plates which feature a removable silicone insert that separates wells where cells are grown. The removal of the insert reveals four 500-micrometer-wide cell-free gaps and one 1,000-micrometer center gap for cells to migrate. When treated with anthelmintic drugs, migration of cervical and breast cancer cells was significantly inhibited compared to untreated control cells. Finally, we used ImageJ software to calculate percent gap closure at different time points. These results suggest that such drugs can be used to combat invasive and metastatic behavior in cancer cells.

**P49 - A methodical quantitation of Nicotinamide Adenine Dinucleotide (NAD), nicotinamide riboside (NR) and nicotinamide mononucleotide (NMN) in extracts from dietary sources.**

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Nicotinamide adenine dinucleotide ( $\text{NAD}^+$ ) is an essential small molecule found ubiquitously in living cells.  $\text{NAD}^+$  is the primary energy carrier to the electron transport chain in the form of  $\text{NADH}$  (oxidized  $\text{NAD}^+$ ), as well as a cofactor for many enzymes including sirtuins, a family of protein deacetylases important for proper remodeling of chromatin and mitochondrial function that have been linked to biological aging.  $\text{NAD}^+$  levels decline steadily with age, indicating it as a central player in biological aging. Restoring  $\text{NAD}^+$  content to young levels, through increased consumption of dietary  $\text{NAD}^+$  precursors, may lead to improved aspects of health and aging such as body weight, eye function, and insulin sensitivity. While the concentrations of nicotinamide and nicotinic acid, referred to as vitamin B3 complex, are well established, existing literature on the concentration of the more recently discovered precursors nicotinamide riboside (NR) and nicotinamide mononucleotide (NMN) in various foods is extremely limited. In this study we selected an array of fruit and vegetables of different genus for initial evaluation. We have simultaneously quantified total  $\text{NAD}^+/\text{NADH}$ , NR and NMN in grocery produce using an enzyme-coupled fluorometric cycling assay. We also successfully replicated the results seen previously in milk, which are the only published measurements for the simultaneous quantitation of NAD, NR and NMN without using liquid chromatography. This demonstrates a rapid and accessible method for assessing the levels of these compounds in foods and provides important information regarding potential health benefits of different diets.

**P50 - Metamorphosis of *Chrysaora chesapeakei*: determining the induction source leading to strobilation**

Kristen Boddy<sup>1</sup>, Dr. Rebecca Helm<sup>2</sup>

**P52 - Assessment Precision of Diatom Communities for Water Quality Analysis**

Anna Agi<sup>1</sup>, Sydney Brown<sup>1</sup>, Kalina Manoylov<sup>2</sup>

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Diatoms, a common group of algae in lotic environments, have cell walls made of hydrated silica, or glass, that survive and function as robust biological indicators for variable aquatic habitats. Accurate identification of diatom species is essential, but also the effort of meaningful number enumerated needs more research. Assessing improvement of aquatic habitats with diatoms can be complementary to other tools like macroinvertebrates and physiochemical data. Fourteen streams with known exposure to nutrients due to agriculture were sampled in 3 different locations within the stream. Following current protocols equal parts of each replica were subsampled and combined in a composite sample. Diatoms were enumerated as 14 composite samples rather than 42 samples. At random, replicas from 3 streams were enumerated and those additional counts were compared to the composite analyses. Within-stream replicas had high similarity, but lower species richness. Within replica counts, individual species abundances were consistent, no taxon had higher than 34% relative abundance. Relative abundance was sample and species specific.

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Jellyfish, members of the class Scyphozoa (phylum Cnidaria) have some of the most complex life cycles of any living creature, capable of undergoing metamorphosis twice. The juvenile polyp metamorphoses through a process called strobilation, a form of asexual reproduction. The polyp elongates and cleaves into many free-swimming juvenile jellyfish, termed ephyra. The mechanisms that drive this remarkable metamorphosis are not well understood. Through transplantation and microdissection, this research aims to reveal the source of the induction signal for the metamorphosis of *Chrysaora chesapeakei*, commonly known as the bay nettle, through strobilation. Future research will be able to build on this to determine the molecular pathways for metamorphosis, leading to a greater understanding of the evolutionary development of Cnidarians.

*Achnanthidium deflexum* (Reimer) Kingston, a common low nutrient indicator stayed at 12-14% relative abundance regardless of valves enumerated. Species richness and Shannon diversity were significantly higher in the composite samples ( $p<0.001$ ). Evenness also stayed relatively high (ranging between 69-90%). Percent similarity based on species composition was high also, 51-59%, which would indicate that diatom analyses for BMP's assessment would be accurate. Results of this research suggest that both: shorter counts from field replicas or longer counts of a single composite sample can be used, dependent on the research objective. From the composite samples, diatom community indices and low abundance of nutrient tolerant diatoms suggested measurable improvement of water quality and reduced risk of harmful algal blooms or lack of ecosystem services. Within stream replicas are a valuable tool to study diatom population ecology.

**P53 - Are there increases in CRISPR/Cas off-target effects in homologous recombination repair deficient *Saccharomyces cerevisiae* cells?**

Salvador Castillo, Irma Santoro

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CRISPR/Cas technology is increasingly being used as a common methodology in many cancer biology studies due to the ease and convenience of the technique. Precise editing of genomic DNA has been achieved upon repair of CRISPR-induced DNA double-strand breaks (DSBs) by homologous recombination (HR). HR repairs DNA DSBs with high fidelity and therefore, deficiencies in HR result in genome instability. These deficiencies have been demonstrated in many cancers. RAD51-dependent HR is a very important pathway for repairing DSBs. Previous studies have shown that genome editing using CRISPR technology relies on the repair of site-specific DNA DSBs induced by the RNA-guided Cas9 endonuclease. Furthermore, previous studies have shown that the efficiency of CRISPR-mediated HR can be improved by the stimulation of HR-promoting factors, such as the RAD51 recombinase. Despite the ease and efficient use of the CRISPR/Cas technology for genome editing, one limitation is the potential occurrence of associated off-target effects. If CRISPR technology is planned to be used to target cancer cells with defective HR capabilities, will off-target mutations be likely to occur? In order to answer this question, a system was developed in *Saccharomyces cerevisiae* using green fluorescent protein (GFP) as a reporter to identify off-target CRISPR-induced DSBs. This study set out to test the number of off-target DSBs that could be introduced by CRISPR-induced genome editing in a RAD51-deficient HR model. We were curious whether loss of RAD51-dependent HR would increase the abundance of off-target CRISPR-induced DSBs in mutant yeast strains as compared to those with a functioning HR-dependent DNA repair pathway. Preliminary findings using this system will be presented.

**P54 - Effects of site preparation method on early plant establishment and soil characteristics in a small prairie meadow**

Nicole Soper Gorden, Shaelyn Roberts, Robert Zinna, Hailey Harwick

*Mars Hill University, Mars Hill, NC*

Prairies are one of the most endangered biomes in North America, with an estimated 90% lost to agriculture, development, invasive species, and other disturbances. Long-standing efforts focus on maintaining and restoring remnant prairies. There is less research on establishing new prairie sites, and most suggested best practices involve expensive site preparation methods and large equipment more suitable for large sites. At Mars Hill University, we are working to establish a small (~1 acre) prairie site for teaching, research, and recreation purposes. Since funding and resources are limited, we began by establishing test plots to determine which site preparation method is best to remove pre-existing vegetation, including several invasive species: smothering with black plastic, smothering with clear plastic, or burning. Plots were prepared in spring of 2021 and planted in summer of 2021 with a seed mix of 80 native prairie species and seedlings of native prairie grasses. Plant species were surveyed in each plot once a month and soil characteristics were monitored. We found that the burned plots had a significantly higher soil pH, lower soil moisture, and lower plant species richness. Plots smothered by clear plastic had significantly higher species richness than any other treatment. Re-invasions have so far been too infrequent to analyze but will continue to be monitored. Additionally, early data from a parallel project found a higher diversity of invertebrates present in clear plastic plots. Our results suggest that smothering sites with clear plastic may be the best site preparation method for establishing small prairies, despite the literature's clear bias towards using fire instead. We hope our final results will provide clear guidance on the differences that may exist between establishing large prairie sites and establishing small prairie meadow sites.

**P55 - Pharmacological and Genetic Manipulation of Longevity Pathways in the Model Organism *Caenorhabditis elegans***

Kaley Nofziger, Michele Malotky

*Guilford College, Greensboro, NC*

Animal aging, a complex and multifactorial phenomenon, is defined by the decline of functional processes that take place in an organism over a period of time. It is characterized by the reduction of essential physiological functions required for survival and fertility, ultimately leading to an increased risk of mortality. There are a great number of predisposing factors that have been associated with poorer healthspan and decreased lifespan in individuals. These factors include genetic deterioration, biological gender, and shortening telomeres. Various lifestyle changes such as eating healthier, increased physical activity, and improved social well-being are linked to increased healthspan and lifespan. In animal models, pharmacological interventions such as rapamycin, a member of the nicotinamide adenine dinucleotides (NAD) family, have been shown to increase longevity in model organisms. Research on *Caenorhabditis elegans*, a nematode with homology to humans, has yielded a number of genes and signaling pathways correlated with increased lifespan. This study looks at two signaling pathways. The first involves a mutant strain, age-1, which interferes with the insulin-like growth factor (IGF) signaling pathway. The second, involves the pharmacological inhibition of the mTOR pathway

induced by rapamycin. The hypothesis being tested is that the combination of both genetic and pharmacological intervention will lead to longer lifespan than either intervention independently.

**P56 - Microbial ecology of sand fly breeding sites: aging and larval conditioning alter the bacterial community composition of rearing substrates**

Nayma Romo Bechara, Kasie Raymann, Gideon Wasserberg

*University of North Carolina at Greensboro, Greensboro, NC*

Phlebotomine sand flies are insect vectors that transmit microorganisms such as *Leishmania* protozoa—responsible for leishmaniasis diseases. With no vaccine available, the use of personal protection and residual spraying with insecticides are used to reduce exposure to sand fly bites. Hence, a more targeted and efficient control method is needed. One promising control strategy is “attract-and-kill”, where the vector is lured to an insecticide using attractants. Sand flies have a terrestrial life cycle. Eggs are laid in organically-rich soil on which coprophagous larvae feed and develop through four instars before pupation and adult emergence. Given that attraction to decomposing organic matter is often mediated by bacterially produced semiochemicals, and that previous literature has demonstrated that gravid sand flies are attracted to their early life stages, we wanted to **(1) test the effect of larval substrate conditioning (feeding and defecating) on bacterial community structure and (2) characterize and compare the bacterial community composition of conditioned and unconditioned substrates across the life-cycle stages of developing sand flies**. Using 16S rRNA amplicon sequencing, we compared the diversity, presence, and abundance of taxa across substrate types at four time points (sand fly larvae life stages).

We hypothesized that, following larval introduction, the bacterial community structure of conditioned and aged substrates would diverge (i.e., become less similar). However, after all pupae had eclosed, we expected the bacterial communities of both substrate types to decrease in diversity and become more similar. Consistent with our hypotheses, we found significant change in bacterial communities within and between substrates over time. So, by temporally characterizing the bacterial composition of fresh, aged, and larval-conditioned substrates, we've provided evidence that substrate decomposition and larval conditioning alters the bacterial community of sand fly rearing substrates, which subsequently may affect the oviposition site selection behavior of gravid sand flies.

**P57 - Riparian Buffer Restoration Within a Pastoral Matrix: Impacts on Soil Microbial Communities and Soil Carbon Sequestration**

Seth McCullough

*James Madison University, Harrisonburg, VA*

Riparian forests have been well documented for their ecosystem functions and services including improving water quality by reducing soil erosion and nutrient runoff, creating biodiversity hotspots, and serving as a natural solution to climate change through carbon sequestration. Riparian forests tend to be more productive compared to their upland counterparts and models have predicted that riparian zones can potentially sequester significant amounts of carbon within their soils. Soil microbes play an important role in the soil carbon cycle but are rarely investigated in studies on carbon sequestration. Additionally, land use change can have profound impacts on soil physicochemical properties but the associated changes in soil microbial communities are poorly understood. In this study, I will investigate the efficacy of riparian buffer restoration for improving soil microbial community composition and function, while also determining how these changes impact long-term soil carbon sequestration. I will determine the belowground response to restoration efforts by surveying a chronosequence (0-30 years) of restored riparian buffers. Each restored riparian buffer will be paired with an upstream, unrestored pasture to account for differences in soil type and land-use history among restoration sites. Soil microbial community composition will be assessed using phospholipid fatty acid analysis (PLFA) while their function will be assessed through soil respiration rates. Additionally, soil physical and chemical properties (moisture, pH, aggregate stability, bulk density, total nitrogen, and total carbon) will be assessed. I predict that restored riparian buffers will have a greater diversity of microbial composition and function and higher soil carbon concentrations than riparian areas where cattle are present. By looking at relationships between age of restoration and soil quality indices I hope to identify belowground impacts of riparian buffer restoration and determine the optimal restoration conditions for the improvement of soil microbial communities and long-term carbon sequestration.

**P58 - Stable Nitrogen Isotopes Document Carnivory in Harvestmen**

Maynard Schaus, Monika Metro, Victor Townsend,Jr.

*Virginia Wesleyan University, Virginia Beach, VA*

Harvestman species are thought to consume a wide variety of available living and dead animal and plant materials, with varying scholarly opinions on the degree to which they rely on carnivory vs. omnivory in their diet. Studies often rely on sporadic field observations or laboratory measures that may not approximate actual field conditions, and are less quantitative measures of the contribution of animal, plant, and fungal material to the diet over time. Stable nitrogen isotope analysis provides an ideal means to quantify the contribution of these materials to the diet over the time scale of tissue turnover. Organisms are typically enriched 2-3‰  $\delta^{15}\text{N}$  over that of their food items, due to preferential excretion of  $^{14}\text{N}$ . Thus, carnivores are relatively enriched in  $\delta^{15}\text{N}$  compared to herbivores, with omnivores having intermediate  $\delta^{15}\text{N}$  signatures that reflect the proportion to which plant and animal materials comprise the diet. We measured  $\delta^{15}\text{N}$  values in the tissues of 3 harvestman taxa from western Belize, and compared them to values of carnivores (scorpions and ctenid spiders), herbivores (grasshoppers and katydids), ants, and termites. Scorpions and spiders typically had mean  $\delta^{15}\text{N}$  values of around 7, whereas katydids and grasshoppers had mean  $\delta^{15}\text{N}$  values of around 5, reflecting a plant-based diet.

In contrast, ants and termites had mean  $\delta^{15}\text{N}$  values of 3.24 and 3.73, respectively. Two of the harvestman taxa (*Erginulus clavotibialis* and *Vonones sp.*) had mean  $\delta^{15}\text{N}$  signatures above 7, indicating that the vast majority of their nitrogen was derived from animal sources. The third harvestman taxon (*Prionostemma sp.*) had  $\delta^{15}\text{N}$  signatures ranging from 6.68-6.83, indicating they were primarily carnivorous, with a maximum of 14% plant material in the diet. Our study provides an initial quantification of harvestman diet and documents that these taxa are primarily carnivorous.

**P59 - The effects of *Alternanthera philoxeroides* (Alligator Weed) on algal communities in Lake Baldwin (Tifton, Georgia)**

Barbara Mcfadden

*LSAMP, Tifton, GA*

*Alternanthera philoxeroides* is an invasive species that originated in South America. It is considered an ecological threat because it can form thick mats along lakes inhibiting the growth of native plants and preventing wildlife's access to the water. We examined the effects of *A. philoxeroides* on algae in Lake Baldwin using a field fluorometer. This device was used to calculate the amount of chlorophyll in water samples at six different sites around the lake in which three contained *A. philoxeroides* and the other three did not. In the sites with *A. philoxeroides*, the chlorophyll levels were lower meaning fewer algae grew in these areas. Future laboratory experiments will include observing how algae respond to *A. philoxeroides* exudates.

**P60 - Do predators use different mechanisms to avoid human presence and human infrastructure?**

Georgia Spann, Emily Prince, Lisa McDonald

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Human disturbance can cause predators to change behavior, such as by altering hunting times, diel activity, or habitat use. These disturbances can be temporary, as with the presence of vehicles or recreational activities (e.g., hiking or dog walking), or they can be long-lasting, as with habitat destruction and fragmentation due primarily to the construction of roads and buildings. In this study, we examined how large predators (i.e., black bears, bobcats, coyotes, and wild boar) respond to temporary human disturbance (THD) and lasting human disturbance (LHD). We predicted that predators would temporally avoid THD by being active at night rather than by leaving the area. We also predicted that in areas of higher LHD predators would be less common but would not alter their diel activity because lasting disturbance can only be avoided spatially. To test our hypotheses, we installed cameras at diverse locations within six sites throughout Upstate South Carolina and analyzed each image by recording the species present and the time of day. We determined THD based on the frequency of images containing human activity at each site. Using GIS, we calculated metrics of LHD using total road length and building area within buffers centered on each camera station. We found that predators were significantly more nocturnal in areas with high levels of THD, but there were no differences in the diel activity patterns in areas with different intensities of LHD. In contrast, we found no correlation between THD and the number of predators sighted per day, but the frequency of predator sightings was negatively correlated with LHD. Overall, predators avoid temporary human presence temporally and lasting human infrastructure spatially.

**P61 - Effects of species identity and time-since-fire on the presence of microbes on leaf surfaces of shrubs in longleaf pine sandhills**

Dallas Nivens, Jennifer Schafer

*Winthrop University, Rock Hill, SC*

Leaf surfaces are often colonized by nonpathogenic microorganisms, which may have beneficial immune and metabolic relationships with plants. Factors such as proximity to the ground and leaf surface moisture and waxiness can influence the types of microbes able to colonize plant leaves. To investigate factors that influence the presence of cyanobacteria and nitrogen-fixing bacteria on leaf surfaces, we studied shrub species with varying growth habits in longleaf pine sandhills in South Carolina. To assess effects of species identity, we measured stem height, number of stems, leaf area, and specific leaf area (SLA) of eight shrub species in burned sites (4 to 6 months post-fire). To assess the effects of time-since-fire, we also measured stem and leaf characteristics of *Gaylussacia dumosa* and *Sassafras albidum* in unburned sites (2 years post-fire). We collected bacteria samples from leaf surfaces, which we will analyze for the presence of cyanobacteria and nitrogen-fixing bacteria using PCR. In burned sites, stem height and number of stems varied among species; *Clethra alnifolia* and *Gaylussacia* had the shortest stems, and *Oxydendrum arboreum* had the most stems. Leaf area and SLA also varied among species in burned sites; *Oxydendrum* and *Sassafras* leaves were larger than other species, and *Clethra* had the highest SLA. While *Gaylussacia* stem characteristics did not differ with time-since-fire, leaves were larger in unburned sites. *Sassafras* had more and taller stems and a lower SLA in unburned sites. Differences in stem and leaf characteristics may impact which microbes can colonize leaf surfaces by influencing habitat accessibility and microclimate. Time-since-fire may influence the colonization and presence of certain microbes on leaf surfaces as the environment changes over time after fire. Knowing how the presence of microbes differs among species and in relation to environmental variation is an important step in investigating beneficial plant-microbe relationships.

**P62 - Plant Pollinator Networks of Greenwood**

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Athens, GA

This research effort aims to explore the pollinator network in the longleaf pine savanna habitat of Greenwood. Greenwood is a historic plantation located in Thomasville, Georgia in south Georgia and represents some of the most pristine remnant longleaf pine savannah habitat in existence. While this habitat is recognized for its remarkable and unique flora, little research has been done in southeastern pine savannas on associate native bee pollinators, even though these systems are known for a relatively high proportion of endemic and rare flora. In this study, we are researching the pollinator network of Greenwood to assess pollinator diversity and ecological services provided by arthropod vectors to plant partners. Pollinating insects are recognized as a critical link in reproductive success and native bees account for the bulk of these pollination activities, both in natural and cultivated areas throughout the world. However, data concerning regional make-ups of these pollinator guilds is severely lacking. Our investigation revealed a correlation between soil moisture and bee population diversity at Greenwood, however additional survey work needs to be done to assess the strength of this relationship and provide land managers with complete picture of critical pollinator services being provided to the plant community of this remnant habitat.

#### P63 - Long-term, Post-Restoration Survivorship and Species Invasion at Rocky Branch

Lauren Willhite

*NC State University, Apex, NC*

With urban stream restoration becoming more common, post-restoration monitoring and research should assess plant communities as a metric of long-term success. Rocky Branch, a mile-long first-order stream in Raleigh, NC, presents a unique opportunity to examine plant communities 13, 17, and 21 years post-restoration. At Rocky Branch, we will assess survivorship and identify some of the best-performing native plants, both planted and volunteer. We will also identify prominent invasives and possible mechanisms for invasion. Other restoration studies suggest that American sycamore, yellow poplar, southern red oak, and green ash perform well in restored areas (Bradburn et al. 2010, Drayer et al. 2017). Furthermore, they note that species that promote canopy closure reduce invasive presence (Paredes and Jones 2001). To evaluate this, we will collect data with transects that include stem count, DBH of native plants, and cover class. Based on the planting lists and restoration literature, we hypothesize that river birch, mockernut hickory, green ash, tulip-poplar, black gum, and various oaks have performed well. Our preliminary survey for invasives indicates that two major species of concern are English ivy and glossy privet. Overall, this research will help inform future stream restoration projects in terms of well-performing native species for planting and strategies for reducing invasion.

#### P64 - Genetic Diversity in Asian Bittersweet (*Celastrus orbiculatus* Thunb.) Near a Site of Initial Introduction Shows No Evidence of a Bottleneck

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Asian Bittersweet (*Celastrus orbiculatus*) is a woody non-native vine first introduced to North America in the 1860s through the Arnold Arboretum (Massachusetts), New York Botanical Garden, and Biltmore Estate (North Carolina). On the landscape it hybridizes easily with its native congener, American Bittersweet (*Celastrus scandens*), and it can also outcompete the native plant to cause extirpations. Little is known about the species' genetic diversity in its introduced range, and whether evidence of a post-introduction genetic bottleneck can be discerned. This study tested the efficacy of newly developed microsatellite markers for Asian Bittersweet, then used those markers to determine genetic patterns in seedlings and adults near one site of introduction. Adult and seedling Asian Bittersweet leaves were collected from geotagged locations within a 150 km of the Biltmore Estate, then DNA was extracted using a modified CTAB method. Next, extracts were PCR amplified at seven microsatellite loci. All loci were reliable, variable, and informative, with only 4-5 needed to distinguish among individuals. Results showed broad genetic similarities between adults and seedlings across sites, and although allelic diversity was higher in adults, the difference was not statistically significant. Levels of genetic diversity were comparable to those found in other vines, and no evidence of drift was seen. This study is one of the first to compare diversity across generations of a non-native species, and results could have implications for management of this and other species. Next steps will include spatial analyses of genetic diversity patterns and testing for cross-amplification with other *Celastrus* species.

#### P65 - Islands in the sky: Evaluating a Constraint-based model of Dynamic Island Biogeography in southern Appalachia

Trinity Johnson, Kimberly Cook, Joseph Burger

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Since its first treatment in 1967, island biogeography has been a crucial theoretical tool for researching the roles that ecology, evolution, and immigration play in impacting biogeography. Despite this being an increasingly popular field, there is still much to be discovered about the historical and dynamic contribution of species' traits toward biogeographic distributions among islands. The Constraint-based Dynamic Model of Island Biogeography (C-DIB) is a systematic attempt to discern the drivers underlying such complexity. Central to the C-DIB model is how an island system's historical and contemporary habitat suitability, island connectivity, and island size affect present species distributions on islands. By evaluating this model in a natural system, we can quantify how ecologically relevant traits interact with environmental constraints over history to produce island biodiversity. We curated 2,367 occurrence records for 12 small mammal species and 12,786 occurrence records for 7 salamander species from the Global Biodiversity Information Facility (GBIF) to identify sampling gaps for 12 "sky islands" in the southern Appalachians. Our

study revealed significant data deficiencies in high elevation sites along the archipelago based on a quantitative summary of geographic and taxonomic biases. The salamander species were relatively well-sampled and there were no community composition discrepancies between the community and museum data, despite the island and species sizes. The small mammal results did show community composition discrepancies on the smaller islands. The small mammal data inconsistencies indicate the need for more data collection along the spruce-fir archipelago to test the C-DIB model, however current salamander species occupancy data can be considered complete. We conclude with future directions to fill data gaps with field survey methods and museum data.

#### P66 - Adaptations to Sun & Shade Exposure in *Helianthus schweinitzii*

Zachary Taylor, Jessica Taylor

*Winthrop University, Rock Hill, SC*

*Helianthus schweinitzii* commonly known as Schweinitz's Sunflower is a federally endangered species of the Asteraceae. The Schweinitz's Sunflower is native to the North and South Carolina Piedmont regions and is regularly seen in disturbed areas, prairies, and under power lines where there is nutrient-poor clay soil. A 10 x 10m prairie plot with approximately 2000 plants was surveyed and split into two sides based on plants exposure to the sun and to the shade. Data collected from the plot indicated a few to no differences in soil pH, temperature, or moisture between the two locations. Morphological differences noted from plants in the sun and in the shade indicate optimal growing conditions based on observed differences in leaf length and width, color, and number of inflorescences and seeds. Determining better growing conditions of the Schweinitz's Sunflower is vital to improving rehabilitation efforts in the Piedmont region.

#### P67 - Comparing Foliar Fungal Endophyte Frequency to Elevation and Host Leaf Functional Traits in the Southern Appalachians

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Foliar endophytic fungi (FEFs) are fungi that live within leaves, between plant cells, without causing visible signs of disease in their host. FEFs display high taxonomic richness, and the majority of undescribed fungal taxa on earth are thought to be endophytic. FEFs are ubiquitous in all temperate tree species, yet the exact nature of the relationships between specific FEF taxa and their hosts is poorly understood. The southern Appalachians are a highly biodiverse region, both in terms of tree diversity and endemic fungal partnerships (e.g., lichens, ectomycorrhizae). As such, they offer rich potential for studying FEF/host tree relationships. This study examines FEFs in the southern Appalachians. We compared leaf functional traits to FEF diversity in sweet birch (*Betula lenta*, Betulaceae) leaves along an elevation gradient, across three forested mountain slopes, near Asheville, NC, in late summer 2022. A LiCor LI-6400 Portable Photosynthesis System was used to measure photosynthetic and transpiration rates, as well as instantaneous water use efficiency. Leaf samples were cultured on agar media to observe FEF frequency and diversity. Cultures were identified by unique morphotype, as a proxy for taxonomic richness. Leaf functional traits were associated with elevation at the site level. FEF frequency was 100% across all sites, and all sites displayed high morphotypic richness. FEF diversity did not differ among sites, but there was some evidence that sites harbored distinct FEF FEF community assemblages. These findings, especially considering the limited sample size and small spatial scale of the study, suggest that the southern Appalachians are a promising region for further FEF research. Ongoing work includes characterizing FEF taxonomic richness using ITS1/ITS2 DNA barcodes and will be presented.

#### P68 - Effects of forest reclamation and landscape features on avian community structure and species diversity in Appalachia

Rebecca Davenport<sup>1</sup>, Christopher Barton<sup>1</sup>, Victoria Burgess<sup>1</sup>, John Cox<sup>1</sup>, Todd Fearer<sup>2</sup>, Jeffrey Larkin<sup>3</sup>, Lauren Sherman<sup>1</sup>, Madoline Varias<sup>1</sup>, Steven

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Surface coal mining and subsequent reclamation efforts transform the ecological characteristics of natural landscapes, often altering the habitat and distribution of native wildlife. The Forestry Reclamation Approach (FRA) is a recent reclamation method that emphasizes best management practices in forestry, such as soil decompaction and the planting of native trees and shrubs. Although the FRA is expected to benefit wildlife communities, no studies have empirically examined the effects of the FRA on avian species. Our study aimed to identify which reclamation approaches and/or landscape features within coal surface mines promote breeding songbirds, particularly forest interior avian guilds and bird species of concern. We conducted traditional point count surveys in high-elevation, red spruce-northern hardwood forests in the Appalachian Mountains of eastern West Virginia. We assessed differences in avian occupancy, species richness, species abundance, and community composition between four site types: 1) young FRA sites, 2) maturing FRA sites, 3) non-FRA reclaimed mine sites, and 4) unmined, mature forest. We found that both FRA sites and reference sites support numerous priority landbirds and species of greatest conservation need in this region. Furthermore, we found that site type had a strong effect on the occupancy and abundance of some species, as well as overall species richness. As the FRA becomes a more widespread and popular reclamation approach in Appalachia, this study will inform future management and reclamation efforts based on considerations for native birds.

#### P69 - Social interactions, temporal clustering, and dominance rank of birds at a water source.

Price<sup>1</sup>

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Intraspecific and interspecific interactions of birds at an artificial water source were studied using a game camera programmed to take a photo every minute from sunrise to sunset from September 2021 through April 2022. We addressed the following questions: 1) which species are the most and the least social?, 2) is the degree of sociality at water linked with body size or dominance rank?, and 3) are bird visits clustered temporally? In the resulting 165,744 photos, bird visits were categorized as either 1) solo, 2) with conspecific, 3) with conspecific and heterospecific, or 4) with heterospecific. The Phi coefficient, a measure of association, was calculated for each of the 15 most frequent bird species. Dominance rank was derived from the literature. To test for temporal aggregation, the period 1-5 PM (EST) was divided into 15-minute increments. A conservative count of unique bird visits per increment was then compared to the expected (Poisson) distribution if bird visits occurred randomly in time. The Phi coefficient ranged from -0.008 to 0.062, with the most interspecifically social species being Eastern Bluebird, House Finch and Chipping Sparrow, while the least interspecifically social species were the Northern Mockingbird, Eastern Phoebe, and Mourning Dove. The Phi coefficient was not correlated with dominance rank (regression,  $p = 0.17$ ); however, less dominant species had a significantly higher percentage of visits to the bath with heterospecifics (regression,  $p < 0.01$ ). Bird visits to the water source were clustered in time, with a significant departure from randomness as predicted from a Poisson distribution (Chi-squared test,  $p < 0.001$ ). Temporal aggregation of birds at water sources could be driven by exogenous factors (sun, temperature, predators) but we propose that the primary driver is social factors, e.g., enhanced vigilance achieved by associating with other birds when bathing and drinking, especially for less socially dominant species.

P70 - Preliminary biodiversity assessment of degraded stream in Giles County, TN

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The creeks in Giles County, Tennessee are significantly degraded from their original ecological function, largely due to the urbanization of the area and non-point pollution sources. A large majority of the county resides in the Richland Creek-Petty Branch watershed, which, as a whole, has been altered significantly by pollution and land-use. The watershed was once home to a large variety of aquatic invertebrates. However, no current data exists on the extent to which these species have been affected by ecological decline of the area. This project's aim is to establish a collaborative network of scientists, state agencies, and community members to i) survey macroinvertebrate biodiversity, ii) improve water quality, and iii) restore the riparian habitat. The project's starting point will be one of the most significantly degraded areas in Pulaski, Pleasant Run Creek. It is located near the city center and is a direct tributary of Richland Creek, where the intake of the city's water is located. The proposed project offers the opportunity for an integrated science, education, and community project that will positively impact a large part of the watershed in the county and the region's economic resiliency.

P72 - Germination of *Eragrostis elliottii* and *Aristida purpurascens* seeds after repeated exposure to smoke-infused water.

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The Pine Rocklands are an endangered ecosystem in southern Florida. Very little Pine Rockland habitat remains, much having been lost to development that inhibits regular fires that are necessary to maintain this ecosystem. Treatment of seeds or soil using a smoke and water solution may supplement the need for fire without endangering nearby developed or inhabited areas. Smoke-water solutions contain chemicals such as karrakinolide, a compound known to increase germination success, and trimethylbutenolide, a known germination inhibitor. Because different plants demonstrate different sensitivities, they likely respond differently to varying smoke concentrations. We examined the effects of smoke-water concentration and application frequency on the seed germination of two species native to Pine Rockland understories, *Eragrostis elliottii* and *Aristida purpurascens*. Neither species demonstrated a significant difference in germination when exposed to repeated applications compared to a single application of the same solution. There was a significant difference in germination success between different concentrations of solution. Smoke-water demonstrated increasingly inhibitory effects on *A. purpurascens* germination as concentration increased. *E. elliottii* seeds demonstrated greatest germination success when treated with 40% smoke solution, and reduced success at other concentrations. Our data suggests that a specific concentration of smoke-water between 20% and 40% would most effectively increase germination success, without overly inhibiting the germination of other species. A solution of this concentration could be effective for large scale application to Pine Rocklands.

P73 - Variable Pollen Production and Hybridization Potential in Imperiled North Carolina Pitcher Plants

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*Sarracenia jonesii* (Mountain Sweet Pitcher Plant; endangered) and *Sarracenia purpurea* var. *montana* (Mountain Purple Pitcher Plant; federal species of concern) are both imperiled carnivorous plants native to North and South Carolina bog environments. These species are sometimes sympatric, and phenotypic hybrids are found in all sites where the parental types co-occur. We compared the timing and quantity of pollen

production of the two species and their hybrids from three sites to determine differences in reproductive effort that could affect future population trajectories. Anthers were collected over three flowering seasons, macerated with Alexander's stain, and homogenized, then viable pollen grains were counted using a hemocytometer. Pollen production and viability varied by week and plant type, but there was broad overlap between species, creating opportunities for interbreeding. Across all sites, *S. jonesii* produced less total pollen than *S. purpurea* var. *montana*, and hybrids made intermediate amounts of pollen. While there was no effect of year or site on total pollen production, viable pollen quantities varied across time and space. Future work should correlate this measure of reproductive effort with seed production and test seed germination across sites and species to better estimate threats posed by hybridization. Knowing reproductive effort, reproductive output, and fitness of parental species and their hybrids can help conservationists understand the long term impact of hybridization on these populations.

P74 - Spatial dynamics of invasive (*Amynthas* sp.) and naturalized earthworms in West Liberty University's campus woods community.

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Pheretimoid earthworms (genus *Amynthas*) from east Asia have been reported in the hardwood forests of the eastern United States with increasing frequency and of greater range. These invasions can have highly negative ecosystem-wide impacts. *Amynthas* sp. are prolific breeders capable of reaching extreme densities in forest soils where they are not native. Invasive *Amynthas* consume large quantities of leaf litter, drain the available nutrients from the forest floor, alter soil conditions, and allow damage to occur in plant communities. As there are no native earthworms in the northeastern United States, the community is composed of naturalized species such as the European *Dendrodrilus* and *Dendrobaena* along with the more recent invasive Asian pheretimoids. Understanding the differences in population dynamics between these earthworms could be able to aid the development of management strategies to prevent or limit the spread of invasive pheretimoid earthworms and reduce the disturbance and destruction of native forest ecosystems. Four lumbricid taxa and one megascolicid (*Amynthas*) have been identified in the study area with *Amynthas* worms being the most abundant. In the summer and fall of 2021 and 2022, earthworm population surveys were conducted in an area of West Liberty University's campus woods community known to be infested with pheretimoid worms. Spatial dynamics were examined using SADIE analysis. It was found that both *Amynthas* sp. and the selected naturalized species showed trends towards patchy aggregations. The temporal patterns of aggregation between the two groups (*Amynthas* and lumbricid) differed, with the patterns of the *Amynthas* showing more irregularity. Better understanding of the spatial dynamics of these worms, particularly the invasive *Amynthas*, could have implications for conservation purposes and forest management.

P75 - What are the impacts of deer browsing and Japanese stiltgrass (*Microstegium vimineum*) on native sapling growth in a residential wetland in southeastern Georgia?

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Wetlands provide a variety of ecosystem services, including stabilizing climate, supporting biodiversity, and regulating water flow, yet they remain vulnerable to the threats of invasive species and overabundant deer. In particular, invasive Japanese stiltgrass (*Microstegium vimineum*) can alter ecosystem conditions by competing for natural resources, while white-tailed deer (*Odocoileus virginianus*) browsing can limit growth of woody species, and hence, recruitment of young trees. In a forested wetland in Georgia, USA, we investigated whether invasive stiltgrass, deer browsing, or both limited the growth of two species of native saplings (*Liriodendron tulipifera* and *Taxodium distichum*). To this end, we removed stiltgrass in 1x1 m plots around 10 individual 2-year old saplings of each species (removal) and left stiltgrass in 10 additional plots (control). Then we established cages around the saplings in half the removal and half the control plots and left the rest uncaged. We measured sapling growth, vegetation cover and stiltgrass cover biweekly from May to November. Overall, 100% of *T. distichum* survived regardless of treatment, compared to only 43% of *L. tulipifera*. *Taxodium distichum* grew 10 cm on average whereas the surviving *L. tulipifera* did not increase in height regardless of treatment. For *T. distichum*, change in height was negatively correlated with stiltgrass cover at its peak in October. Preliminary analysis showed that cages had no effect on any measure of growth for either species. Our study suggests that deer herbivory had no measurable effect on either species during the growing season, while invasive stiltgrass, when present in high abundance, limited growth of *T. distichum*. The environmental conditions of this wetland did not support high survivorship of *L. tulipifera* saplings. Further studies should be conducted to evaluate the effects of invaders and deer browsing on tree growth at other life stages.

P76 - Using bioacoustics to evaluate biodiversity of traditional and sustainably managed cattle farms in temperate and dry tropical forests

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may be a more important factor than the number of trees scattered across a farm. Time since implementing tree conservation practices on experimental farms may also be relevant. Panama farms had only been enrolled in conservation for 11 years while Virginia farms ranged from one to 20 years. We suggest that although tree conservation programs may be effective in increasing carbon sequestration, cattle pastures may not provide sufficient habitat to support a diverse wildlife community, emphasizing the need for nearby forest fragments in highly deforested regions.

**P77 - Co-Occurrence Analysis and Species Distribution Modeling in the Blue Ridge Mountains for the Management of Invasive Plant Species**

Daisy Ryan, Mike Madritch

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The Blue Ridge Mountains support several endemic communities, such as the well-known spruce-fir forest. However, the encroachment of invasive plants into its higher elevations may threaten biodiversity and increase susceptibility to disease. Modern management plans commonly target individual invasive species, however, a single-species approach is ineffective for the Blue Ridge Mountains, which hosts more than 23 invasive plant species. Our research conducts an invasive species co-occurrence analysis on the Blue Ridge Parkway (BRP; NC and VA), to track the occupancy of multiple species and use interspecies presence relationships as a proxy for identifying invasive species of concern for mitigation planning. Surveys were conducted along an elevation gradient from 650 to 6,000 feet and paired with long-term precipitation and temperature data to predict the future directions and rates of spread for all observed invasive species. The co-occurrence analysis indicates that several species along the Blue Ridge Parkway are positively correlated with one another providing them with an establishment advantage when another correlated species is present. Ecological niche models were conducted to identify areas of favorable habitat that currently host or have the potential to host these invasives with changes in climate over time. This study will provide land managers with new, updated information on invasive plant habitation and provide actionable information on which invasive species have positive correlations or high invasion risk. This will improve prevention practices and accuracy on occurrence reporting for the BRP.

**P78 - Environmentally Friendly Pest Control: Queensland Fruit Fly Chemical Ecology**

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Pest insects cause serious threats to horticultural industries by damaging produce and contaminating exports, thereby threatening access to international and domestic markets. In recent years, the invasive Queensland fruit fly (Qfly) has become incredibly destructive. The current national annual cost of Qfly damage is estimated to be ~300 million AUD (Yazdani, 2022). This equates to ~213 million USD per year. Current methods of pest control include harmful pesticides which poison the environment and the crops where the Qfly lays its eggs. The Applied Biosciences Research Laboratory at Macquarie University has undertaken a project to create environmentally sustainable insect pest management strategies. To do this, components of kairomones were extracted from a known Qfly predator, *Oecophylla smaragdina*, the green ant. Of the green ant kairomone components, the Qfly reacted most adversely to the alcohols octanol and nonanol. These alcohols were then used to create natural insecticides and were microencapsulated to prevent rapid evaporation. To test the efficacy of these products, 50 mature male flies and 50 mature female flies were placed in a mosquito net with four bell peppers. Each bell pepper was coated with either a treatment substance, octanol or nonanol, or with a control substance, water+T20 or water alone. After 24 hours, the bell peppers were removed. The puncture sites were counted and the eggs were extracted and counted. In the treatment groups there were significantly lower amounts of puncture sites and eggs than in the control groups. Lower puncture site counts and lower egg counts suggest repellent success. This indicates that treatments of microencapsulated octanol and nonanol act as repellents for the Qfly.

**P79 - Microplastics in Freshwater Food Webs and Their Potential as Substrates for Algal Growth**

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Plastic products have exploded in popularity and use since the mid-1900s, resulting in plastic pollution. A common form of plastic pollution is microplastics (25µm-5mm). The productivity of marine communities can be linked to resources needed for the proliferation of primary producers

The beef cattle industry occupies two-thirds of land and is a major cause for deforestation worldwide, resulting in habitat and biodiversity loss. Cattle pastures are increasingly recognized for their potential to combat climate change through reforestation. However, the effect of incorporating more trees into cattle pastures on wildlife diversity is largely unknown. In this study, we quantified acoustic diversity within 19 active cattle pastures in Virginia and 15 in Panama. In the summer of 2022, we set up automated recording units (ARUs) on each of the surveyed farms representing one of two management types: traditionally managed (control) or enrolled in a tree conservation program (experimental). One ARU was set up at each farm where tree density was greatest record sound at dawn and dusk for a period of three days. Two control farms and two experimental farms were simultaneously recorded during each three-day period. We predicted farms enrolled in conservation programs would have a higher tree density which would then correlate to higher acoustic diversity.

Our preliminary results suggest that farm management has no significant effect on acoustic diversity indices in either Panama or Virginia. Other factors may be more important to increasing wildlife diversity in regions dominated by cattle pastures. The presence of a nearby forest fragment

like algae. While we often recognize chemical (nutrient) resources as limiting, spatial constraints for algal growth can also impact productivity. Biofilms can be important food sources for primary consumers and microplastic pollution could be a substrate for algal biofilm growth. Yet plastics can also release chemicals that limit algal productivity. Production can be measured using chlorophyll *a* concentration ([Chla]) and/or the number and form of algal cells (live, dead, single, clumped). If microplastics function as a spatial resource for algal biofilms, then [Chla] and live clumped cell counts should reflect that enhanced growth. However, microplastic pollution is a concern because they can be mistaken for food by foraging animals, and this is more likely to occur when plastics are camouflaged by biofilm. Here, we grew algal cultures in the presence of high-density polyethylene (HDPE) microspheres and microfibers. We used these as food sources for laboratory-reared copepod populations. In preliminary studies using smaller microspheres (45um), we found algal growth did not differ as measured by [Chla] concentration ( $p=0.100$ ), and the frequencies of different algal cells (single, clumped, live, dead) ( $p=0.713$ ) did not change. Copepods did not consume a larger proportion of live/dead cells in the presences of microplastics ( $p=0.946$ ), nor did the number of algal clumps change in the presence of microplastics ( $p=0.699$ ). We repeated these trials with larger microspheres (77um) and microfibers (2-5mm) to determine if larger surface areas of microplastics have similar results.

#### P80 - Microbial Bioindicators of Nonpoint Source Sewage Contamination in Karst Springs

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Karst springs are biodiverse hydrologic features that are both under-studied and under-sampled. Due to the direct connections between sinkholes, sinking streams, and underlying cave systems, these springs are often impacted by human land use activities on the surface. A preliminary biodiversity survey of the microbiome inhabiting Mill Creek Springs (Southwestern Virginia) was conducted in 2021 using eDNA metabarcoding, and bacteria associated with sewage were the predominant microbes detected. The goal of this 2022 project was to determine if any specific sources of fecal contamination could be detected in groundwater upstream of Mill Creek Springs. We hypothesized that the source of contamination may be septic systems serving residences and schools and/or the infiltration of agricultural runoff. Five sites upstream from Mill Creek Springs were sampled using two techniques – water filtration and glass beads suspended in the water to collect periphyton. The highest concentration of *Escherichia coli* was detected from Mill Creek Springs using IDEXX Colilert tests, however, each of the upstream samples were elevated above levels deemed safe for recreational contact. Environmental DNA metabarcoding indicated the microbiomes of all the sites were similar, with no significant differences. While these results did not allow us to determine the source(s) of the fecal contamination, we cannot rule out the additive contributions of multiple sources of surface water and groundwater that enter the cave system already contaminated with feces, most likely due to shallow soils and poorly-maintained septic systems.

#### P81 - Seed Viability and Germination Rates of Two western North Carolina Pitcher Plants and Their Hybrids

Alyssa Lynch, Dr. Jennifer Rhode Ward, Dr. Rebecca Hale, PhD, Caroline Kennedy

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Two western North Carolina native species of pitcher plants, *Sarracenia jonesii* and *Sarracenia purpurea* var. *montana*, co-occur in mountain bogs and seeps. Both are currently of conservation concern; with *S. jonesii* declared as federally endangered and *S. purpurea* var. *montana* as a state endangered species. These species are experiencing population declines due to habitat destruction and they also readily hybridize when in sympatry. Hybridization poses a problem for the conservation of these two species because of the possibility of hybrids outcompeting their parental species and because gene flow among these populations could result in the loss of genetically distinct gene pools for the two parental species. In late summer 2022, seeds from all three plant types were collected from a western North Carolina site. A subset of seeds were weighed and seed viability was determined using tetrazolium assay. Seeds were germinated in growth chambers with a 14 h light:10 h dark light cycle at 20°C and 70% humidity, and checked for radicle emergence every 2 days. Seeds were also germinated at the field site in both peat moss and seed bags. Analysis of variance was used to determine if there were significant differences in seed size, seed viability, or germination rates among the three plant types. This study complements previous research on pollen production among these plants to obtain a more complete understanding of the current reproductive status of *S. jonesii*, *S. purpurea* var. *montana* and their hybrids at this site.

#### P82 - Effects of temperature exposure on endurance swimming capacity of juvenile white sturgeon

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Despite a decline in global sturgeon populations, little is known about how well sturgeon navigate water diversion sites within the highly altered waterways they inhabit. Assessing sturgeon swimming abilities may help us to understand how water diversion sites might impact these populations. Increases in water temperature in these rivers due to the impacts of climate change could influence the swimming capacity of sturgeon. Therefore, we tested the effects of temperature on the swimming performance of juvenile white sturgeon (*Acipenser transmontanus*) using endurance, or fixed velocity, tests. Juvenile white sturgeon ( $9.5 \pm 0.5$ cm total length  $\pm$  SD;  $n=64$ ) were exposed to two temperatures, 18 C and 23 C for two weeks and then tested for endurance swimming capacity at their respective temperatures. Fish were tested at speeds of 35, 42, 49, and 56 cm/s. Our results show that temperature has an effect on endurance swimming. Surprisingly, fish exposed to warmer temperatures were able to maintain swimming performance longer during endurance swimming tests. These results suggest that the threshold temperature for juvenile white sturgeon endurance swimming capacity is above 23 C. These results will help to inform managers about the swimming capacity of juvenile white sturgeon at different

temperatures so they can make more informed decisions when designing diversion sites and fish passageways. Additionally, this information adds to our knowledge about how climate change will affect white sturgeon.

**P83 - Population Genetic Structure of Tricolored Bats(*Perimyotis subflavus*) in the Coastal Plains of the Southeastern U.S.**

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The tri-colored bat (*Perimyotis subflavus*) is a species heavily affected by white-nose syndrome. Significant population declines have occurred in the Appalachian Mountain and Cumberland Plateau regions. Little is known about tri-colored bat populations in the Coastal Plain region of the southeastern US where the species has been documented to inhabit transportation structures in addition to tree roosts, caves, and mines. Summer and winter habitat preferences by the tri-colored bat are not well understood, particularly regarding the use of manmade roosting structures. The size and distribution of these culvert-dwelling populations are currently unknown but could represent a significant remnant population. The objective of this study was to determine genetic connectivity of tri-colored bat populations roosting and/or hibernating in transportation structures and relatedness of Georgia Coastal Plain bats with the heavily impacted tri-colored bat populations in the Appalachian Mountains. Bats were collected by hand. Standard morphological measurements were recorded. Oral swabs and hair samples were collected. Samples were stored in silica gel desiccant at 0°C in the field; then transported to Kennesaw State University and stored at -80°C. DNA was isolated with a DNeasy kit (Qiagen). We surveyed 6 microsatellite markers and will sequence from the non-coding HV1 region of the mitochondrial genome. This genetic assessment of *P. subflavus* population structure will reveal dispersal patterns among sampled sites, identify isolated populations or connectivity among populations. These data will be combined with geospatial distribution data and *P. destructans* presence/absence data to understand factors associated with disease spread, and susceptibility to white-nose syndrome.

**P84 - Long-term effect of riparian restoration using macroinvertebrates as indicators**

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The Shenandoah Valley encompasses some of the highest agricultural producing regions in Virginia, many of which are large contributors of nutrients and sediment. The Conservation Reserve Enhancement Program (CREP) assists landowners in the installation of riparian restoration projects in which cattle are fenced out and a riparian buffer is planted. We examined the long-term temporal effects of riparian restoration and the impact of upstream landuse on quality from one farm for 12 years. We hypothesized that water quality would continue to improve as time passed after restoration. Water quality was quantified by measuring the benthic macroinvertebrate assemblages using the Hilsenhoff Biotic Index (HBI), Virginia Stream Condition Index (VA-SCI), and Shannon Diversity Index. Positive relationships between VA-SCI or Shannon Diversity and time since restoration signifies better stream health while a negative relationship between HBI and time since restoration signifies better stream health. Linear regressions showed a negative relationship between HBI and time since restoration ( $p<0.001$ ,  $R^2=0.215$ ) and a positive relationship between VA-SCI and time since restoration ( $p=0.037$ ,  $R^2=0.078$ ). There was no significant relationship between Shannon Diversity and time since restoration. An unexplained drop in water quality occurred in 2013, so data was reanalyzed to look at improvement after the drop. The linear regression showed a significant positive relationship between VA-SCI and time since restoration with a higher correlation ( $p=0.004$ ,  $R^2=0.194$ ) while not much changed for HBI and Shannon Diversity ( $p=0.003$ ,  $R^2=0.202$  and  $p=0.09$ ,  $R^2=0.072$ ). The restored stream continued to improve after the initial restoration.

**P86 - Assessment of microplastic contaminants in *Faxonius cristavarius* across various levels of habitat degradation**

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Microplastics, commonly categorized as particles <5 mm in diameter, are becoming an increasingly researched topic in ecotoxicology. Most of what is understood about environmental microplastics stems from coastal and oceanic ecosystems. Freshwater systems are currently understudied even though their close relation to urbanization can lead to a considerable amount of microplastic introduction from various sources (runoff, wastewater, atmospheric deposition, and fragmentation of plastic debris). This lack of research in freshwater habitats presents a knowledge gap involving microplastic retention within freshwater organisms. To accurately understand their occurrence in freshwater habitats and bioaccumulation in freshwater organisms, it is important to examine aquatic invertebrates due to their placement in the sediment and water column, where microplastics have been recovered from streams. Crayfish make an ideal study taxon due to their prevalence in regional streams. Additionally, crayfish have been experimentally observed to retain microplastics within tissues that originate from their habitat. In our study, we use a common, introduced species of crayfish, *Faxonius cristavarius*, due to their unique feeding habit of filtering sediment through their gills, increasing their susceptibility to microplastic intake. Collecting *F. cristavarius* across different levels of habitat-degraded streams throughout Montgomery County, VA, can inform us how urbanization may influence microplastic abundance within stream organisms. We predict a higher level of microplastics in crayfish collected from degraded streams due to increased urbanization reflective of those present in sediment and water sampled, indicating accumulation can arise from both ingestion and ventilation. Determining microplastic retention rates in crayfish, at the base of the aquatic food chain, can provide an understanding of potential bioaccumulation to upper trophic levels. Additionally, examining the habitat degradation gradient can determine areas of concern for potential biomagnification of microplastics.

P87 - Assessing competition and identifying optimal reintroduction conditions for the American Chestnut (*Castanea dentata*)Seth McCullough*James Madison University, Harrisonburg, VA*

After the introduction of two pathogens in the late 19th and early 20th centuries, American Chestnut (*Castanea dentata*) populations were drastically diminished, and remaining individuals were reduced to recurrent stump sprouts. American-Chinese chestnut hybrids and transgenic American chestnuts now offer hope of reintroducing a form of this species back into Eastern forests. Most studies on chestnut growth have been conducted in orchards and few short-term studies have evaluated their performance in forest environments. This is the first study that has evaluated the longterm comparative success of hybrids in a heterogeneous forest environment. In this long-term experiment (15 years), we compared the growth and survival of planted American and hybrid chestnuts to Tulip Poplar (*Liriodendron tulipifera*) and Chestnut Oak (*Quercus montana*). Treatments were designed to evaluate effect of varying site conditions (light and moisture availability) on growth and regeneration of the three species. There were significantly greater *Liriodendron tulipifera* saplings in all treatments except small xeric plots ( $p<0.001$ ), while within small xeric plots, there were significantly more *Castanea dentata* and *Quercus montana* saplings ( $p<0.001$ ,  $p=0.021$ ). Additionally, *Liriodendron tulipifera* trees had significantly lower relative basal areas within xeric treatments compared to mesic treatments ( $p=0.038$ ,  $H(2)=8.452$ ), and *Liriodendron tulipifera* had significantly greater relative basal area compared to the other two species within both mesic treatments but did not differ significantly within xeric treatments ( $p=0.034$ ,  $H(2)=6.788$ ). Our results suggest that when resources are abundant (e.g., high-light, mesic sites) fast-growing competitive species, such as *Liriodendron tulipifera*, outperform slower growth species, like chestnut and oak. However, in areas with low resource availability (e.g., low light, xeric sites), *Castanea dentata* and *Quercus montana* outperform *Liriodendron tulipifera* in the long-term. We suggest that forest management practices, such as initially reducing the presence of competitive understory species, may benefit the success of chestnut plantings in more optimal environmental conditions.

P88 - Biomonitoring of zebra mussels (*Dreissena polymorpha*) in a reservoir lake using environmental DNA (eDNA)Amy Rodriguez, Margi Flood, Hannah Fontenot*University of North Georgia, Oakwood, GA*

Environmental DNA (eDNA) recovery and detection via polymerase chain reaction (PCR) is an emerging tool in biomonitoring studies and may be appropriate for the detection of difficult to detect target species, including freshwater organisms. Zebra mussels (*Dreissena polymorpha*) are a freshwater invasive species of concern with a rapidly expanding range in North America. Early detection of newly established invasive populations may minimize ecological damage and improve management outcomes. In April 2021, a substantial number of zebra mussels were discovered attached to a boat destined for a reservoir lake (Lake Sidney Lanier) located in North Georgia. This watershed has neither a known zebra mussel population nor an established biomonitoring system to detect an introduction. In June and July 2022, water samples were collected from six defined site locations (three bi-weekly collections per location). We tested for the presence of *D. polymorpha* eDNA using Qiagen's DNeasy PowerSoil Pro kit for eDNA isolation and species-specific mitochondrial cytochrome b gene region PCR primers. Additional samples were tested from singular collections at three site locations near heavily used marinas (identified as likely sites of potential introduction through collaboration with Georgia Dept. of Natural Resources personnel) and the Tennessee River, a site of known zebra mussel infestation. Target DNA was successfully recovered and detected from the known inhabited site. No target DNA was identified in samples collected from the lake under investigation. Results indicate that the study design and methodology is an effective model for future studies that may benefit from the aquatic recovery of eDNA. The study further supports continued investigation and development of eDNA as a tool to identify the presence of difficult to detect species of interest, which may help guide efforts for watershed management and conservation.

## P89 - Evaluating Prescribed Fire for Blight Resistant B3F3 Hybrid American Chestnut Seedling Introduction

Felix Stith, Mike Madritch*Appalachian State University, Boone, NC*

The functional extinction of American chestnut (*Castanea dentata*) from invasive chestnut blight (*Cryphonectria parasitica*) and widespread wildland fire suppression has significantly impacted the diversity and resiliency of the deciduous forests of the southern Appalachian region. Several American chestnut restoration strategies are being tested. Breeding programs have created Chinese-American B3F3 hybrid chestnut trees that have some level of resistance to chestnut blight and physical growth close to pure American chestnut. This hybrid holds potential to aid in restoration of the American chestnut on a forest scale. A knowledge gap exists regarding hybrid chestnuts' response to various environmental factors within natural settings. American chestnut has several fire-adapted traits and may be fire dependent in certain ecosystems. We hypothesized that B3F3 hybrid chestnuts would have higher growth rates when planted in areas that had previously been treated with prescribed fire. We tested this by establishing plots in Yancey and McDowell Counties, NC in the southern Appalachian Mountains. Part of each plot was burned and then planted with two-year-old B3F3 hybrid chestnut seedlings. Unburned control plots were established adjacent to burned plots and were also planted with two-year-old B3F3 hybrid chestnut seedlings of the same genetic stock. Soil CN samples were taken on a 10x10m grid throughout the plots. Light measurements and CN leaf samples were collected for each seedling in the study. After two growing seasons, B3F3 hybrid chestnut seedlings in the burned plots were taller and had larger stem diameters than seedlings planted in unburned plots. Understory light availability was the primary driver of stem growth speed. These findings suggest that prescribed fire may be an effective silvicultural tool for introducing B3F3 hybrid chestnuts in certain forest communities.

P90 - Barcoding reveals cryptic diversity within the *Cambarus dubius* species complex

Quentin LaChance, Michael Gangloff, John Wells

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This research project is a phylogeographic investigation of *Cambarus dubius* in Western North Carolina. *Cambarus dubius* is a species complex with a wide range of morphological traits, primarily concentrated in the central and southern Appalachian mountains. In recent years new species have been identified within this complex based on genetic data and unique color morphs to diagnose cryptic taxa. Recent descriptions include *Cambarus adustus* (Dusky Mudbug- brownish orange) from northeastern Kentucky, *C. pauleyi* (Meadow River Mudbug- blue) from south-central West Virginia and *C. loughmani* (Blue Teays Mudbug) from the ancient Teays River Valley in West Virginia. These taxa are genetically divergent from topotypic *C. dubius* ss (orange) in the Allegheny Mountains and central Appalachian Plateau. The goal of our research project is to provide additional insights into the taxonomy and phylogeography of the *C. dubius* species complex in the southern Appalachian mountains. To date, we have examined mtDNA sequences from 11 specimens from the New and 2 specimens from the Catawba drainages. All of these individuals are bright blue and exhibit a high degree of genetic divergence when compared with taxa in the northern Appalachians, including topotypic *C. dubius*. We hope to examine specimens from other Interior and Atlantic Slope drainage including the Yadkin and Watauga drainages in the near future to assess regional genetic diversity, delineate geographic species boundaries, and diagnose morphological traits for what appears to be a cryptic taxon in the southern Appalachians.

P91 - Implementation of a Water Monitoring Program in the Irwin Creek Watershed of West Charlotte, North Carolina

Rashawna Huntley, Mark Dugo

*Johnson C. Smith University, Charlotte, NC*

The systematic monitoring and assessment of water quality in urban surface waters is important for maintaining public health and preserving ecological integrity of our watersheds. Urban creeks are frequently burdened by a variety of environmental stressors that can be ameliorated through stewardship that implements best management practices. Johnson C. Smith University (JCSU) is situated in the Irwin Creek subwatershed of the Catawba River Basin in West Charlotte. Irwin Creek is listed as impaired according to Section 303d of the Clean Water Act. Herein, we present on the early stages of a budding water monitoring program for Irwin Creek that is being initiated by JCSU's Center for Renewable Energy and Sustainability. Our focal area in the Beatties Ford Road Corridor of West Charlotte is comprised predominately of African Americans. Our water program is largely intended to serve as a means to educate and engage the community in order to improve stakeholder engagement in local environmental issues. It is predicted that a localized water monitoring program will provide the necessary data and community participation to best inform decisions aimed at improving the overall water quality of Irwin Creek. Our water monitoring program includes a) citizen science-based monitoring for E. coli along greenway corridors, b) JCSU student research and training in the use of eDNA technology as a diagnostic tool, c) physicochemical water quality monitoring using handheld meters and the deployment of long-term data loggers at strategic locations throughout the subwatershed. We predict that this program will help increase agency among local stakeholders in a manner that is reflective of the population residing in the area and contribute to an improved understanding of the current state of water quality in Irwin Creek. Ultimately, we anticipate that these efforts will benefit the long-term goals of restoring and maintaining ecological integrity of Irwin Creek.

P92 - Natural history observation of the Appalachian primary burrowing crayfish *Cambarus* aff. *dubius* across the central portion of its range.

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*Cambarus* aff. *dubius* (*C. dubius* from herein) is a widespread species complex that occurs in the Central and Southern Appalachian Mountains. Seepage wetlands, stream banks, mesic forests, and riparian corridors all harbor *C. dubius* populations, as well as anthropogenic habitats like roadside ditches and agricultural fields with high water tables. This habitat plasticity results in a wide *C. dubius* distribution throughout the Appalachians. During the summer of 2022 a survey was initiated to collect *C. dubius* from the southern and central portion of its range, gathering specimens for morphological and phylogenetic analysis with the intent to describe unnamed taxa and revising the complex. During these surveys, several previously unpublished ecological observations were made. Herein we document habitat preference and burrow morphology for *C. dubius* and attempt to create a distribution map for each phenotype. Results from this study will educate managers on the biology of this understudied species, as well as provide a map to guide future survey efforts.

P93 - Dune Building and Cross-Island Connectivity on Virginia Barrier Islands

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<sup>1</sup>*Virginia Commonwealth University, Richmond, VA*, <sup>2</sup>*University of Virginia, Charlottesville, VA*

Barrier islands exhibit multiple distinct habitats including the beach, dune, and swale that are differentiated by changes in elevation, morphology, and vegetation. As an important part of barrier island resistance and resilience, dune development can influence swale habitat productivity and species richness. The goal of this research is to understand 1) patterns of sediment accretion, soil characteristics, and changes in dune structure and

2) effects on swale biota. Plots were established on two barrier islands with different disturbance responses (Hog and Metompkin), and were placed in newly formed embryonic dunes, on the dune face, as well as in the swale. At each plot, percent cover, stem count, and sediment accretion were measured seasonally. Sediment cores were collected to assess organic matter content, soil chlorides, and bulk density. Annual drone flights were conducted to create digital elevation models, which were used to calculate least cost paths from beach to swale. Our results suggest that dune building is occurring at a higher rate on Hog Island as a result of higher percent cover and higher stem densities among dune building grasses. Increased rates of dune building reinforce protection of the swale habitat from disturbance events, which can allow for higher rates of productivity. Least cost path analysis suggests that mean path cost on Hog Island is increasing with time, while it is decreasing on Metompkin Island. A higher path cost means that it is more difficult for disturbance driven saltwater or sediment to reach the swale habitat. Future changes in sea level rise, intensity and frequency of severe weather, and species distribution will continue to modify the barrier island ecosystem. Improving our understanding of island response and habitat connectivity will help with predictions of future changes on regional, island-specific, and ecosystem scales.

#### P94 - Exploring the Evolutionary Advantage of Superfetation using a State-Dependent Life History Model

Dr. Rebecca Hale, PhD, Andie Lambeth

*University of North Carolina Asheville, Asheville, NC*

Superfetation, the ability to carry multiple broods at once, has been observed in many species, but is most commonly found in *Poeciliidae* fish. Past research used a state-dependent life history model in R to realistically predict the reproductive decisions of the mother, but did not result in superfetation as a viable decision. Further research will modify this model by adjusting the minimum resource requirements for viability and resource allocation among embryos. Previously, resources were spread evenly across embryos regardless of their age, while realistically more resources would go towards larger, older embryos. This new model will hopefully show what environmental factors trigger the evolution of superfetation and produce testable predictions for future work with *Poeciliid* fish.

#### P95 - Impact of Impervious Surface on Tree Growth at NC State

Nicole Garcia, Steph Jeffries

*NC State University, Raleigh, NC*

Trees provide important benefits in urban environments, but also face challenges. They contribute many ecosystem services, such as cooling, carbon capture, and stormwater control. However, urban environments are hard places for trees to grow. For example, impervious surfaces can negatively affect trees due to their hard surfaces limiting water and nutrients and restricting root growth. Although the NC State campus has more than 10,000 trees, it also has various types of impervious surfaces. These include patios, decks, streets, parking areas, driveways, and sidewalks. We wanted to measure the effect of impervious surface on tree growth at NC State. We tagged more than 100 trees and measured their diameters at breast height (DBH) in 2019, 2021, and 2022 to estimate growth. In addition, we calculated the percentage of impervious surface surrounding each tree using the pace-to-plant method (Youngsteadt et al. 2016). We were surprised to discover no relationship between the percentage of impervious surface and the percent diameter growth over three years. Our observation is that there are many other factors that can impact tree health and growth such as disease, location, and tree management that might supersede the effects of impervious surface. These factors not only determine overall tree health and growth, but also help us understand how to better care for our trees on campus and promote sustainability. P96 - Environmental and biotic drivers of disease presence in Floridian and Puerto Rican coral reefs

Brianna Casement, Nicholas Green

*Kennesaw State University, Kennesaw, GA*

**Question:** We investigated environmental factors driving coral biodiversity and disease prevalence in the Florida Keys and Puerto Rico. Coral reefs face numerous threats related to anthropogenic pollution and climate change. Recent disease outbreaks within South Florida populations of stony coral (Cnidaria: Scleractinia) have been the cause of significant concern; however, the causes and factors underlying the spread of these diseases remain little understood. One hypothesis that may explain disease prevalence in these corals is the dilution effect, which states that as host communities become more diverse, pathogen prevalence decreases because pathogens will be less likely to encounter a suitable host. **Hypothesis tested:** We tested the hypothesis that disease transmission among coral reefs is affected by coral colony community structure because more diverse communities may have lower disease transmission rates due to the dilution effect. **Methods:** We utilized publicly available coral monitoring data in conjunction with data on geographic, climatic, hydrologic, and other environmental variables to examine what factors might contribute to the transmission of these diseases. We used hierarchical Bayesian generalized linear mixed models (GLMM) to account for environmental factors and analyze the effects of species evenness on the likelihood of diseases being present. **Results:** Coral status was driven by latitude, bottom rugosity, and depth. Across all coral species, disease prevalence was driven primarily by latitude and coral community evenness within each reef.

**Conclusions:** Sites with greater species richness and evenness typically had lower disease presence, while sites with lower species richness and evenness were more likely to have diseases present. This finding supports our hypothesis that coral reef diseases among Floridian and Puerto Rican reefs exhibit a host dilution effect. These findings may provide new insight for management practices in these regions.

#### P97 - Canopy Gap Influence on Heterotrophic and Autotrophic Components of Soil Respiration in a Southern Appalachian Mixed-

Oak Forest

Kevin McCarthy<sup>1</sup>, Jodi Forrester<sup>1</sup>, Chris Maier<sup>2</sup>, Tara Keyser<sup>3</sup>

P98 - Effect of Increased Salinity Concentrations on Gill Morphology and Gene Expression in Redear Sunfish (*Lepomis microlophus*)

Joel Haley

*Winthrop University, Rock Hill, SC*

Changes in gill morphology have been observed in certain species of freshwater fish in response to increases in salinity. The purpose of this study was to observe the salinity tolerance and the osmoregulatory responses through gene expression and changes in gill morphology of a local freshwater fish (Rear sunfish, *Lepomis microlophus*). Four treatment groups with six fish each were designed to test both short term (24 hr) and long term (96 hr) exposure to hypersaline water (17 ppt). Fish exposed to 17 ppt salt water had significantly increased plasma osmolality and interlamellar cell mass to gill length ratios compared to control groups. Early testing indicates the expression of genes associated with stress and osmoregulation as well. In total, Redear sunfish exposed to increased salinity seem to display some level of osmoregulatory behavior as demonstrated by gill tissue changes and expression of relevant genes, however, fish were unable to correct the osmotic change by 96 hours sustaining dangerously high plasma osmolality.

P99 - Survey of Disjunct Red Spruce in the Alarka Laurel Basin, North Carolina

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<sup>1</sup>*University of North Carolina at Chapel Hill, Chapel Hill, NC*, <sup>2</sup>*Highlands Biological Station, Western Carolina University, Highlands, NC*

Red spruce (*Picea rubens*) occurs in disjunct populations in the southern Appalachians. Red spruce populations in these low latitudes are likely the first to show the impacts of climate change, and thus act as models for understanding past and future impacts of a changing climate. We conducted a vegetation survey in the Alarka Laurel basin in the Nantahala National Forest to assess changes in the red spruce population and other vegetation in this basin since an initial survey in 2007. Using five 10-m-wide belt transects, we measured the diameter at breast height (d.b.h.) and recorded the location of adult red spruce, measured the height of seedlings, and sapling d.b.h. We also recorded the d.b.h. of other trees that were present in the canopy. Every 20 m, we recorded the percent coverage of understory vegetation in a 5 m x 10 m plot, noting the percent of open sky present. We analyzed adult red spruce for evidence of pest infestation. Adult red spruce accounted for 23.9% of overstory trees, with hardwoods dominating the remainder. Great rhododendron (*Rhododendron maximum*) and mountain laurel (*Kalmia latifolia*) made up most of the understory, limiting light availability. Red spruce seedlings and saplings were present throughout the basin, with a weak positive relationship between height of seedlings and percent of open sky in observed understory plots. Our assessment indicates that the mean d.b.h. of adult red spruce trees in 2007 was significantly greater than with our study ( $p = 0.0443$ ); however, there was little change in the height of red spruce seedlings ( $p = 0.653$ ) compared to 2007. We suggest that this population of red spruce has remained stable with a large seedling bank and is unaffected by climate, despite its southern latitude and relatively low elevation.

P100 - Hybridization and Genetic Swamping of *Packera serpenticola* by *Packera anonyma*

Katie Dyson<sup>1</sup>, J. Brandon Fuller<sup>2</sup>

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Soils in mature forests are highly dynamic carbon pools influenced by a myriad of above- and belowground factors. Maintaining the net carbon balance of these soil pools requires greater sequestration and storage than loss through respiration. Understanding how these components respond to disturbance is important with predicted future increases in canopy disturbance rates with climate change, invasive insects, and diseases. This project sought to quantify the influence of experimental canopy gaps on the component sources of soil carbon dioxide (CO<sub>2</sub>) efflux. Canopy gaps change light availability, air and soil temperature, soil water availability, and gross primary production, all of which directly influence biological processes regulating soil respiration. Due to increased light availability and throughfall, we hypothesized that 1) soil respiration would be greater in gaps than the surrounding forest and most variable at gap edge, 2) the size of the opening and the distance from gap edge would influence the magnitude and direction of the component sources, 3) the heterotrophic component would be stimulated by the higher soil temperatures, while the autotrophic component would be depressed by the decreased belowground carbon allocation of plants competing for light. Instantaneous soil CO<sub>2</sub> flux was measured in two gap sizes (0.1 ha and 0.4 ha) and in an unharvested control in Pisgah National Forest, NC, June–November 2022. Paired root exclusion (RE) and non-root exclusion (NRE) collars were measured at 5–6 locations along a transect starting in gap center and extending north into the surrounding closed canopy forest. Soil temperature and moisture measurements were collected concurrently with soil flux. Analysis of variability of flux components and microclimate responses by location and size are underway. By isolating the components of soil respiration, this study will contribute important detail to our understanding of how canopy gaps influence the carbon balance of mature mixed-oak forest ecosystems.

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*Packera serpentifolia* (Boufford, Kartesz, S.H. Shi & R.C. Zhou) is endemic to a single location, the Buck Creek serpentine barrens of Clay County, North Carolina. Serpentine barrens are characterized as ultramafic with elevated levels of nickel, cobalt, and chromium. *Packera anonyma* (W.A. Weber & Á. Löve) is known to occur on serpentine soils and co-occurs at Buck Creek alongside *P. serpentifolia*. *Packera anonyma* readily hybridizes with other *Packera* species and genetic swamping by *P. anonyma* has impacted congeners that are also geologically constrained, such as *P. millefolium* (W.A. Weber & Á. Löve). This makes *P. serpentifolia* susceptible to hybridization and genetic swamping from *P. anonyma*, which could place further stress on its small and geologically constrained population. While hybrids have not officially been recognized at Buck Creek, the authors of this study and others have observed individuals with intermediate traits of both parent species. Vouchers will be collected in March of 2023 when individuals will be in flower for *P. serpentifolia*, *P. anonyma*, and putative hybrids. To determine the extent at which hybridization is impacting the population of *P. serpentifolia*, we will use NextRAD genome skimming to perform a hybrid parentage analysis, which will generate the percent parentage of *P. serpentifolia* and *P. anonyma* in our samples. We will also map out hybrid zones at Buck Creek, which can be used to aid in conservation efforts to target areas that are potentially impacted more by hybridization and genetic swamping. We suspect that genetic swamping of *P. serpentifolia* by *P. anonyma* is occurring at Buck Creek and could potentially cause negative effects on *P. serpentifolia* populations.

#### P101 - Does stream restoration benefit aquatic insect communities?

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*Davidson College, Davidson, NC*

Data are equivocal on the efficacy of stream restoration projects to restore degraded streams and benthic macroinvertebrate communities, especially taxa sensitive to stream degradation, sedimentation, and organic pollution, such as stoneflies. Small streams in the North Carolina Piedmont near Charlotte are often deeply incised and highly degraded. Grading and stabilization of incised banks is usually part of restoration projects but are extremely disruptive. In-stream modifications such as deflectors, weirs, step-pools, and engineered log jams create habitat heterogeneity, but may not succeed if degradation, erosion, and sedimentation continue in the watershed. A one-size-fits-all approach to restoration will not achieve the same results in every stream, yet tends to be the proposed solution by environmental engineers. To test the effects of stream restoration projects and collect aquatic insect community data pre-restoration, we assessed aquatic benthic macroinvertebrates in three streams. Ramah Creek flows through the ~1,000 acre Ramah Creek Preservation Area protected by conservation easements and has undergone limited in-stream habitat modifications and improvements. Torrence Creek in the Torrence Creek Greenway has undergone several restoration projects over the past decade. West Branch Rocky River in Abersham and Fisher Farm Parks is currently undergoing a major stream restoration project. Using standard kicknet protocol, we found that Ramah Creek had the greatest diversity and abundance of EPT taxa, especially of the sensitive stoneflies. West Branch Rocky River had the second most EPT abundance, and Torrence Creek had the lowest EPT diversity and abundance. We found little evidence supporting the effectiveness of restoration efforts in Torrence Creek, to the detriment of overall stream habitat quality. Heavy development in the watershed contribute to erosion and high sediment loads entering the stream, which reduce the effectiveness of restoration efforts. Without watershed-level protection, the planned restoration to restore West Branch Rocky River will similarly not achieve the desired results. P102 - 2022 Population Review of Three Federally Listed Outcrop Species

Caitlin Brown<sup>1</sup>, Melissa Caspary<sup>2</sup>

<sup>1</sup>*Georgia Gwinnett College, Lawrenceville, GA*, <sup>2</sup>*Georgia Gwinnett College, Athens, GA*

For only a few early months out of the year, the exposed granite outcrops throughout the Piedmont (ranging from Alabama up through South Carolina) support rare plant life in ephemeral pools. This habitat is threatened by quarrying, development, agriculture, a changing climate, invasive species, traffic, and dumping. *Gratiola amphiantha*, *Isoetes melanospora*, and *Isoetes tegetiformans* are three Federally listed plant species that only occur in these seasonally temporary pools. The purpose of this study was to locate and re-evaluate a number of these populations, previously surveyed 2007-2008 (Caspary 2008), focusing primarily on the populations that were identified in that survey as the most threatened. Visits to pristine sites served as a guideline for the optimal environmental conditions these species need to survive. The density of plant populations, pool sizes, water samples, and local threats were observed at each site visited. Water samples were analyzed for carbon, nitrogen, and pH. This data was compared to the 2007-2008 survey data, which helped establish the changes in the sites over time. We expected to find that in sites noted as threatened in the 2007 - 2008 survey, the conditions would not have improved in the most poorly rated sites, or that the plants previously identified at these sites could be gone entirely. We also hypothesized that plant populations at conserved sites remained at equilibrium and that their rating would not change after reviewing. The results of the survey demonstrate a clear trend towards further and continued degradation and reduction of available habitat for the federally listed species as evidenced by decreasing site quality and species population data.

#### P103 - Assessing Bacterial Diversity in Local Aquatic Environments: A Metabarcoding Study of Polluted/Unpolluted Surface Freshwater Bodies and Adjacent Sediments

Jacqueline Valiente, Analuz Nieves, Tammy Laberge

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In the environment, a wide range of organisms, including plants, animals, and microorganisms, leave behind genetic material termed environmental DNA (eDNA). Environmental DNA can be collected in local habitats, for example in air, water, sediment, snow, soil, etc. In our study, aquatic samples were obtained from a diversity of freshwater bodies that were collected from previously polluted and traditionally clean sampling sites

(estuaries, tributaries, man-made waterways, lacustrine environments, and lentic environments). A multifaceted approach was employed for the collection and filtration of water samples, which involved the use of two different filters with different pore sizes (0.22 µm and 0.45 µm). Further, in an exploratory effort, sediment samples were collected from the surrounding areas of the previously obtained water samples. Following collection, samples were processed for DNA extraction and quantification analyses. Recently, the V4 hypervariable region of the prokaryotic 16S ribosomal RNA (rRNA) gene was amplified by polymerase chain reaction (PCR) using a universal primer set (515f and 806r). PCR and gel electrophoresis provided information about the amplicon size and quantity of the amplified DNA fragments. As a result of our analyses, our current research focuses on sequencing approximately thirty candidate PCR amplicons using next-generation sequencing and using bioinformatics tools to perform DNA metabarcoding. Our goal is to determine the local bacterial diversity and abundance present in each sample. Studying the diversity and abundance of freshwater local microbiomes is crucial as they play a vital role in ecosystem functioning, impact larger organisms' health and survival, and aid in identifying environmental stressors and conservation efforts.

**P104 - Evidence of More than Two Variations of Eastern and Western Meadowlark (*Sturnella magna* and *Sturnella neglecta*)  
Vocalizations in Areas of Geographical Overlap Suggests Further Speciation or Hybridization of These Two Species.**

Amanda Cordle, Dr. John Quinn

*Furman University, Greenville, SC*

There are three avian groups known to have the capacity for vocal learning; specifically the families Trochilidae, Psittacidae, and Passeriformes. Based on the diversity of songbird species, it has long been hypothesized and theoretically demonstrated that vocal learning is a leading indicator of speciation. Because bird song functions as a tool for mate attraction, it has been suggested to be the most important prezygotic isolation mechanism in avian species. Since vocal divergence often drives speciation, it can be hypothesized that vocal divergence may lead to species hybridization. To test if song can serve as a leading indicator of species differentiation, we tested if songs of Eastern and Western Meadowlarks (*Sturnella magna* and *Sturnella neglecta*) in North America could be broken into sub-groups and perhaps sub-species. These two closely related species, which are separated in part by their distinct songs, co-exist in central North America. Through unsupervised classification of thousands of meadowlark field recordings and the use of ML software to group the recordings, we found evidence of variation within and between meadowlark species in the regions, adding evidence that there may be more than two species of meadowlark in the area. This also provides evidence that new data improves our ability to detect speciation occurring in avian communities.

**P105 - Comparison of environmental factors and macroinvertebrate community structure in small, human-constructed ponds.**

Helena Loucas, Kenneth Fortino

*Longwood University, Farmville, VA*

Small human-constructed ponds are an abundant habitat globally and can function as biodiversity and biogeochemical hot spots. However human-constructed ponds are not structurally or functionally homogeneous due to variation in design, watershed characteristics, and organic matter and nutrient loading, etc... The diversity and abundance of macroinvertebrates in aquatic systems have been shown to be sensitive to abiotic conditions and disturbance and therefore we may expect macroinvertebrate communities to vary with differences in the structural and functional characteristics of human-constructed ponds. Alternatively, dispersal limitation or other landscape-scale factors may limit macroinvertebrate community structure and obscure the effects of local environmental conditions. We compared macroinvertebrate community structure and structural and functional characteristics of two human-constructed ponds of contrasting design. Campus pond is a 0.1 ha. shallow (0.7 m, maximum depth) storm-water retention pond enclosed by a vertical concrete wall in an urban watershed. Chalgrove lake is a 1.2 ha. pond with a 4 meter maximum depth in a less developed watershed. We described macroinvertebrate community structure using Hester-Dendy samplers placed in the littoral zone of each pond. Environmental conditions in the pond were characterized by continuous high-frequency sampling of dissolved oxygen, temperature, and conductivity. Additionally, we monitored depth in Campus Pond, since it could vary dramatically during rainfall events. We found that the ponds differed in their environmental characteristics with Campus Pond showing much greater variation in all measured variables, including frequent and extended anoxia. Although the ponds contained similar taxa richness, Chalgrove Lake had greater taxa diversity which would be consistent with a limiting effect of the environmental conditions of Campus Pond. Understanding the ways that the design and placement of human-constructed ponds affects biodiversity can inform decisions around pond construction and conservation goals.

**P106 - Crayfish of the Cumberland Overthrust Block and its influence on crayfish dispersal into the northern realms of North America**

Roger F. Thoma

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In the Appalachian Mountains a geologic formation called the Cumberland Thrust Block (CTB) is found between the southern portion (NC, TN, GA) of the mountain range and its northern areas (VA, WV, PA). The structure follows the strike of the Appalachian Mts in a southwest to northeast direction with its southern edge just south of the TN/KY-VA border and its northern border at the WV, VA, & KY border intersection. It is located at the eastern edge of the Cumberland Plateau. The CTB appears to have served as a migratory corridor for ancestral southern Appalachian crayfish into northern North American territories. Examination of the crayfish faunas associated with CTB reveals notable relationships between southern representatives of Cambarid species complexes and their northern representatives. Species groups discussed and illustrated in this paper are *Cambarus* aff. *dubius*, *Cambarus* aff. *sciotoensis*, *Cambarus* aff. *veteranus*, *Cambarus* aff. *distans*, *Cambarus* aff. *chasmodactylus*, *Cambarus* aff.

*robustus*, and *Cambarus* aff. *bartonii*. The genus *Faxonius* does not appear to have used the CTB as a dispersal route. Geologic and genetic timelines are compared.

P107 - Evaluation of headwater tributaries in a historically industrialized corridor of the Black Warrior River watershed in Birmingham AL.

Mark Meade, Whitney Dollins, Nicholas Foster, Lawrence Fountain, Albert Key, Sophie Lovett, Deanna Palermo, Owen Phillips, Cassandra Ruiz, Noah Scoggins, Louis Smith, Joseph Swing, Caroline Teal, Eli Vyhlidal, Paige Washington

*Birmingham Southern College, Birmingham, AL*

The city of Birmingham Alabama is well known as an industrial city. Beginning in the 20th century, steel and coal industries were the dominant revenue producers and, from their inception, resulted in major impacts to the flora and fauna of the region. Although the steel and coal industries have subsided, their impact is still prevalent in the region. We used EPA Rapid Bioassessment Protocols (RBPs) to examine fish assemblages and habitats in four major headwater streams (Village creek, Five-Mile creek, Beaver creek, and Turkey creek) in the NE corridor of the city to assess current status in comparison to historical fish assemblage data. These streams can be found in the center of Birmingham (Village creek) and increase in distance until reaching Turkey creek approximately 15 mi. from the city center. Two teams of surveyors worked at each site collecting fish downstream and upstream at any site as well as conducted habitat assessment along each surveyed reach. Water quality parameters were also examined at each site and included D.O, temp, pH, conductivity, TDS, chlorine, copper, nitrate, and phosphorus measurements. In total 1,471 fish comprising 18 species were captured, identified, and released following enumeration at each site. Most sites had poor IBI scores and were dominated by common headwater tolerant minnow and sunfish species including stoneroller (*Campostoma oligolepis*), creek chub (*Semotilus atromaculatus*), green sunfish (*Lepomis cyanellus*), and bluegill sunfish (*Lepomis macrochirus*). Sensitive fish taxa, such as darters, were lacking at sites near the city center but gradually were observed farther in the suburbs. The majority of darter species were observed in Turkey creek and included the statelisted Vermilion darter (*Etheostoma chermocki*) and Rush darter (*Etheostoma phytophilum*). Habitat and water quality scores were poor/marginal for most sites except for Turkey creek. To date, none of these sites show improvement compared to historical data. P108 - Quantifying cyclic di-AMP in *Staphylococcus aureus* during stress using a competitive ELISA

Carissa Her, Noela Moraga, Melinda Grosser

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*Staphylococcus aureus*, a Gram-positive bacterium, is one of the leading causes of life-threatening bacterial infection in the US, partly due to its antimicrobial resistance. If infected by this bacteria, diseases like pneumonia, skin and soft tissue infections, sepsis, and endocarditis may arise. *S. aureus* uses a second messenger called cyclic di-adenosine monophosphate (c-di-AMP) to regulate many cellular functions such as peptidoglycan synthesis, stress response, and more. Our data show that high c-di-AMP levels can be toxic during nitric oxide (NO<sup>-</sup>) stress, and others have shown that low c-di-AMP levels result in reactive oxygen species (ROS) toxicity in *S. aureus*. Thus maintaining normal c-di-AMP is essential for proper cell functions, especially during stress. In this research, we aim to optimize a competitive enzyme-linked immunosorbent assay (ELISA) to quantify specific concentrations of c-di-AMP in *S. aureus*, with the ultimate goal of understanding how c-di-AMP levels change during stress and what levels are optimal. For assay development, we used three strains: *S. aureus* containing a vector-control of an anhydrotetracycline (aTc)-inducible plasmid (pRMC2), and *S. aureus* over-expressing the genes encoding DacA (produces c-di-AMP) or GdpP (hydrolyzes c-di-AMP) from pRMC2. Growth conditions and lysate purification methods were tested with these strains in conjunction with a commercial c-di-AMP ELISA kit, and a protocol was developed to successfully detect different concentrations of c-di-AMP in *S. aureus*. Currently, we are using this new assay combined with stepwise induction of DacA or GdpP via aTc (in the above strains) to determine the precise concentration range of c-di-AMP that can be tolerated in the presence of stressors. We are also using ELISA to calibrate a fluorescence resonance energy transfer (FRET)-based biosensor for c-di-AMP, another means of measuring c-di-AMP in *S. aureus* in real-time. Long-term, these findings may contribute to the development of drugs that alter signaling pathways of antibiotic-resistant bacteria.

P109 - The impacts of nutrient pollution on macroinvertebrate communities in Accokeek Creek and Potomac Creek in Stafford Co. Virginia

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Agricultural practices and suburban development can contribute excess nutrients to freshwater systems and negatively impact biodiversity. Tidal streams, due to high surface area to volume ratios, are more susceptible to effects of excess nutrients, thus making them useful water quality indicators for entire watersheds. Macroinvertebrates are good bioindicators of stream health because they are accessible, sensitive to environmental changes, and taxa can be organized based on pollution tolerance. In this study, we compare macroinvertebrate assemblages in Accokeek (AC) and Potomac Creeks (PC) to better understand the relationship between land use, nutrient pollution, and macroinvertebrate community structure. Both creeks border the forested Crow's Nest Nature Preserve but the AC watershed is more suburban upstream whereas the PC watershed is primarily agricultural. Macroinvertebrates were collected at a total of 10 sites (6 AC and 4 PC) during July 2021. At each site, three core sediment samples were taken, sieved, and stored in 90% ethanol. Organisms were identified to the family level except for Oligochaeta. Water quality parameters including temperature, pH, conductivity, dissolved oxygen, nitrate and orthophosphate were measured at each site. We found no significant differences between AC and PC for the mean nitrate, orthophosphate, and pH. Potomac Creek had significantly higher dissolved oxygen than

Accokeek Creek ( $p=0.037$ ). In our preliminary results, AC had a significantly higher average Shannon diversity compared to PC ( $p=0.019$ ). Oligochaeta was the dominant taxa in PC while Chironomidae was most abundant in AC.

P110 - Classifying the antibiotic resistance of *Escherichia coli* isolates

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Although antibiotics have been vital in treating bacterial diseases, their usefulness is limited by the rising prevalence of antibiotic resistance. Previously, we isolated *Escherichia coli* strains from students at Davidson College, and this research study sought to classify the antibiotic resistance of these natural (as well as laboratory) strains through the disk diffusion and minimum inhibitory concentration techniques. For the disk diffusion assay, isolates were spread on Mueller-Hinton plates and exposed to paper disks impregnated with different antibiotics. Following an overnight incubation, the zones of inhibition were measured. Each isolate was tested against penicillin, chloramphenicol, streptomycin, tetracycline, and ampicillin in their respective dosages. This testing was expanded upon through the minimum inhibitory concentration (MIC) technique, which tested the strains against chloramphenicol, streptomycin, tetracycline, and ampicillin. As expected, the data for penicillin for all isolates consistently suggested resistance, so this antibiotic was not included in further testing. MIC testing was carried out via a microplate array and varying dilutions of each antibiotic, using TSB wells as a control. These arrays were read using a spectrophotometer, then incubated overnight before being read again to measure levels of bacterial growth in each well. As expected, isolates showed higher growth with lower concentrations of antibiotics, although the ranges of growth varied per sample and per antibiotic. Using these data, we were able to classify the antibiotic resistance of numerous *E. coli* strains to enable future research. Furthermore, dual testing allowed for a comparison of the results from the disk diffusion and MIC techniques, allowing further insight into the reliability of disk diffusion in comparison to the more precise MIC testing. P111 - Microbial Diversity of the Cumberland River

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Microbial communities in freshwater habitats are incredibly diverse, and understanding their ecological effects is important in recognizing their role in the environment. However, there is a scarce amount of research done on microbes that are contained within urban waterways, and the type of microorganisms found is important for many reasons, including the public health of populations that use that as a source of drinking water. A study done by the World Health Organization states that more than 5 million people per year die from waterborne diseases<sup>1</sup>. The Cumberland River is a major waterway whose microbial inhabitants have been understudied. Further investigation of the Cumberland's microbial diversity is important because this river is a major source of drinking water for the surrounding regions. A study done in 2021 shows that wildlife are a major source of microbes in American rivers<sup>2</sup>. Animals such as dogs and waterfowl are considered major sources of bacteria, such as *E. coli*<sup>3</sup>. Knowing what microbial contaminants are contained in water could better improve the effectiveness of water treatment by water treatment facilities, improving the quality of drinking water that is consumed by the masses.

P112 - Assessing Biofilm Formation of *S. sanguinis* and Examining its Role in Dual-Species Biofilms

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Many bacterial species are known to form an organized community that can adhere to surfaces, known as a biofilm. *S. sanguinis* is one of the most common species of bacteria found in the oral biofilm better known as dental plaque. This species is thought to be a pioneering species whose colonization aids in the attachment of other microorganisms that are associated with the formation of cavities. Assessing the role of *S. sanguinis* in dual-species biofilm formation will provide insight into oral pathology of dental caries. To create a biofilm, a 96-well plate was used to inoculate a 25-hour culture of *S. sanguinis* in BHI broth and BHI broth with 1% sucrose. After 17 hours of growth, a crystal violet assay was performed to assess biofilm formation. A 0.4% crystal violet solution was used for the assay and a spectrophotometer was used to measure the absorbance values. The results indicate that the methods used were successful for creating and analyzing a biofilm and that *S. sanguinis* had greater biofilm formation in BHI broth supplemented with sucrose compared to BHI broth with no supplement. This information proves that this procedure is effective and can be used for further research on *S. sanguinis* and examining its role in dual-species biofilm formation.

P113 - The Creation and Implementation of a Watershed Management Plan for a Creek with a 200-Year-Old History of Coal Mining Located in the Upper Ohio River Valley

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Watershed Management Plans (WMPs) are created and implemented across the United States to reduce non-point source pollution in waterways. Identification of pollutants and sources of impairment is the first step in managing load reductions and implementing restoration projects. WMPs also outline monitoring efforts, community support/education, financial requirements, and timelines. The Wheeling Creek watershed is increasingly utilized for recreation and is recognized as an economical benefit to local communities. However, Wheeling Creek has a long history in coal mining

and other industry. Bituminous coal in the Pittsburgh coal seam was discovered in early 1800 with the first coal mines opening in Wheeling, WV in 1810. Mine waste went unregulated until 1976, allowing active and abandoned mines to contribute substantial impacts to the creek. Sediments, sulfates, and metals from mine waste increased stream water conductivity and reduced biodiversity.

Additionally, the Wheeling Creek watershed is impacted by high levels of *E. coli*, algal blooms, and biological impairments. Rain events commonly over-burden sewer systems and combined sewer overflows (CSOs) discharge sewage contaminated stormwater into Wheeling Creek and its tributaries. In some parts of the watershed, agricultural run-off is substantial and often includes fecal material, pesticides, and fertilizers- a major contributor of algal blooms from nutrient loading. Furthermore, run-off from roadways and parking lots contains road salt and particulate matter, increasing conductivity.

Like many waterways, the Wheeling Creek watershed crosses state lines, flowing northeast from southern Pennsylvania into the Northern Panhandle of West Virginia. Contaminants from agricultural run-off and other pollutants are carried from PA into WV, but each state has different regulatory structures and policies complicating efforts to improve water quality. Here we document our efforts to create a WMP for the Wheeling Creek watershed- the first step in a multi-state remediation effort for this ecologically, economically, and historically important creek.

#### P114 - Adapting a FRET Based Cyclic-di-AMP Biosensor for *Staphylococcus aureus*

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Methicillin-resistant *Staphylococcus aureus* is a Gram-positive bacterium and an opportunistic human pathogen. It causes infections such as skin abscesses, sepsis, urinary tract infections, and meningitis. Due to antibiotic resistance, *S. aureus* is a major public health concern. One of the contributing factors to its survival in a host is cyclic di-adenosine monophosphate (c-di-AMP). This second messenger regulates many aspects of cellular function in Gram-positive bacteria including the maintenance of osmotic pressure and cell wall homeostasis. In *S. aureus*, an enzyme called DacA generates c-di-AMP from two ATP molecules. C-di-AMP is degraded into 5'-pApA by the phosphodiesterase enzyme GdpP. The deletion of DacA causes growth arrest and sensitization to antibiotics, which indicates the importance of c-di-AMP for bacterial survival. Most c-di-AMP quantification methods require purification of cell lysates; however, to understand how dynamic changes in c-di-AMP are related to antimicrobial resistance and stress responses, it will be useful to have a fluorescence-based biosensor that can be monitored in real-time in live cells. In this project, we are adapting and testing a fluorescence resonance energy transfer (FRET)-based c-di-AMP biosensor created for *Listeria monocytogenes* for use in *S. aureus*. The biosensor includes cyan fluorescent protein (CFP) and yellow fluorescent protein (YFP) linked by a truncated c-di-AMP binding protein (Imo0553) from *L. monocytogenes*. When c-di-AMP is present at high levels, Imo0553 changes conformation to bring CFP and YFP into close proximity, allowing FRET and a change in emission spectra to occur. We are testing the expression of the biosensor from the *S. aureus* plasmid pCN52, with expression driven by either the constitutive *rpsJ* or *rpoD* promoters. The resulting vector will be tested in *S. aureus* strains overexpressing DacA or GdpP and in conjunction with an ELISA to calibrate a standard curve relating FRET results to c-di-AMP concentration.

#### P115 - Water Quality Trends in a 2nd Order Stream Located in the Upper Ohio Valley

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Long Run Creek is a second order stream with a watershed of approximately 18 km<sup>2</sup> located in the Upper Ohio River valley, in Wheeling WV. Forests cover approximately 50% of the watershed while 17% is urbanized and less than 5% is agricultural. Due to this stream's many water quality impairments (chemical and bacterial), water chemistry data has been collected weekly for approximately 5-years as part of water quality monitoring project at West Liberty University, starting in the winter of 2018. Here we report water quality trends in Long Run from 209 datapoints, with the goal of better understanding the stream's physiochemical chemical profile. Mean water temperature was 13.2°C (range -0.2 – 27.9°C) and mean pH was 8.3 (range 7.4 – 9.1). Dissolved oxygen in the stream ranged from 4.84 mg/L to 19.7 mg L<sup>-1</sup> with a mean 11.3 mg/L. Specific conductivity (SPC) and chloride (Cl) concentrations were generally high, with mean SPC 631  $\mu\text{S cm}^{-1}$  (range 326 – 825  $\mu\text{S cm}^{-1}$ ) and Cl 71 mg L<sup>-1</sup> (range 23 –

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Within the Gulf of Mexico, forage fish species serve as an important link between lower trophic levels and higher trophic levels, such as economically valuable predator fish, birds, and mammals. Despite the key ecological role of forage fish, fisheries management efforts are often directed elsewhere; as a result, input data for fisheries models is unavailable for many forage fish species. To fill this knowledge gap, we sought to produce species-specific caloric content values for four forage fish species in the Gulf of Mexico: Atlantic Croaker (*Micropogonias undulatus*), White Trout (*Cynoscion Arenarius*), Bay Anchovy (*Anchoa Mitchellii*), and Gulf Menhaden (*Brevoortia Patronus*). We additionally investigated the impacts of body condition, body length, month of collection, as well as environmental conditions such as temperature and river discharge to determine the response of caloric content to physiological, temporal, and environmental variation. Using bomb calorimetry, we were able to determine the dry energy density of the four forage fish species. Additionally, the dry energy density of both adult Gulf Menhaden, juvenile Gulf Menhaden, and adult Atlantic Croaker was found vary temporally by month and peaking at different times of the year. Furthermore, we found that the dry energy densities of Gulf Menhaden and Atlantic Croaker are positively correlated with fork length. We are currently in the process of investigating the potential relationship between environmental conditions and dry energy density but are encouraged by our completed analyses. Ultimately, our species-specific caloric content values for forage fish in the Gulf of Mexico add much need specificity to current fisheries models, especially since our caloric content values are available for a variety of temporal, environmental, and physiological conditions.

250 mg L<sup>-1</sup>). We found a modest correlation between the Cl and SPC (R<sup>2</sup> = 0.19). When analyzed by season, winter (December, January, and February) and summer (June, July, and August) correlations between Cl and SPC were stronger (R<sup>2</sup> = 0.27 and R<sup>2</sup> = 0.37). This study supports the efforts by the City of Wheeling and a local water quality group to better understand sources of pollution in the watershed and improve water quality in Long Run.

P116 - The bacterial loads of various leafy vegetables and the washing practices to reduce the loads

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In today's fast paced word, bagged ready-to-consume leafy vegetables have become more popular due to the nutritional benefits of leafy vegetables, the convenience, and the perceived safety. Zooming in on the perceived safety, this study aimed to: (1) compare the abundance of the bacterial loads between bagged leafy vegetables and their bunched counterparts, more specifically, kale, spinach, and collard greens, and (2) explore an effective way to clean these leafy vegetables before consumption. We used tryptic soy agar to compare the heterotrophic bacteria, eosin-methylene blue agar to compare the Gram-negative bacteria, and 3M™ Petrifilm™ EC count plates to compare the coliforms and *Escherichia coli* dislodged from vegetables. There were significant differences between bunched and bagged versions of spinach (p < 0.05), kale (p < 0.05), and collard greens (p < 0.05). However, there were no significant differences in Gram-negative bacteria or coliforms among vegetables. We also found that water and the commercially available Veggie Wash dislodged a similar number of bacteria when washed 3 times (p > 0.05). Compared to water and Veggie Wash, solution of one part vinegar and three parts water was the most effective alternative to kill dislodged bacteria from vegetables. To further investigate the effectiveness of vinegar, we treated spinach leaves from bags, due to their high bacterial loads, in a serial of diluted vinegar solutions for various durations. Our preliminary results showed that it took 3.125% vinegar 7 hours to exhibit bactericidal effect. In summary, our results showed that bagged leafy vegetables have higher bacterial loads than their bunched counterparts and washing leafy vegetables with diluted vinegar solutions can effectively lower the bacterial loads and make them safer to consume. Future research will finetune the washing practice with various concentrations of vinegar and treatment times and extend the practice to other produce.

P117 - Understanding the physiological, temporal, and environmental determinants of forage fish caloric content in the Gulf of Mexico

Rebecca Walsh<sup>1</sup>, Robert Leaf<sup>2</sup>

P118 - Honeybee Wings: Antibiofouling and Antimicrobial surfaces

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Natural surfaces with remarkable properties and functionality have become the focus of intense research. Heretofore, the natural antimicrobial properties of insect wings have inspired scientists. The wings of cicadas, butterflies, dragonflies, and damselflies have evolved phenomenal antibiofouling and antimicrobial properties. These wings are covered by periodic topography ranging from highly ordered hexagonal arrays of nanopillars to intricate "Christmas-tree" like structures with the ability to kill microbes by physically rupturing the cell membrane. However, there is a lack of information in the literature on antibiofouling and antimicrobial associated with honeybee wings. Thus, the aim of the present work was to evaluate antibiofouling and antimicrobial properties of honeybee wings. Here, through antimicrobial and electron microscopy studies, we showed that pristine honeybee wings displayed no microbes on the wing surface. Also, the wings displayed antimicrobial properties that disrupt microbial cells, inhibiting their growth. The antimicrobial activities of the wings were extremely effective at inhibiting the growth of Gram-negative bacterial cells compared to Gram-positive bacterial cells. Electron microscopy revealed that these wings were studded with rough, array of sharp pointed pillars distributed on both the dorsal and ventral sides enhancing anti-biofouling and antimicrobial activities. This makes honey wings biomaterials capable of killing bacteria which can be translated into healthcare materials.

P119 - Facilitating nursery production of *Spartina alterniflora* through assessment of fecundity and phenology in coastal Georgia

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Because of the important ecosystem services provided by salt marshes, recent efforts to restore these habitats have increased. Living shorelines, which use green infrastructure and native biota to stabilize shorelines, have been shown to reduce erosion, facilitate marsh accretion, and restore ecological processes. *Spartina alterniflora* is often included as a biotic component in these projects due to its vital role in salt marsh growth and maintenance. Studies show genetic and phenotypic diversity in *S. alterniflora* populations both across its native range and within marshes. Thus, it has been suggested that nursery stock be sourced from local marshes and exhibit relatively high genetic diversity (i.e., propagated by seed) to maximize restoration success. To facilitate nursery production of a local, genetically diverse plant nursery stock in coastal Georgia, information on the best time and marsh location for seed collection and methods of seed propagation is needed. Thus, the goal of this research was to characterize phenology and fecundity of *S. alterniflora* in coastal Georgia. Reproductive status was assessed bimonthly in multiple salt marshes in Chatham County, Georgia, from September to November 2022, and mature spikelets (seed stalks) were collected when available. Collected spikelets were

then processed to collect seeds, total number of seeds per spikelet were counted, and seeds were stored at 10°C in four different water salinities (0, 10, 20, and 30 ppt) for at least three months to break dormancy and examine the effect of salinity on fecundity. Assessment of seed viability and germination under salinity treatments are currently being conducted, and analysis of phenology data is underway. The results from this study will provide valuable information on the best site and time for seed collection and proper storage and germination conditions to facilitate the nursery production of local *S. alterniflora* for marsh restoration projects in coastal Georgia.

P120 - Assessing the Effects of Prescribed Fires on Mycorrhizal Fungal Communities in Upland Mixed Oak-Pine Forests in DuPont State Recreational Forest, NC.

Ari Puentes

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Fungi are often overlooked when planning forest management practices such as prescribed fires, but fungal communities are incredibly important to ecosystem health. Almost all trees have been found to rely on mutualistic fungal symbionts, known as mycorrhizal fungi. This study investigated the effects of prescribed fire on mycorrhizal fungi in Dupont State Recreational Forest in western North Carolina. Prescribed burn units in mixed oak-pine communities that differed only in the time since they were last burned (unburned, 1, 2, and 13 years) were surveyed monthly from June to September 2022. Surveys consisted of photographing, counting, collecting specimens, and taking notes on all mycorrhizal fungi found within the three 0.1 ha<sup>2</sup> research plots per burn unit. Field collection was done by citizen scientists from the Asheville Mushroom Club and UNC Asheville student volunteers. The collected specimens were brought back to the lab for identification and detailed descriptions before they were preserved. Shannon-Weaver diversity index, species richness, and community composition were calculated and compared between burn units. There were no statistically significant differences in any of the diversity indices between the burn units, though the recently burned areas had higher diversity and higher richness than the unburned ones. There were different communities present in the recently burned and unburned areas indicating that certain species react more to fire than others. This preliminary study encourages further research so that forest management methods can take the mycorrhizal fungal communities into account. Viable specimens with DNA barcodes were deposited into the newly created UNC Asheville Fungarium. This information was digitized on the UNC Asheville Fungarium Mycology Collection Portal (MyCoPortal) and uploaded to iNaturalist. The results of this project provide many resources for citizen scientists, students, and researchers to utilize for continued mycological research.

P121 - Molecular analyses and benthic distribution of shallow-water zooxanthellate zoantharians (Cnidaria: Hexacorallia) and their symbionts Symbiodiniaceae along the north-eastern coast of Toco, Trinidad

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Zooxanthellate cnidarians form symbiotic associations with dinoflagellates of the family Symbiodiniaceae, which allow this mutualistic relationship to thrive in nutrient-poor shallow waters of marine habitat. With changing habitat and increased anthropogenic effects, annual surveys of benthic coral reef components are essential ways to determine cnidarian percentage cover, and biodiversity on coral reefs. One species that is most commonly found on Caribbean reefs are zoantharians. Although zoantharians are visibly difficult to accurately identify, molecular analyses present one method to assist in this dilemma. Past studies in this region are only now identifying zoantharians, specifically for two species: *Palythoa* and *Zoanthus* spp. This study assessed zoantharian benthic coverage, species identification, and biodiversity of symbiotic dinoflagellates of the family Symbiodiniaceae. This study utilized benthic line intercept point surveys to determine zoantharian percentage cover at three sites: Grande L'Anse, Pequelle, and Salybia Bays along the northeastern coast of Toco, Trinidad. Molecular analyses used DNA markers, specifically mitochondrial cytochrome oxidase subunit I (COI), mitochondrial 16S ribosomal DNA (mt 16S rDNA), and the nuclear internal transcribed spacer region (ITSrDNA). A total of 5 polyps were collected from separate colonies at three sites with 1-2 polyps used to extract DNA from these cnidarians and their dinoflagellate symbionts. Results showed that zoantharian benthic coverage ranging from 47-74%, with *Palythoa caribaeorum*, *Zoanthus pulchellus*, and *Zoanthus sociatus* as common zoantharians observed at sites. *Palythoa* sp. associates predominantly with *Cladocopium*, and *Zoanthus* sp. with *Symbiodinium* respectively. Although dinoflagellate diversity was observed at the family level, it is important that the next step focus on ITS-2 makers, which can reveal extensive diversity of Symbiodiniaceae at a finer level. Understanding diversity of zooxanthellate cnidarians and their symbionts, may highlight host-symbiont species resistant to trending environmental changes.

P122 - Initial Mycological Survey of Tater Hill Plant Conservation Preserve Fleshy Fungi

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In order to protect biodiversity, one of the foundational steps is to survey protected lands to fully describe the current biodiversity and monitor future changes. To enhance the preservation efforts on the Tater Hill Plant Conservation Preserve, many surveys of the preserve's flora and fauna have been conducted. This project serves to survey the preserve for above ground fleshy fungi (Basidiomycota and Ascomycota), to document both rare and common taxa for the first time. The preserve was surveyed 14 times between 5/4/2022 and 10/18/2022 by walking the entire area using the existing logging roads as well as contour lines. This amounted to ~70 hours of active search time. The main diversity centers were at the low elevations near a creek and mountain seeps as they traveled up in elevation. The high elevation grassy bald and rock outcrops pose a more extreme environment and less fungi were observed in these areas. Roughly 150 above ground fungal specimens were collected, some of which represent

rare species, prized medicinals as well as sought after edibles. These specimens were photographed and their locations were recorded. The majority of these samples are vouchered into the Appalachian State Herbarium to further document biodiversity on the Tater Hill Plant Conservation preserve. This baseline examination of the fungal community should enhance future research directions. Now fungal diversity can be continually monitored and studied alongside other conservation efforts. Tater Hill hosts a diverse and unique fungal community that should remain in conservation.

P123 - Assessing the role of flavonols in driving stomatal aperture in response to diffuse light in tomatoes

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Improving agricultural productivity requires continually exploring how plant function is affected by its ever-changing environment. Recent work has found that the angular quality of light (diffuse) can alter the stomatal opening and rate of photosynthesis. But the leaf-level mechanism for this remains unexplained. Guard cell aperture is mediated by flavonols which reduce reactive oxygen species (ROS), driving changes in guard cell turgor. In this study we sought to understand how light environments, both direct and diffuse, alter flavonol production and ROS accumulation leading to changes in stomatal aperture and photosynthetic rates. To assess flavonol response to light environment, we used genetically altered lines of tomatoes where flavonols are either over-expressed (*oe3*) or under-expressed (*are*) relative to a wild-type control (VF36). Each plant was exposed leaves to 4 hours of a specific light condition (direct or diffuse) to quantify photosynthesis and stomatal conductance using the LI-6800 Portable Photosynthesis Machine. Leaves were immediately excised and stained with DPBA (3 hours) to determine flavonol accumulation. To quantify ROS species, an epidermal peel was stained with DCF to detect a variety of ROS species. Both methods were imaged using confocal fluorescence microscopy. We find that flavonol production affects stomatal aperture under diffuse light. Stomatal conductance and photosynthesis were increased in *oe3* tomatoes most prominently. We further find that *oe3* has increased flavonols and decreased ROS in response to diffuse light across the 3 mutants. This work reinforces that flavonols drive stomatal aperture and are responsive to novel light environments like the angular quality of light. Our future work will further quantify how diffuse light can be used to improve carbon gain and tomato production under novel future climate scenarios.

P124 - The Examination of a Fairy Ring: Cultural, Biological, and Ecological Significance

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Mushrooms have the capability of growing in abundant, circular formations emerging from the same mycelium network, commonly known as "fairy rings". In various cultures, folklore surrounding fairy rings have attempted to teach lessons about the potential harms of ingesting poisonous mushrooms. From an ecological perspective, fairy rings are vital in energy conservation, maintenance of biodiversity in grasslands, and the cycling of nutrients into the soil. The purpose of this experiment was to examine a "fairy ring" and describe its morphological and molecular identity. On two separate occasions, wild fruiting bodies of two generations of mushrooms were isolated in Tuskegee, Alabama. Morpho-identification using field guides, staining and light microscopy techniques were implemented. To confirm identity, polymerase chain reaction (PCR) amplifying the internal transcribed spacer (ITS) region was performed. Following PCR, the sequence was blasted in GenBank database, and phylogenetic analysis using Molecular Evolutionary Genetics Analysis (MEGA 11) software was performed. The results of BLAST and phylogenetic analyses revealed that the fairy ring DNA showed 100% similarity to *Chlorophyllum molybdites* (KU049673) and 51% bootstrap support in the phylogenetic tree. The samples were preserved and sequence deposited into GenBank for future studies. *C. molybdites* is also known as False Parasol, a poisonous basidiomycete fungus. The ability to distinguish edible versus poisonous mushrooms is key to human survival. Understanding the historical/cultural context of the fairy ring along with biological analysis may explain the cultural diversity of superstitions.

P125 - Physiological responses of striped bass (*Morone saxatilis*) to natural temperature variations

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The response of striped bass to seasonal changes in water temperature has been well documented for populations north of the Carolinas, showing fish participating in seasonal coastal migrations following preferred temperature ranges. However, populations in the Southeastern United States are non-migratory, with fish remaining in rivers and estuaries that provide limited options for thermal refuge.

Over the last decade, striped bass in Georgia's rivers have shown decreased reproductive success and are no longer self-sustaining. My study seeks to describe the effects of natural cycles in temperature on the physiological performance of southern striped bass to examine if exposure to temperatures above the thermal optimum is a plausible cause for the population decline. Thirty-six hatchery-reared striped bass were acclimated to one of six thermal treatments for two weeks that reflected the natural temperatures in the Savannah river over early (20°C), mid (25°C), and late (30°C) summer. Fish were acclimated to a stable temperature or a matched thermally variable environment (stable temp  $\pm$  2°C daily). Intermittent flow respirometry measured resting and maximal oxygen consumption rates at each acclimation temperature. These measurements were used to calculate aerobic metabolic scope (AMS) as a predictor of physiological performance. Analysis of data from the experiment is currently underway. The information gained from studying the effects of thermally variable environments on southern populations of striped bass will help inform management agencies on possible causes of the decreased reproductive success in southern populations of striped bass. It will also provide insight into how striped bass may acclimate or adapt to an increasingly warming environment due to global climate change. P126 - Usability of Biodiversity Databases to Answer Questions About Plant Hybridization

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With rates of hybridization increasing due to ecological disturbance, it is essential for plant scientists to understand the potential outcomes of hybridization at all spatial scales. Mapping the distribution of hybrid plants could provide a multifaceted means of studying the impacts of hybridization. Occurrence data available through floras, digitized herbaria, and other sources are a starting point for mapping hybrid distributions, but many issues, such as hybrid identification difficulties, bias in the field, and incompleteness of digitized herbarium records, can leave holes in reconstructed distributions. We compared occurrence data to assess the usability of several commonly used biodiversity databases: GBIF, iDigBio, SERNEC, vPlants, and BONAP. Occurrence data by county were extracted from each database for Georgia, North Carolina, South Carolina, and Virginia for three hybrids (Fabaceae), chosen for their distinct nativity and commonality: *Lespedeza x nuttallii*, *Baptisia x serenae*, and *Wisteria x formosa*. Presence was marked for either a county occurrence point (BONAP) or a representative herbarium sheet (GBIF, iDigBio, SERNEC, vPlants). Cronbach's Alpha consistency tests were used to test for consistency of occurrence data between the databases. Tests show that the databases have varying levels of consistency; consistency is high between the herbarium-based databases, while BONAP has little to no consistency with the herbarium-based databases both individually and combined. GBIF, while showing acceptable consistency with the other herbarium-based databases, has notably lower numbers of occurrence points for all three hybrid species. BONAP, while reporting the highest number of occurrence points, is based on a vastly different array of data references. Overall, the results suggest that there are discrepancies between databases that warrant investigation before use in hybrid distribution mapping. Considerations should include how databases handle various names for a single hybrid, what sources of data are being used, and the number of herbaria from which the vouchers are included.

**P127 - A Chernobyl Field Camera Trap Application: The Influence of Ambient Settings on Deep Learning Effectiveness**

Rachel Maile<sup>1</sup>, Matthew Duggan<sup>2</sup>, Timothy Mousseau<sup>3</sup>

detect 16 different object classes (14 animal species, humans, and fires) across 9,576 images taken from camera traps placed in the Chernobyl Exclusion Zone. After analyzing wind speed, cloud cover, temperature, and image contrast, there was a significant positive association between CNN success and temperature. Furthermore, we found that the model was more successful when images were taken during the day as well as when precipitation was not present. We show that external variables at camera trap locations have a noticeable effect on CNN accuracy. Qualitative sitespecific factors can confuse quantitative classification algorithms such as CNNs. This study suggests that further exploration into the causes of error in classification modeling is necessary given the unique challenges posed by the analysis of camera trap imagery.

**P128 - Preliminary Vascular Plant Flora of the Redstone Arsenal, Madison County, Alabama.**

Curtis Hansen<sup>2,2</sup>, Alfred Schotz<sup>1,3</sup>, Leslie Goertzen<sup>1,2</sup>

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Camera traps have become in-situ sensors for collecting information on animal abundance and occupancy estimates. When deployed over a large landscape, camera traps have become ideal for measuring the health of ecosystems, particularly in unstable habitats where it can be dangerous or even impossible to observe using conventional methods. However, manual processing of imagery is extremely time and labor intensive. Because of the associated expense, many studies have started to employ machine learning tools, such as convolutional neural networks (CNNs). One drawback is that for the majority of networks a large number of images (millions) are needed to devise an effective identification or classification model. This study examines specific factors pertinent to camera trap placement in the field that may influence the accuracy metrics of a deep learning model that has been trained with a small set of images. False negatives and false positives may occur due to a variety of environmental factors that make it difficult for even a human observer to classify, including local weather patterns and daylight. We transfer-trained a CNN to

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American Ginseng (AG, *Panax quinquefolius*, L.) is one of the highest unit value cash crops and one of the most well-known medicinal plants originating from North America. After 300 years of foraging, wild populations have become scarce. Nearly all ginseng is now commercially cultivated. One of the greatest challenges faced by ginseng producers is control of fungal pathogens such as blight, damping off, root rotting, and anthracnose. The goal of this project is to survey the bacterial soil microbiome of healthy versus diseased ginseng to search for differences. Our hope is to identify soil bacteria with anti-fungal properties associated with healthy plants that could be developed into an organic fungicide. Soil associated with healthy and diseased plants were collected from commercial farms in Garrett County, Maryland and Marathon County, Wisconsin. Environmental DNA metabarcoding was used to compare the microbiomes of healthy versus diseased plants from both locations. Beta diversity comparisons identified statistically significant differences between the states. Analysis of Maryland samples identified statistically significant differences between soil samples taken from healthy and diseased plants. *Mycoplasma* sp. was the predominant bacterium in soil associated with healthy plants from Maryland. In diseased plants, an unknown bacteria predominated. In samples from Wisconsin, there was no statistically significant difference between healthy and diseased samples. The most common bacteria in soils associated with both healthy and diseased plants was unidentifiable. However, a bacterium in the genus *Rheinheimera* was exclusively associated with healthy plants. In Wisconsin, soil samples were collected from two cultivation methods - field and woods. There was a statistically significant difference in their microbiomes.

**P131 - Survey of Anisopteran Species of the Rocky River Nature Park**

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A survey of vascular plants from Redstone Arsenal, Madison County, Alabama, is presented. Owned by the U.S. Department of Defense, Redstone Arsenal became the center for missile and rocket research after World War II. Redstone Arsenal is over 15,000 hectares and located southwest and adjacent to Huntsville, Alabama. It is situated in the eastern Highland Rim physiographic province, part of the Interior Low Plateau and adjacent to the Cumberland Plateau. Redstone Arsenal maintains areas of high impact including roadways, industrial buildings and missile testing ranges. There are also nine ecologically sensitive areas that are largely restricted from heavy development and human impact including, natural springs, ponds and streams, rocky-sloped mountains, deciduous forests and limestone bluffs above the Tennessee River. Results of surveys across these diverse habitats have resulted in a rich vascular flora which included 586 species in 355 genera from 122 families. Seventeen Alabama Natural Heritage-listed species were documented from this survey including, *Agastache nepetoides*, *Aplos priceana*, *Astragalus canadensis*, *Carex decomposita*, *Cotinus obovatus*, *Desmodium ochroleucum*, *Elymus churchii*, *Eriogonum harperi*, *Geum virginianum*, *Hottonia inflata*, *Leavenworthia uniflora*, *Muhlenbergia sobolifera*, *Penstemon kralii*, *Perideridia americana*, *Schoenolirion croceum*, *Silphium brachiatum* and *Trillium pusillum*. *Aplos priceana* is also a federally threatened species.

P129 - Mortality of Eastern Bluebirds following a severe winter storm: Did they starve to death?

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Eastern Bluebirds (*Sialia sialis*) often roost together during the winter months, possibly to conserve heat. During severe winter storms, food resources can be covered with snow making them inaccessible causing the birds to have trouble finding the energy they need to live through harsh conditions. After these events, it is not uncommon for bluebirds to be found dead in nest boxes. If the bluebirds starved because they were unable to find food, we would expect their stomachs to be empty when they died. Ninety Eastern Bluebirds were found dead in nesting boxes following a severe winter storm in 2021. Our goal was to dissect the birds and analyze their stomach contents. We gathered information about the age, sex, weight, and general condition of each bird, then we removed the proventriculus and ventriculus of each bird. The dead bluebirds, when ranked, were found to be in fair to poor condition. A healthy bluebird weighs about 30 g, by comparison, the dead bluebirds on average weighed 21.7 g. This suggests that they were having trouble gathering food and maintaining a healthy weight. In the year following the severe winter storm, breeding success was approximately 70% lower than previous years. Eastern Bluebird monitoring programs would benefit from checking nest boxes following severe winter events to gain a better understanding of how these events impact the overall population dynamics of Eastern Bluebirds.

P130 - Soil Microbiomes of Healthy vs Blighted Cultivated Ginseng (*Panax quinquefolius*) Roots

Isabella Maggard<sup>1</sup>, Kayla Deel<sup>1</sup>, Ying Gao<sup>2</sup>, Bruce Cahoon<sup>1</sup>

Micah McKnight, Rocky Nation

*Anderson University, Anderson, SC*

Dragonflies are an important bioindicator species for aquatic habitats (Silva, 2010). Sampling them is standardized, however, knowing the opportune time for sampling is important to ensure accurate sampling results (Santos and Fernandes, 2021). An understanding of the daily fluctuations in dragonfly numbers would help to ensure such accuracy. In this study we sampled the species of a man-made lake at different times to see when they would be most active. Different species showed different preferences for both temperature and time of day. Further studies are required and a larger data pool for conclusive results.

P132 - MIPS-3 Gene Expression and CoSuppression in *Arabidopsis thaliana*

Maureen Besneatte-Cullinane

*Emory & Henry College, Emory, VA*

This study focused on *l-myo-inositol 1-phosphate synthase 3* (*MIPS-3*), and its effects on root length in genetic mutants versus the genetic wildtype in the model plant *Arabidopsis thaliana*. In this study, I focused mainly on phenotypic analysis assays of the root length of *MIPS-3* genetic mutant and wild-type plant strains by plating seeds from 2 different mutant seed strains and the wild-type strain directly onto two MS media conditions - half with added sucrose and half without - and allowing them to grow on the plates for in two different temperature conditions - 19degrees C and 24-degrees C - for 14 days before harvesting the seedlings to use for future qPCR analysis. From these plants, I photographed the differences in the root length at 10 and 14 days, measured the data using ImageJ software to gain an accurate average for the root length over this period, and then analyzed the data to calculate accurate error bars in order to better understand the differences between each strain, and between the different environmental conditions. From this research, I concluded that there were statistical differences in the root length of my *MIPS-3* mutant stains when compared to WT plants when the seedlings were grown in all conditions, with the exception of the seedlings grown in 24-degree C temperature conditions without sucrose. For future work on this particular subject and with these plants (and specific genetic strains), I will be performing a qPCR test on tissues collected from the plants on day 14 during their seedling stage.

P133 - Endocast of *Hyaenodon raineyi*: Implications for Neuroanatomy in Hyaenodonta

Elyse Zeffiro, Heather Ahrens

*High Point University, High Point, NC*

Cranial endocasts are a three-dimensional representation of the endocranial cavity and allow for the visualization of internal anatomical features within the skull. Early studies of endocranial variation were based on naturally preserved stone endocasts found within fossil crania. The increased accessibility of high-resolution computed tomography (CT) scanning of fossils has allowed for more widespread modeling of digital endocasts and assessment of evolutionary patterns within the mammalian brain. We used Avizo Lite 9.4 to create a digital endocast of *Hyaenodon raineyi*, a small-bodied mammalian predator from the late Eocene (36.5 – 33.7 mya). We estimated the body mass as 4.93 kg, utilizing an osteological correlate of the first lower molar. Description of the endocast provides insight regarding brain morphology, including the extent of neocorticalization, or expansion of the telencephalon. Due to damage of the fossil, both the rostral and caudal portions of the endocast could not be reconstructed. However, some inferences could still be made regarding the arrangement of neocortical sulci and several cranial nerves. Three neocortical sulci were identified in *H. raineyi*, including the sulcus marginalis, sulcus suprasylvius, and sulcus ectosylvius. The rhinal sulcus, which delineates the paleocortex from the neocortex, was positioned relatively ventrally. The position of the rhinal sulcus, in conjunction with the three neocortical sulci, suggests that *H. raineyi* had a relatively expanded neocortex similar to the hyaenodontans *H. crucians* and *H. horridus*. The piriform lobe was clearly identifiable and indicates reliance on the olfactory system in *H. raineyi*. The cranial endocast also allows for visualization of cranial foramina and suggests that the alisphenoid canal was absent in the taxon. The identification of these anatomical structures provides a comparison, and visible variation of sulci, across known North American species of the genus *Hyaenodon*.

P134 - Diffuse light increases photosynthesis and stomatal conductance in deciduous ecosystems

John Grosscup, Carter Berry

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Photosynthesis is one of Earth's vital processes that moves massive amounts of carbon. This process relies on basic components of the environment, one of them being light. But not all light is created equal; recent work has demonstrated that diffuse light can drive changes in leaf photosynthesis, but the mechanisms are still unknown. Diffuse light is a common occurrence particularly in the presence of clouds, high water vapor, or atmospheric pollution. We ask two questions: (1) how does photosynthetic rates vary under direct and diffuse light and (2) what are the structural or physiological mechanisms driving these changes? We measured the effect of diffuse light on photosynthesis in 7 deciduous species using a portable photosynthesis system and a 3D-printed light diffusion tool. We paired these data with microscopic measures of leaf structure and biochemistry. We find a wide array of strategies for handling diffuse light. *Liriodendron tulipifera* and *Acer rubrum* significantly increased photosynthesis under diffuse light while *Magnolia grandiflora* had very little change. Notably, all species saw a shift in productivity when exposed to different diffuse light environments. These patterns largely seem to be driven by stomatal conductance and variation in leaf structure. The results require that we reevaluate our understanding of local and global photosynthetic productivity regarding diffuse light environments.

P135 - Continued efforts to develop microsatellite markers within the genus *Dicentra*

Zackary Childers, Matt C. Estep

*Appalachian State University, Boone, NC*

The genus *Dicentra* (Fumariaceae, Bernhardi 1833) is a small group of herbaceous plants, with several species located in the Southern Appalachians. Three species; *D. eximia*, *D. cucullaria*, and *D. canadensis* formed through a polyploid series, where *D. eximia* is a diploid (2N=16), *D. cucullaria* is a tetraploid (2N=32) and *D. canadensis* is an octoploid (2N=64). The purpose of this project is to establish a set of polymorphic microsatellite markers that can be used to investigate genetic diversity, population structure, and the possibility of recurrent polyploidy. This work builds on past attempts to identify polymorphic markers.

P136 - Analyzing the Potential Spread of Invasive Sugarcane Throughout the United States

India Hughes<sup>1</sup>, Steven Hughes<sup>2</sup>, Rima Lucardi<sup>3</sup>, Travis Marsico<sup>4</sup>, Emily Bellis<sup>5</sup>

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*Saccharum spontaneum* (wild sugarcane) is a regulated Federal Noxious Weed (FNW) that has been shown to alter native plant communities postintroduction. *Saccharum spontaneum* is rhizomatous, grows in thick stands, can choke out native species, and has been found to be a fire-promoting species, impacting restoration and reforestation efforts. We ask if there is significant risk of invasion in the U.S. We have conducted genomic comparisons of existing populations of naturalized sugarcane in Florida, the only U.S. state where colonies of *S. spontaneum* are documented. We took tissue samples from these stands and extracted and sequenced full genomic DNA. Using an alignment-free bioinformatic approach to analyze the sequences, genetic analyses for three populations were consistent with expectations for hybrids of *S. spontaneum* and *S. officinarum* (domesticated sugarcane). In contrast, out of 12 sampled stands, 9 were genetically identified as *S. spontaneum*. In addition to naturalizing wild sugarcane stands in Florida, we have discovered other potential U.S. invasion routes. The research team collected reproductively viable propagules attached to refrigerated shipping containers that passed through the Panama Canal, where *S. spontaneum* is widespread. These

samples clustered more closely with *S. spontaneum* than with individuals of hybrid origin. We also procured samples of “dried naturals” containing imported grass infructescences, including grasses morphologically indistinguishable from wild sugarcane, at nationwide craft stores, a potential invasion path of high risk. It is our goal in this ongoing project to test the viability of these propagules through several germination and growth experiments under differing environmental conditions, utilizing a secure greenhouse.

P137 - Remnants of the American red wolf: A spatial and genomic analysis of the Arkansas coyote population

Zachary Campbell

*Arkansas State University, Jonesboro, AR*

The American red wolf (*Canis rufus*) is a North American endemic species that once represented a successful reintroduction story following its 1980 extinction in the wild. However, this species continues to face conservation challenges, including anthropogenic forces and hybridization with coyotes (*Canis latrans*). As the American red wolf faced extirpation, the coyote experienced an opportunistic range expansion throughout the southeast, and hybridization events increased. Recent evidence indicates that some wild *Canis* species located in the historical American red wolf range harbor ancestral genetic elements no longer found in the contemporary population. As targeted genetic rescue techniques continue to advance, remnant genetic elements could prove integral for the future conservation of this critically endangered species. To date, there has been no comprehensive genomic study on coyotes in Arkansas—a state once fully nested within the historical American red wolf range. By sampling coyotes at a statewide scale, we aim to determine (1) the population genetic structure of coyotes throughout Arkansas and (2) whether Arkansas coyotes are reservoirs of American red wolf genetic elements that have been lost in the breeding population. We are also working with collaborators to observe spatial genetic variation in coyotes across the contiguous United States. Hybridization events are expected to scale with reintroduction efforts, making it increasingly important to improve methods of differentiation between wolves, coyotes, and hybrids. This report presents preliminary data on genetic sampling and morphometric analyses in our efforts to disentangle genotypic and phenotypic variation of American red wolves and coyotes and inform future recovery efforts.

P138 - Characterizing plant biochemical responses to pathogenic stress: spotlight on red leaf spots

Anna Ferraro, Maggie Salley, Nicole Hughes *High*

*Point University, High Point, NC*

Red leaf spots commonly accompany pathogen infection in plants. Yet, the function of red anthocyanins pigments in infected tissues is unknown. We characterized biochemical responses of two plant species, red-tip photinia (*Photinia glabra*) and Indian hawthorn (*Rhaphiolepis indica*), to infection by the pathogenic fungus, *Entomosporium mespili*. We used analytical HPLC and LC-MS to identify and quantify anthocyanins and photopigments in red leaves or tissues (including red expanding leaves, red senescing leaves, and red spots) and green tissues (green areas around red spots, fully-expanded green leaves) of the same plants. Maximum quantum yield efficiency of PSII ( $Fv/Fm$ ) and xanthophyll cycle pigment ratios was measured as a proxy for sustained high-light stress. DPPH radical scavenging assay assessed low molecular weight activity (LMWA) in red spots versus surrounding green tissues. The major anthocyanins in red tissues of both species were cyanidin-3-*O*-galactoside and cyanidin-3-*O*-arabinoside. Red tissues of photinia also contained small amounts of three additional cyanidin mono-glycosides (cyanidin-3-*O*-glucoside and two unidentified cyanidin-*O*-pentosides); red spots lacked cyanidin-3-*O*-arabinoside. All red tissues had significantly lower chlorophyll and  $Fv/Fm$  than green tissues, suggesting increased vulnerability to photo-oxidative stress. Photoprotective carotenoids did not differ between red and green tissues, likely due to small sample size. Antioxidant activity was 1.5-3.4x higher in red spots versus surrounding green tissues; follow up assays with purified anthocyanins suggested anthocyanins were responsible for ca. 1/3 antioxidant activity in red spots. From these results, we predict the function of anthocyanins in red spots is similar to anthocyanins in young and senescing leaves, and this function likely includes photoprotection (i.e. light attenuation and antioxidant defense). We are currently investigating an anti-microbial function by culturing *Entomosporium mespili* in water agar plates treated with cyanidin-3-*O*-galactoside. Optimal conditions are being determined to avoid anthocyanin degradation. P140 - Sexual dimorphism in the musculoskeletal system of domestic cats

Katharine Spence, Latavia Foster, Jeremy Morris

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Across many animal species, mating success is influenced by competition, territory holding and resource access. Intrasexual selection is a form of Sexual Selection in which males define traits passed on through same sex male competition that would increase reproductive success (R.R. Lawler, et al. 2005). Domestic cats are one such species that are polygamous and rely on intense male to male competition for the best access to mates and territory. Sexual dimorphism is widespread in polygamous species where one sex of the same species is larger than the other. Sexual dimorphism is passed on to the strongest competitor allowing for natural selection and evolution of certain traits. We tested the hypothesis that because fighting involves grappling with the forelimbs and accelerating toward an opponent with the hindlimbs, males in contrast to females are predicted to have adaptations in the musculoskeletal system that improve their ability to carry out these physical tasks when fighting. Because the forelimbs are used to a greater degree when fighting, we predict that the degree of sexual dimorphism in the forelimbs will be greater than in hindlimbs. We dissected and removed 5 major muscle groups in house cat limbs (*Felis catus*) actively used in fighting: biceps brachii, triceps brachii, wrist extensors, wrist flexors, gastrocnemius, and a control of the Tibialis Anterior. By measuring skeletal and vertebral length to muscle mass we found that relative to body length males have larger muscles in forelimbs compared to skeletal size. The specific purpose for collecting this data is to determine if there were differences in muscle mass and skeletal shape in house cats.

P141 - Characterization of Isolates for Development of Canavanine-based Gross Chromosomal Rearrangement Assay in *S. pombe*

Canavanine resistance provides pathways for the development of an assay that works in *Schizosaccharomyces pombe* to isolate cells with a gross chromosomal rearrangement mutation so that the causes and effects of these mutations may be studied in a laboratory context. The assay depends on the deletion of a cassette containing *ade1* and arginine permease genes *cat1* and *aat1*, which allows cells to be isolated as GCRs if they grow pink on minimal media with canavanine and low amounts of adenine present. After the development of this assay, its efficacy has been tested by carrying out a fluctuation test with 4 variations of 16 strains. The variations are the placement of the cassette, which is either on chromosome 1 or 2, and the presence of an inverted repeat, which is either present along with the cassette or not. The fluctuation test indicated what the rate of gross chromosomal rearrangements was in each strain and variation, and results were further and explored via PCR. Importantly, regardless of which chromosome the cassette was placed on, the presence of an inverted repeat was found to increase the rate of gross chromosomal rearrangements significantly, demonstrating that the assay successfully indicates when mutagenic or unstable elements are present. The most common extent of mutation verified by PCR was between 97 and 120 kilobase pairs from the telomere on the chromosome. Demonstrating efficacy of this low-cost assay can bolster scientific research due to its accessibility and ease of use, and it can provide information about the mechanisms of human genetic diseases and cancer.

## P142 - The impact of mental health and mental stress on cardiovascular health in a diverse population.

Zoe Snider, Jennell Talley

*Georgia Gwinnett College, Lawrenceville, GA*

Cardiovascular disease (CVD) is the leading cause of death in the United States. Recent studies have suggested that poor mental health and increased mental stress may contribute to hypertension, which is related to cardiovascular and coronary artery disease (CAD). Too often studies related to human health rely on data collected from a narrow group of people that does not accurately represent the diversity of our population. In 2015, the National Institutes of Health worked with many scientists to start the *All of Us* project, which aims to recruit 1 million study participants from diverse backgrounds. We used information from demographic surveys, electronic health records, and other survey data from the *All of Us* database to determine if people who report feeling depressed, anxious, or stressed OR those who have been clinically diagnosed with depressive or anxiety disorders are more likely to also have CVD, CAD, or hypertension.

P143 - Evaluating genetic intermediacy of a naturally occurring hybrid blueberry, *Vaccinium x Marianum*

Laura George, Lisa Wallace, Devani Jolman

*Old Dominion University, Norfolk, VA*

Hybridization is common in plants and is a mechanism for increasing plant biodiversity and vigor. Detecting hybrids in nature can be accomplished by genetic data. The basis of this research is to evaluate if the highbush blueberry, *Vaccinium x Marianum*, is a hybrid between the parental species, *Vaccinium fuscum* and *Vaccinium formosum*. Morphological data suggests *V. x Marianum* has traits intermediate to the parental species, and this research project used genetic data to evaluate differences between the parents that are expected in *V. x Marianum* if it is a hybrid. Leaf samples of the three forms were collected from four locations in Virginia. They were identified in the field using a diagnostic key. Fourteen microsatellite loci were selected to test with the samples. Data from seven loci were evaluated for this study, although one locus was not found to be consistently scorable and thus was removed prior to analysis. Our preliminary analysis suggests that *Vaccinium x Marianum* is genetically intermediate to the parental species. The parental species are genetically distinguishable from one another and little evidence of introgression was found in these samples. Samples of *V. x Marianum* show extensive variation among samples, suggesting that it may represent multiple hybrid generations and diverse origins. More work is needed to examine the genetic variation of the three forms, and this semester I will use chloroplast markers to evaluate the maternal origin(s) of *V. x Marianum*. Hybridization between blueberry species is common in nature and studying hybrids can further the understanding of genetic diversity, adaptation, and evolution of blueberries species.

P144 - Cranial variation within and between *Canis rufus* and *Canis latrans* in North Carolina

Alexandria Ritz, Hannah Smith, Rebecca Fasanella, Heather Ahrens

*High Point University, High Point, NC*

*Canis rufus* is an endangered North American canid that has experienced population decline since the early 1900s. A wild population of red wolves has been reestablished in eastern North Carolina where they coexist and hybridize with the coyote, *Canis latrans*. Thus, our objective was to compare the cranial anatomy of *Canis rufus* and *Canis latrans* from North Carolina and examine morphological variation within and between these two sympatric species. Our sample included 45 specimens housed at the North Carolina Museum of Natural Sciences collected from both eastern and western North Carolina. To evaluate cranial variation, we conducted principal components analyses (PCA) using two-dimensional geometric morphometrics based on 35 landmarks in both lateral and ventral views. In lateral view, the two *Canis* species overlap in PCA morphospace. Principal component one was associated with variation in the length and position of the occipital crest and position of the suture between the jugal and squamosal bones. Principal component two was associated with variation in the height of the occipital crest and position of the parietal suture. In ventral view, *Canis rufus* and *Canis latrans* occupy distinct areas of PCA morphospace. Principal component one was associated with changes in the position of midline sutures of the sphenoid complex, as well as of the posterior dentition. Principal component two was also associated with changes in the position of the midline sutures of the sphenoid complex, as well as the width of the zygomatic arch. In this preliminary analysis,

sutures of the basicranium and width of the zygomatic arch provide the clearest anatomical delimitation of *Canis rufus* and *Canis latrans*. P145 - Reverse Genetic Analysis of Karrikin-Induced Seed Germination in *Arabidopsis*

Rose Catherine Livinus, John Stanga

*Mercer University, Macon, GA*

Germination is the process by which seeds begin to grow, entering the early development of roots, shoots, and leaves. Karrikins (KARs) are exogenous molecules derived from smoke that promote seed germination. Strigolactones (SLs) are small-molecule plant hormones that stimulate germination in several species of parasitic plants, such as *Striga hermonthica*, as well as other non-parasitic plants, like *Arabidopsis thaliana*. Both KARs and SLs activate germination through a shared molecular mechanism that can bypass gibberellin acid (GA), which is the dominant hormone for germination stimulation. While it is known that SLs promote parasitic seed germination through the KARRIKIN INSENSITIVE 2 (KAI2) pathway, the mechanism by which SLs bypass GA signaling is not fully understood. A 2020 study identified ninety-eight *S. hermonthica* and *A. thaliana* genes that are responsive to KARs during the first 24 hours of germination (Bunsick et al., 2020); while other studies have detected transcriptional responses within the first 8 hours. Among these, we hypothesize that genes with GO terms relating to regulation of seed germination, hormone signaling, and other signal transduction functions are more likely to function in early seed germination than genes with GO terms relating to photosynthesis, which represent thirty-eight percent of the ninety-eight genes. Transcriptional analysis in early germination of *Arabidopsis thaliana* seeds is underway using qPCR to identify expression patterns that differ among karrikin signaling mutants during the early stages of the first 24 hours of germination. Genes that express the earliest transcriptional responses are candidates for CRISPR mutagenesis. Induced mutants that can no longer germinate through the KAI2 pathway but are still capable of germinating through the GA signaling pathway would represent karrikin signaling molecules.

P146 - Ethanol and nicotine cross state dependent learning in *Caenorhabditis elegans*.

Jonathan Lindsay

*Emory & Henry College, Emory, VA*

State dependent learning occurs when expression of a learned behavior is less effective due to shifting from one internal state to another during learning and recall. Though state dependency can occur in a variety of organisms, including zebrafish, rodents, humans, and *Caenorhabditis elegans* (*C. elegans*), very little is known about the molecular mechanism of the behavior. *C. elegans* make great models for investigations in animal behavior: they are capable learners, mutant strains are readily available, and worms have a simple nervous system. To demonstrate state dependent learning in *C. elegans*, N2 lab strain worms are tested for olfactory learning (OL), which is the diminished response to an odor after subsequent exposure to the same odor in the absence of food. State dependency is observed when worms exposed to ethanol during learning and recall show normal OL, while worms exposed to ethanol during learning, but deprived of ethanol during recall, show impaired olfactory learning. Previously, we identified genes required for associative learning tasks, *hen-1* and *scd-2*, are also required for state dependent learning. Furthermore, we observed that an alternate circuit consisting of neurons novel to the olfactory learning mechanism are required for state dependency. Here, we investigate cross state-dependent learning, learning while exposed to ethanol and testing while exposed to nicotine in *C. elegans*. We will assay for cross state dependency by exposing N2 worms to 150 mM ethanol during learning and observe behavior following two separate conditions during chemotaxis - 50  $\mu$ M nicotine or no treatment. Impairment of olfactory learning in the absence of nicotine indicates state dependency. We hypothesize crossstate dependency will require the *hen-1* and *scd-2* genes. Our goal is to test the similarities and differences between state dependency and crossstate dependency to further our understanding of the state dependent learning mechanism.

P147 - Unlocking the Power of Genomics Statistical Modeling for Insights in the Relationship Between Trait Genetic Architecture and the Allelic Frequency Distribution

Carina Bravo-Chan

*University of North Carolina Greensboro, Greensboro, NC*

Adaptation in allele-variant organisms tells a story about how they change and adapt to their environment. The genetic architecture of adaptation is a critical determinant of the evolutionary rate and phenotypic outcomes under different allelic starting points. The higher the initial allelic variance, the faster the evolutionary rate, suggesting that more diverse populations are more likely to start with traits that help them adapt than populations with less allelic variance. Understanding this relationship is invaluable in predicting evolutionary trajectories and how future populations might adapt. In this study, we made two simulations to examine how traits change over time and how these changes affect the molecular basis for trait divergence. We focused on a few aspects of adaptation: how quickly new large-effect loci appeared from the existing variation in a population, how long these new alleles remained, and at what frequency. To examine that, in our first simulation, the mutation variance rate was lowered to a quarter of the standard observed rate. This was done to see how quickly large-effect loci reached a high frequency in a population. The second step looked at how much the final distribution of alleles depended on the presence of large-effect loci and whether they persisted in the population. To test the impact of large-effect loci, a second simulation was done with a selection variance rate four times higher than the usual rate. We used RStudio and Nemo, a mathematical modeling environment, to calculate how the number of alleles changes over time. We expect to find a pattern between the number of genes involved in trait variation and how much the initial molecular architecture affects how alleles are distributed in a population. Understanding the genetic architecture of a trait can provide insights into the evolution of a trait and inform genetic manipulation. P148 - Methods for Saliva Collection in Canines

Matthew Hegler, Joni Criswell

*Anderson University, Anderson, SC*

One of the most sought after tests in vet medicine is one that can predict the existence or onset of serious illness. Serum cortisol concentration (SCC) is a highly researched and promising technique for prognostic diagnosis (Yuki et al., 2019). However, SCC requires drawing blood. This can be invasive, traumatic, and sometimes impossible depending on the dog's receptiveness to the procedure. Nevertheless, recent research has proposed a healthier alternative. Saliva cortisol levels have shown to be both comparable and more positively received by canines. The only setback to implementing this technique is the difficulty in obtaining enough saliva per sample (Damián et al., 2018). About 1ml of saliva is needed to accurately measure and verify cortisol levels in a saliva sample. This is attainable without anything other than a polyester swab. However, it takes significant work and restraint time for predominantly insufficient results. In this study, an alternative acquisition method using citric acid was proven to be exponentially more effective.

**P149 - Ice, ice, baby: morphological, genomic, and physiological characterization of a novel psychrophilic bacteria *Sphingomonas nivalis* sp. nov isolated from snow**

Amerah Odeh, Shawn Brown

*University of Memphis, Memphis, TN*

Late season temperate snows make up a large amount of the earth's surface and contain many unique and endemic microbial taxa that are poorly studied. To investigate these taxa, a snow culture library of fungi and bacteria was generated from a permanent snowfield near Lyman Glacier, Glacier Peaks Wilderness Area, Wenatchee National Forest, in Washington State. Here, we describe a novel psychrophilic bacterial species backed by genomic resolution that we are putatively naming *Sphingomonas nivalis* sp. nov., a rod-shaped Gram-negative bacteria. Here we present morphological, physiological, and nutrient utilization capabilities using a combination of Scanning Electron Microscopy coupled Energy Dispersive X-Ray Spectroscopy (EDS) and culture based Phenotype MicroArray plates targeting carbon, nitrogen, phosphorus and sulfur utilization metabolic pathways. Further, EDS analyses suggested there was an unusual aggregation of Zinc (Zn) and Silica (Si) associated with cultures of this bacteria possibly suggesting sequestration or utilization that may enhance growth. To investigate if this was occurring, we performed differential growth assays on solid media with different concentrations of Zn and Si, and compared growth with Zn or Si to cultures without added Zn or Si using multiple regression analyses. This suggested Zn inhibits growth while Si is neutral to growth. *S. nivalis*, being a putatively novel species in which investigations are ongoing, by studying this bacteria we can expand our understanding of snow ecosystems. Because snow-inhabiting microbial taxa are poorly studied, this work is crucial to elucidate the evolutionary history and biological functions they hold, and their interactions with the environments.

**P150 - Fighting and Jump Performance in Male Crickets**

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*Wofford College, Spartanburg, SC*

Male crickets engage in intrasexual competition to compete for access to resources such as territory and access to potential mates (Andersson, 1994). Often these competitions lead to sexual selection in males so they are better equipped for fighting. These traits could include things such as greater body mass or better weaponry. These traits could also be important for other activities. Crickets use the same muscles for fighting as they do for jumping. During fights, male crickets attempt to push their opponent over using their hindlimbs as leverage and wrestling around by locking mandibles (Hack, 1997). We hypothesized that males with a greater jump velocity would be better fighters. We also hypothesized that males with morphological traits such as greater body mass and better weaponry would be more likely to win fights as they are better equipped.

**P151 - Creation of a Knockdown Strain Library for *Staphylococcus aureus* via CRISPR interference**

Gennie Bassett<sup>1</sup>, Claudia Prieto Alcaide<sup>1</sup>, Melinda Grosser<sup>2</sup>

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In bacteria, gene knock-down libraries are a useful tool in investigating the roles of essential genes that cannot be investigated via loss-of-function mutation. However, essential gene knock-down libraries are often not readily available for non-model organism bacteria, including *Staphylococcus aureus*, despite being a useful research tool. *S. aureus* is one of the leading bacterial pathogens worldwide. Its infections vary in presentation (e.g., skin and soft tissue infection, endocarditis, osteomyelitis) and can be either community or hospital-acquired. *S. aureus* is difficult to treat due to the emergence of multi-drug resistant strains, such as Methicillin-Resistant *Staphylococcus aureus*, or MRSA. The goal of this project is to use CRISPR interference (CRISPRi) to eventually create a 579-strain library for *S. aureus* that can be used to research the function of essential or nearly essential genes that are not represented in the current commercially-available Nebraska transposon mutant library. CRISPRi is a relatively new technique that has shown success in creating knock-down libraries. It employs a catalytically inactive version of Cas9 (dCas9). When CRISPRi and dCas9 associate with DNA, instead of cleaving the target site, they sterically block transcription by preventing RNA polymerase from interacting with the DNA at that site. This research focuses on optimizing a user-friendly protocol for creation of knock-down strains that can be performed by undergraduate students in an upper-level molecular biology course. To date, proof-of-principle experiments have shown that knock-down strains were successfully created for genes *srrA*, SAUSA300\_1526, and SAUSA300\_0984, where qRT-PCR confirmed a reduction in target gene activity.

Future directions for this project include expanding and improving protocols to scale up the creation of knock-down strains and using these strains in phenotypic and growth assays. Ultimately, use of the resulting strains to study the function of essential genes may help elucidate targets for novel antibiotics.

P152 - The effect of upland pasture on longnose dace (*Rhinichthys cataractae*) time in flowing water

Ethan Moses, Kimberly Bolyard

*Bridgewater College, Bridgewater, VA*

Longnose dace (*Rhinichthys cataractae*) are found widely within the streams and rivers of the Shenandoah Valley and typically inhabit moderate to fast flowing water. They are an important food fish for larger fish like smallmouth bass and trout, and serve as a key link between algae and large fish in aquatic ecosystems. Assessments of waterways across the U.S. show that many are below recommended standards for human use and for aquatic life due to excess nutrients, sedimentation, and chemical pollutants such as pesticides, herbicides, and heavy metals. Agricultural practices often contribute to degraded water quality despite implementation of best management practices (BMP) to mitigate these effects. Polluted water can impact the health and behavior of the fish in it. We examined the sublethal impact of upland pasture use on the behavior of longnose dace from two different streams in the Shenandoah Valley. The streams varied in the percent of upland pasture that drained into them but had similar levels of BMP implementation. We collected dace with a backpack electroshocker and brought them into the laboratory for testing. Submersible pumps generated a one-way water current (0.20-0.75m/s) in half of a test area (30cm x 60cm, 10cm deep) of a larger tank. As expected, we found that dace from the stream draining less upland pasture spent more time in the flowing water than in the still water, but that dace from the stream draining more upland pasture spent equal amounts of time in the flowing and the still water. The fish did not spend different amounts of time swimming. A histological examination of the fish is in progress to better understand what physiological changes may co-occur with the behavioral differences we observed.

P153 - *Escherichia coli* O157:H7 present in Rocky River and Cox Creek of Anderson, SC

Georgiana Walters, Canon Allen, Carrie Koenigstein

*Anderson University, Anderson, SC*

*Escherichia coli* O157:H7 is a pathogenic serotype of the broader *E. coli* species that can cause severe intestinal infection when ingested. Following South Carolina Adopt-A-Stream (SCAAS) protocols, we collected water samples from Rocky River and Cox Creek in Anderson, SC over the course of four weeks. Bacterial samples were isolated using Petrifilms and sorbitol-MacConkey agar to identify potential *E. coli* O157:H7. The isolated bacteria were analyzed using a Western Blot to confirm the presence of the H7 flagella protein, typical of the pathogenic O157 serotype. The level of *E. coli* collected from each water source was within the acceptable range as defined by SCAAS standards (<1000 CFU/100 mL). Additionally, our results indicated a relatively low level of *E. coli* O157:H7 in each water source as compared to the overall *E. coli* levels.

P154 - Bad Egg: Mimicking Brood Parasitism Among Brown Thrashers and Northern Mockingbirds

Emily Gardella<sup>1</sup>, Savannah Steet<sup>2</sup>, Dr. John Quinn<sup>2</sup>

<sup>1</sup>*Furman University, SC, <sup>2</sup>Furman University, Greenville, SC*

Obligate brood parasitism is the reproductive method of laying eggs in other birds' nests to rear as their own. Hosts will reject parasitic eggs to prevent repercussions of foreign fledgling(s) consuming valuable resources. Currently, ornithology research categorizes hosts into "rejector" or "acceptor" species depending on the rate of rejection. The Northern Mockingbird (*Mimus polyglottos*) and Brown Thrasher (*Toxostoma rufum*) are two classified "rejecter" species found in a majority of the United States. As evolution persists, host species such as the Northern Mockingbird and Brown Thrasher have adapted diverse defense mechanisms to prevent parasitism. We aim to replicate brood parasitism during the breeding season to evaluate Northern Mockingbird and Brown Thrashers as "rejector" species and explore the potential mechanisms of rejection and adapted behaviors of defense. We conducted our research by systematically parasitizing Northern Mockingbird and Brown Thrasher nests using mimetic 3D modeled eggs. We recorded the longevity of the fake egg- labeling eggs as rejected if no longer present after parasitization or accepted after five days untouched. Correspondingly, we recorded host behaviors and proximate mechanisms during parasitization. In totality, 36 nests were parasitized during the span of research. Northern Mockingbird parasitization (n = 8) had a rejection rate of 37.5% and Brown Thrasher parasitization (n = 23) had a rejection rate of 82.6%. From observation, defense adaptations increased the likelihood of rejection, however, no physical difference of the mimetic egg played a role. Further research should evaluate Northern Mockingbirds as "rejecter" species, as the low percentage of rejection could suggest an alternate mechanism for rejection than previously assumed. Research such as this reveals the distinct types of adaptations and evolutionary models that parasites and their host counterparts can take.

P155 - Identification of Bacteria in Contaminated Dog Kibble

Logan Eure

*Guilford College, Greensboro, NC*

Proper storage of dog kibble (food) is essential to the health and well-being of all canines that consume it. The aim of this study was to identify bacteria that could possibly contaminate canine kibble while in storage which may lead to serious illness/disease. Five different scenarios were constructed to mimic different kibble storage techniques, each with a distinct environment. Samples were collected and grown in four different media. This was followed by biochemical testing, DNA extraction, PCR, gel electrophoresis, and sequencing of the PCR product to identify the organism/s. The data showed that *Escherichia coli* and *Salmonella* sp were found within the contaminated kibble. This supports the hypothesis that when stored improperly, canine kibble could be contaminated with *Escherichia coli* and *Salmonella* sp which may cause life-threatening illnesses. It is hoped that this research and analysis will encourage dog owners to properly store their animal's food in order to prevent serious illness. Future experiments could entail testing additional pet food storage environments as well as identifying different sources of contamination.

#### P156 - Influence of Urbanization on Southern Appalachian Insectivorous Bats and Arthropods

Noa Meiri<sup>1</sup>, Rachel Maunus<sup>2</sup>, Lindsey Zarecky<sup>3</sup>, Malcolm Schug<sup>4</sup>, Rada Petric<sup>5</sup>

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As urbanization and concomitant pollutants rise in conjunction with the human population, freshwater sources are increasingly becoming compromised by urban development and chemical contamination. Degraded environments reshape the diversity and abundance of arthropod communities, which may in turn influence the composition of insectivorous bat communities. Bat populations are similarly sensitive to changes in urbanization, making it essential to understand how changing land cover and land use influences bat and their arthropod prey community dynamics. In this study, we investigated nocturnal arthropods and insectivorous bats at 32 water bodies along a rural-exurban-urban gradient using light traps and acoustic recorders in the southern Appalachian Mountains of western North Carolina, USA. We found that total bat activity was the highest in areas with a medium disturbance level; however, individual species responded uniquely, and sometimes oppositely, to disturbance. Arthropod biomass was highest in areas of low disturbance, demonstrating that both bat activity and arthropod biomass are affected by disturbance. There was no significant correlation between activity and biomass at varying disturbances. The results of this study highlight the importance of a species-specific approach to bat conservation and points to the need for future research on the intricacies of the relationship between bat activity, arthropods, and anthropogenic disturbance.

#### P157 - Identification of Antibiotic Producing Bacteria in Wastewater Obtained from the Harnett County Wastewater Treatment Plant

Thomas Rush

*Campbell University, Buies Creek, NC*

The emergence of antibiotic-resistant pathogens has become a major public health concern. Antibiotics are an essential, first line component of modern medical treatment but their overuse and misuse can provide the selective pressure necessary for multidrug resistant bacteria to develop. The discovery of novel antibiotic producing bacteria in wastewater could provide additional tools to combat resistant microbes. In this study, a sample of mixed liquor with suspended solids was collected from the Harnett County Wastewater Treatment Plant. Potential antibiotic producing bacteria within the sample were cultured, isolated and identification was attempted through 16s rRNA sequencing and Biolog Gen III microplates™.

#### P158 - Preliminary Assessment of the Function of the Growl Call of Chinese Blue-breasted Quail (*Coturnix chinensis*).

Edward Mills

*Wingate University, Wingate, NC*

The Chinese Blue-breasted Quail (Galliformes, *Coturnix chinensis*) produces several different calls associated with courtship once they become sexually mature. Both males and females give loud crow calls related to territoriality and courtship. The low, raspy growl call, however, is only emitted by reproductively mature males. Several spectral characteristics of the growl call were measured during this study including call length (sec), low and high frequency (Hz), band width (Hz) as well as average and maximum power (dB). This acoustic signal is of medium volume (58.35 dB), has a low frequency (555.0 – 1581.6 Hz) and has an average duration of ~1.5 seconds. No harmonics are formed along with the main call. Given in close proximity to females, the males often exhibit a low wing-droop display along with the call. Some researchers believe the growl to be a separation call, given when a male-female pair are separated. Others hypothesize that this call is part of courtship, given by unmated males attempting to secure mates. In this study, unmated, sexually mature males were recorded producing growl calls and displays in response to the presence of female quail (caged separately, but within view) and when stimulated by the digital playbacks of growl calls. These results suggest that the growl call is given by males as part of the courtship process in an attempt to acquire a mate, but does not rule out the possibility that this vocalization may also be used as a separation call.

#### P159 - Identification of *E. coli* Proteins Required for Shape Transformation and Growth at Low Temperature

Jireh Pinkney, Molly White, Pamela Jones

*Winston-Salem State University, Winston-Salem, NC*

Cellular stress occurs upon exposure of cells to unfavorable growth conditions, such as temperature changes. Physiological changes are induced to counteract the damaging effects of the stress on cellular structures and processes. A physiological change that occurs upon shifting *Escherichia coli* cells from high temperature to low temperature is a transformation in morphology from rods to small rods. The aim of the research study is to understand the adaptive basis for the alteration in cellular shape by identifying cellular factors required to facilitate the small rod morphology and growth at low temperature. In accordance with this goal, we have identified *E. coli* mutants that fail to form small rods at low temperature. In contrast to the normal growth and small rod morphology of the wild-type, incubation of the mutants at low temperature resulted in cold-sensitive growth and the formation of filamentous cells. Furthermore, introduction of a plasmid encoding the wild type protein in each of the mutants resulted in normal growth accompanied with the appearance of small rods. In addition, an extragenic multicopy suppressor of the cold-sensitive phenotypes of one of the mutants has been identified. Therefore, *E. coli* has a set of proteins that facilitates the rod to small rod morphology, which is adaptive for growth at low temperature.

P160 - Effect of prolonged Cannabidiol oil exposure on *Mus musculus* locomotion and instinctual anxiety-related behavior

Emily Evans, Yesica Landaverde, Holly Boettger-Tong, James Rowan

*Wesleyan College, Macon, GA*

Cannabidiol oil (CBD) is an anti-inflammatory, anti-arthritis, and analgesic drug that targets the endocannabinoid system of *Mus musculus* and other mammals. CBD reacts on multiple dopamine-producing receptors and has a low affinity for the CB1R receptor in the endocannabinoid system. Endocannabinoids have been shown to mediate memory formation using the CB1R receptor, where an agonist leads to memory loss or an antagonist reduces the amount of memory loss. In a previous study in our lab, adult male and female mice were treated daily, via oral administration over a six week period, with either CBD or the vehicle, coconut oil, to ascertain the influence of CBD on a single alternation learning and memory task. In the current study, the long-term effects of this prior CBD treatment on mouse behavioral traits were assessed in an open-field testing environment. While most studies have addressed either acute oral cannabidiol effects or long-term effects via intraperitoneal routes of administration, this study addresses the long-term effects of prolonged oral CBD exposure on mouse locomotor and anxiety-related behaviors. As many human uses of CBD are achieved via oral administration, assessment of the long term effects of this pharmacologic route are important in understanding this drug's full therapeutic potential.

P161 - South East Regional Network of Expertise and Collections (SERNEC) Data Portal: Seeking Collections & Data Users!

Carrie Barker<sup>1</sup>, Herrick Brown<sup>2</sup>, Michael Denslow<sup>3</sup>, Jennifer Kluse<sup>1</sup>, Shawn Krosnick<sup>4</sup>, Laura Lagomarsino<sup>1</sup>, Zack Murrell<sup>5</sup>, Katie Pearson<sup>6</sup>, Lindsay Walker<sup>6</sup>, Jenn Yost<sup>6,7</sup>

Public access to digitized vascular plant records accessioned in Southeastern US herbaria is provided through the SERNEC (South East Regional Network of Expertise and Collections) portal, hosted by the Symbiota Support Hub. Herbarium specimen data is entered directly into the portal (live) or as uploads (snapshots) from institutional databases (such as Specify). There is much functionality built into the portal, such as uploading community science transcriptions, image loading, collaborative georeferencing, and data publishing tools to share records from the SERNEC portal to broader data aggregators, such as GBIF (Global Biodiversity Information Facility). The SERNEC portal and its data contributors provide a powerful tool for the scientific community to study natural history collections over 200 years of time at a global scale. The portal and its many data discovery tools (e.g., checklists and dataset creation) have also become invaluable for conservationists and land managers stewarding the highly diverse flora of the Southeastern US region. In 2014, the SERNEC Thematic Collections Network (NSF Award #1410069) launched the SERNEC portal, which has grown to include 233 SE USA herbaria with 123 of these herbaria contributing data to the SERNEC portal, totaling over 5.5 million specimen occurrences with 90% of these records being imaged and 85% identified to species. As a community, we rely on students and volunteers to assist with generating data (see sernecportal.org for how to get involved) and herbaria staff by becoming members of Southeastern United States of America Chapter of the Society of Herbarium Curators (SHC-SEUSA) to network. As users of this platform and members of SHCSEUSA, we are presenting this poster in hopes of providing a point of contact to answer any questions in person.

P162 - Epigenetic consequences of poor parenting can be partially ameliorated by environmental enrichment in a rat model

R. Adam Franssen, Delaney McMahon, Hailee Banks, Josh Bain, Alayna Palamar, Kiersten Witte *Longwood*

*University, Farmville, VA*

As in humans and other species, not all rat mothers are good at motherhood. Based on the extensive literature, "good" maternal behavior can be defined as high levels of licking, grooming, nursing, retrieving, and protecting pups. Conversely, "bad" maternal rats perform these tasks less

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frequently. The epigenetic consequences of good or bad maternal behavior can last generations in terms of memory, anxiety, and resiliency. Our lab has been working to determine if environmental enrichment – adding chew toys, nesting materials, and hiding places to the home cage – can alter behavioral outcomes for rat pups of bad mothers. For this experiment, four conditions were created: 1) good mother + enriched environment, 2) good mother + control environment, 3) bad mother + enriched environment, and 4) bad mother + control environment. We then assessed rats' behavior in four different paradigms: the elevated plus maze, an object location test, a novel object preference test, and a forced swim test. Analyses of behavioral measures from these paradigms assessed the effects of parenthood and enrichment on boldness, resilience, and memory. We found some evidence of improvements in memory and resilience, suggesting that it is possible to partially overcome bad parental behavior with enrichment. These preliminary results offer evidence that specific environmental interventions could be used to improve specific negative behavioral outcomes in the offspring of bad mothers.

#### P163 - Morphometric analysis of the *Packera dubia* (Spreng.) species complex

Elizabeth Fleming

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*Packera dubia* is a member of the *Packera* genus, known for its morphological variability and difficult classification. The species is widely distributed across eastern North America, with disjunct populations along the gulf coastal plain and midwestern United States. Variation in morphological traits by regional groups (categorized as Atlantic coastal, gulf coastal, granite outcrops of the piedmont, and western) was examined to determine if there is any separation between these distinct populations. Vouchers of *Packera dubia* identified and collected from states in the eastern and midwestern United States in the spring and summer of 2020, as well as specimens obtained from the UNC Herbarium, were measured based on several vegetative and floral characters. The morphological data were categorized and visualized using a clustering analysis, principal coordinate analysis, and linear discriminant analysis in RStudio. The field data showed sufficient variation between the populations, especially in indumentum variability and basal leaf data. The herbarium data also demonstrated the predicted variation but with a lower spread. The relatively small population sample sizes and uncertainty of the validity of traits measured contributed to the inconclusive results. Further work on this project will measure traits deemed significant in this study with expanded taxonomic sampling for target populations not covered by the initial sampling.

#### P164 - Examining the potential costs of vibrational signals: Who's listening?

Ainsley B. Shan, Jake Ramalho, Michael Kingston, Jennifer A. Hamel

*Elon University, Elon, NC*

Animals communicate to facilitate behaviors such as mating, foraging, and predator evasion and defense. However, communication can be costly: signals can be detected by both intended and unintended receivers, including eavesdropping predators. In a plant-feeding insect (Hemiptera: *Entylia carinata*), females with eggs or offspring produce vibrational signals during simulated predator encounters, and males produce vibrational signals in a mating context. Because most invertebrate predators of *Entylia* are sensitive to substrate-borne vibrations, we hypothesized that a potential cost of vibrational signaling for this species is the attraction of eavesdropping predators. To test this hypothesis, we examined the responses of a common, generalist insect predator (Coleoptera: *Hippodamia convergens*) to mating signals, maternal antipredator signals, and silence. We played vibrational stimuli to predators through the stems of potted plants using linear resonant actuators, and we monitored the stimuli that were played using microaccelerometers. We found that the amount of time until predators located the stimulus leaf differed among treatments (one-way ANOVA:  $F:3.790$ ,  $P:0.048$ ), but pairwise tests were not significant (Holm-Sidak method: NS). However, a total of 7 predators located the stimulus during treatments in which signals were played, and no predators located the stimulus during the silence treatments. In addition, we found no differences between treatments in the amount of time predators spent moving on plants or in the amount of time that predators remained on plants. We conclude that the vibrational signals produced by *Entylia carinata* have the potential to attract insect predators. Our findings provide evidence that vibrational communication may impact ecological interactions. However, further work is needed to uncover the costs and benefits of vibrational communication for this species.

#### P165 - A Floristic Inventory of the Sheffield Wildlife Management Area, Paulding County, GA

Katherine Dobson<sup>1</sup>, Sebastian Hagan<sup>1</sup>, Will Freyberger<sup>1</sup>, Heather Sutton<sup>1</sup>, Aliya Davenport<sup>2</sup>

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Sheffield Wildlife Management Area (WMA) is a 2,232 ha property located in northwest Paulding County, Georgia. It is situated within the Talladega Upland ecoregion, which is characterized by its distinctive geology. The Sheffield property is home to several hundred hectares of an increasingly rare plant community type – montane longleaf pine woodland, and areas in the WMA are being managed for optimal montane longleaf conditions. To inform conservation and restoration efforts, a comprehensive floristic inventory was conducted to assess the vascular plant diversity present within the Sheffield property. Over a span of four years, the WMA was surveyed weekly or biweekly during the growing season (March through October). In addition to montane longleaf pine woodland, habitat types surveyed include: mixed upland forest, mesic forest, bottomland forest/floodplain, disturbed areas and riparian zones. Plants were collected in triplicate when possible, and GPS coordinates were recorded. Plants were identified to species level using regional floras, and vouchers will be deposited at Reinhardt University (REH), the University of Georgia (GA) and Kennesaw State University. Currently, 380 species have been identified across 101 families of vascular plants. Of these, 60 species are classified as rare or uncommon in the Georgia piedmont, and 18 of these rare or uncommon species are endemic to the southeastern United States. Four

additional species have never before been reported from the Georgia piedmont. This species list provides numerous county records for Paulding County, a previously understudied area of NW Georgia.

P166 - Viral Spillover from Honeybee Apiaries in Native Bee Populations

Matthew Johnson, Maryam Ahmed, Mary Frances Burrows, Emma Rasco, Daniel Ledrowski, Jennifer Geib *Appalachian*

*State University, Boone, NC*

Over the past 20 years native bee populations have been on the decline. One potential class of stressors affecting native bee populations include novel pathogens that originate from commercial colonies of *Apis mellifera* (honey bees). Recent studies documenting honey bee virus spill over into native bee populations have generally been limited to eusocial species, e.g. bumble bees. This study seeks to determine the degree to which native solitary bees are infected by common diseases carried by commercial honeybee populations: Kashmir Bee Virus (KBV), Israeli Acute Paralysis Virus (IAPV) and Deformed Wing Virus (DWV). I have collected native bees within and outside of the foraging range of managed *Apis mellifera* apiaries and will quantify infection rates by KBV, IAPV, and/or DWV. 73% of the collected samples are part of the family Halictidae which are commonly referred to as sweat bees with the next largest family being Apidae (honeybees and bumblebees) at 21.7% of collected samples. Samples will be homogenized and total RNA will be extracted then converted into cDNA which will be amplified using q-PCR and oligo primers specific to each virus. I hypothesize that distance plays a role in the intensity of viral spillover from managed honeybees to native solitary bee species. Furthermore, I predict that native solitary bees within the foraging range of *Apis mellifera* will be infected with KBV, IAPV, and DWV more frequently than those outside the foraging range. Native solitary bee species are critical pollinators for the plants indigenous to the area, making them an important component of healthy native ecosystems. Understanding how honeybee viral spillover affects our solitary bees is a key piece of information that can be used to help implement strategies to prevent apiaries from passively infecting solitary bee species and maintain our indigenous wildlife.

P167 - The Consequences of Pollination Competition on the Reproduction of *Astragalus bibullatus*

Ashley Gereben

*Middle Tennessee State University, Murfreesboro, TN*

Morphological and phenological similarities of flowers may result in species competing for pollinators, and this competition may be particularly intense for rare species. *Astragalus bibullatus* is a federally endangered legume endemic to the cedar glades of middle Tennessee. It co-occurs with a nearby flower homolog, *Pediomelum subacaule*, which is abundant and significantly overlaps in flowering time. In this study, the competitive effects of *P. subacaule* on the reproduction of *A. bibullatus* were assessed by comparing pollinator visitation rates, quantifying the constancy of pollinator taxa, and determining the frequency of interspecific pollination via field stigma collection. The most common pollinator of *A. bibullatus* and *P. subacaule* is the Eastern bumble bee, *Bombus impatiens*. This pollinator had a significantly higher visitation rate to *P. subacaule* than *A. bibullatus*, indicating that *P. subacaule* may be favored by *Bombus impatiens* and thus reducing pollination of *A. bibullatus*. Field observations on pollinator movement were collected to quantify the species constancy of pollinators using a Constancy Index with a scale of 0 (completely inconstant) to 1 (completely constant). The average constancy of all pollinators was 0.899. To determine the frequency of foreign pollen deposition, 100 stigmas of *A. bibullatus* and *P. subacaule* were collected, and the number and species of pollen grains were recorded. The frequency of foreign pollen grains present in samples was significantly higher in *A. bibullatus* (22% of stigmas) than in *P. subacaule* (4%). *A. bibullatus* samples had significantly more conspecific pollen grains on their stigmas than *P. subacaule*, with averages of 63.11 and 30.97, respectively. The results of this study indicate these two plant species are competing for pollination services, consequently reducing pollinator visits to *Astragalus bibullatus* and increasing the frequency of foreign pollen deposition. Future studies will aim to quantify the reproductive loss of *A. bibullatus* due to foreign pollen deposition.

P168 - Smart SOD: The Smart Soil Organism Detector

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Soils are the most biodiverse ecosystems on earth yet are the least understood and poorly protected. Forming the basis of almost all terrestrial ecosystems, soils are critically important to life on earth with their function mediated by an astonishing diversity of soil organisms. Studying these organisms is difficult, however, and is complicated by the inaccessibility of belowground systems and diversity in the soil. The Smart Soil Organism Detector (Smart SOD) is an instrument and pipeline that can non-destructively extract, isolate, count, sort, and identify live organisms directly from soil at rates of more than 1,000 per minute. Here we highlight how this instrument works and discuss preliminary successes accurately identifying a suite of nematode species and soil microarthropods with a focus on applications to soil biodiversity, species discovery, and conservation.

P169 - Predicting Oxalic Acid in Commonly Eaten Food Source in South Georgia

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Oxalic acid is a highly toxic organic compound that is widely found in plants consumed as food products. Humans do not possess the enzymes needed to metabolize oxalic acid, and this potentially toxic compound may accumulate in the body and absorb into the urinary tract to form urine, or it can combine with calcium, forming insoluble calcium oxalate, which is eliminated in feces. However, it has been shown that calcium oxalate may cause kidney stones which is associated with several risk factors including diet, climate, race, and gender, with men having a higher risk of development.

The objective of the study was to quantify the concentration of oxalic acid in plant food products eaten in South Georgia. Plant tissues were prepared from spinach, beet greens, kale, sweet potatoes, and turnip greens, dried at 70°C for 48 h, ground, then filtered through a 1-mm filter screen and stored at 25°C. Total oxalic acid in the sample was extracted with distilled water and HCl, respectively. Samples were further heated in boiling water bath for 18 min, filtered (0.45 µM) and adjusted to pH 3.0. Determination of oxalic acid contents was performed by solid phase extraction of analytes by ion exchange phase highly polar column HPLC. Sample of 10 µL were chromatographed at 30°C using 15 mmol NaHCO<sub>3</sub> as eluent at a flow rate of 1.5 mL min<sup>-1</sup>. Preliminary results indicated that all six-food products contained oxalic acid. This suggest that consumption of large quantities of these food products may contribute to disease conditions and should be investigated more thoroughly. P170 - SEM-based Study of Fecal Samples from the Harvestman *Leiobunum formosum* (Arachnida, Opiliones)

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Harvestmen are generalist predators of small invertebrates with omnivorous diets that may also include fungi and fruit. Relatively little is known about prey handling or the composition of the diet for most species, including common, southeastern species such as *Leiobunum formosum*. From 2 September-18 November 2023, we observed 360 harvestmen inhabiting mixed deciduous forest on the campus of Virginia Wesleyan University. Surveys were conducted in 2 hours increments each week during morning, afternoon and evening hours. Adults were inspected for the presence of food items being held in the chelicerae with items collected and preserved in 70% ethanol for further analysis. Observed prey included ants, coleopteran larvae, millipedes, snails and dead conspecifics. Additional harvestmen were collected and housed in the laboratory on campus and fed different foods including blueberries, slices of mango, live termites or small pellets of dried dog food. We made 31 observations of food handling and collected 71 fecal samples from these individuals (35 prior to feeding and 36 after feeding). In an effort to gain insight into food handling, we used scanning electron microscopy (SEM) to examine food items after feeding. To further characterize the diet of these harvestmen, we also used SEM to survey the composition of the fecal samples.

P171 - Potential use for Acetyl Salicylic Acid on the Germination and Root Formation of *Pisum sativum* and *Lactuca sativa*

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This project will investigate the potential use of Aspirin as a stimulate seed germination and root production. Aspirin is one of the most commonly used medications in the world, and its chemical properties have been investigated by for their benefits to plant growth in both scientific and home settings for years. Aspirin, C<sub>9</sub>H<sub>8</sub>O<sub>4</sub>, is a synthetic version of acetyl salicylic acid which will be diluted to concentrations of 10<sup>-3</sup>Mmol, 10<sup>-4</sup>Mmol, 10<sup>-5</sup>Mmol, 10<sup>-6</sup>Mmol, and 10<sup>-7</sup>Mmol then applied to *Pisum sativum* and *Lactuca sativa*. The seeds will be sterilized using a chemical wash and plated to grow on a nutrient-free gelatin agar for intervals of one-week. Each agar plate will have 10 seeds of the same species lined along the center of the plate allowing for root and shoot growth at both ends and seeds will be oriented in the same direction of root and shoot growth. Allocation of 1mL aspirin concentrations will be applied to each agar plate and allowed to soak in before placing the seeds on plate. At the end of each week, root growth and hypocotyl length will be measured using the online program Image-J. The date of cotyledons emergence will also be recorded for comparison purposes. The development of the seeds exposed to treatment will be compared to that of a control group of seeds of the same species without the Aspirin solution in the same conditions. The goal of this experiment is to establish whether acetyl salicylic acid is effective in promoting seed germination, and if it is, what dilution is it most effective. The results of this experiment could later be utilized for horticultural and conservational purposes where increased seed germination is needed.

P172 - Beavers and Bugs: Can Beaver Dams Help Improve Macroinvertebrate Diversity in Streams?

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American beavers, *Castor canadensis*, are adapted to living in urban streams and can restore the channels to pre-degraded conditions. Beaver dams slow the flow of water in the stream, retain stormwater, filter out sediment, and can return channels back to their original shape, which can improve the overall habitat and quality. Beaver dam analogs (BDAs) are a restoration technique that utilizes man-made beaver dams in degraded stream locations where beavers are not as abundant, in an attempt to restore the stream channel and improve the water quality. Macroinvertebrates serve as bioindicators of stream health, and the presence and diversity of pollution-sensitive taxa can indicate the quality of the stream. In Atlanta, Georgia in 2018 and 2021, we sampled stream macroinvertebrates both upstream and downstream of a few recently installed BDAs at the Blue Heron Nature Preserve. Riffle kicks and bank sweeps were performed at both sites and the samples were stained to dye the organisms. The contents of each sample were sorted to pick out the macroinvertebrates, which were then identified to family and genus. The Shannon-Weiner Index, Hilsenhoff Biotic Index, and the Adopt-A-Stream Index were calculated for both the upstream and downstream sites to compare the amount of diversity and the quality of the water between 2018 and 2021. Trends showed that the samples taken downstream of the BDA in both 2018 and 2021 had a higher taxa diversity than upstream samples. The highest Adopt-A-Stream and Shannon-Weiner index values were calculated for the downstream site in

2021, indicating less pollution and greater macroinvertebrate diversity. Since the installation of the beaver dam analogs in 2020, the overall data for this stream suggests that the BDAs are effective in filtration of pollution and sediments, therefore increasing the water quality and levels of biodiversity.

P173 - Testing the Efficacy of *Methylobacterium oryzae* Bacterial Inoculation to Improve the Quality of Hydroponic, Salt-Stressed Tomatoes

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Salinity stress can dramatically impact crop yield and quality, especially in hydroponic tomato cultivation. Nutrient solutions with artificially-raised salinity can improve tomato fruit quality and chemical profile, but at the tradeoff of reduced yields. Increasing market demand for hydroponic produce therefore requires methods to modify fruit quality without significantly compromising yield. *Methylobacterium oryzae*, a leaf-borne symbiotic bacteria, has been shown to relieve the negative impacts of salinity stress on inoculated crops through the production of Indole-3-acetic acid (IAA) hormone and the enzyme 1-aminocyclopropane-carboxylic acid (ACC) deaminase. Using *M. oryzae* as a bioinoculant could therefore provide a means of increasing hydroponic crop salt tolerance. During the spring and fall of 2022, we conducted two greenhouse trials consisting a two-way factorial design with high salinity (EC = 3.5 (Fall) or 5.0 (Spring)) and foliar inoculation of talc-immobilized *Methylobacterium oryzae* as experimental treatments. Tomato plant health was continually monitored via chlorophyll fluorescence measurements. Tomato fruits were harvested upon ripening, weighed, and assessed for the presence of Blossom End-Rot (BER) Disease. Fall tomatoes were additionally analyzed for sugar content percentage. Salinity significantly reduced fruit weight and fresh plant biomass and significantly increased fruit sugar percentage and the frequency of fruit BER, but it had no significant effect on chlorophyll fluorescence measurements in either trial. Fruit number was not significantly affected by either treatment, with the salt/inoculated treatment having 40.1% more and 4.78% fewer fruits than the salt/non-inoculated treatment group during the spring and fall trials, respectively. Inoculation significantly reduced the frequency of BER, but only in salt-stressed plants during the spring trials. Further trials manipulating the method of inoculation (seed inoculation, foliar spray, etc.) are needed to fully assess the potential of *Methylobacterium oryzae* as a biological control against salt stress in hydroponic settings.

P174 - A Pleistocene age scaly cricket, *Cycloptilum carolinensis* sp. nov. (Orthoptera: Gryllidae: Mogoplistinae), of the Martin Marietta Locality

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The Pleistocene Martin Marietta locality of southeastern North Carolina (34° 22. 368' N, 77 ° 50.356' W) has produced a plethora of floral diversity, while also including the rare remains of a mummified insect. The specimen was identified as a scaly cricket, *Cycloptilum*, representing the first occurrence of the genus in the fossil record. Preservation of scales, maxillary palps, and complete leg (including foretibia, hind tibia, and femur) were key characters for identification to genus, with differences in pronotum size used for species determination. Common scaly crickets are staples in southeastern North Carolina ecology, and represent an important floral herbivore-decomposer and faunal food source. Discovery of a scaly cricket at the Martin Marietta locality indicates that the ecology and climate during the Sangamon interglacial was similar to modern southeastern ranges.

P175 - Preliminary data in the Species Delimitation of the Native, Southeastern Early Blue Violet (*Viola palmata* L.) species complex using morphological, ecological, and genetic datasets

Jonathon Osborne

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Members of the genus *Viola* are widespread throughout the Northern Hemisphere and high elevations in the Southern Hemisphere. With ~545 species among 31 sections, *Viola* is one of the largest genera in the world. Despite this, the ecology and classification of the genus has been understudied in the Southeastern US. The *Viola palmata* group in particular has been historically confused, split, and/or synonymized by past researchers even since Linnaeus's initial description in his *Species Plantarum* (1753). With only cursory or confounding information having been brought forth to describe this species group, the Alford Lab at USM in conjunction with the Ballard Violet Lab at Ohio University have set out to clarify this complex species group. We seek, through traditional as well as modern genetic, ecological, and morphometric evaluative methods, to determine the number and circumscription of species in this group. Genetic analyses include sequencing of the nuclear ITS and plastid *trnL-F* gene regions. A multitude of morphological characters among sampled populations and herbarium specimens are being evaluated; these morphological characters include chiefly seed coat texture and colour, petal trichomes, and lobed leaf dimensions. Preliminary data suggests some consistent morphological character differences among large-scale ecoregions in the southeast (Piedmont, Gulf Coastal Plain, etc.). Thus far, 5-10 individuals from 11 sampled populations have been brought to USM for a common garden experiment and closer observation. Along with these live specimens, more than 200 herbarium specimens of *V. palmata* L. and allies from the region are on loan to USMS for further examination and to supplement any geographic gaps in sampling.

P176 - Preliminary genetic analysis of microbial endosymbionts in kudzu bugs, *Megacopta cribraria* (F.), from 2012 to 2021.

Erika Niland, Ethan Hathcock, Lauren Hall

*Wingate University, Wingate, NC*

Co-evolution of insects and plants have been extensively studied, and invasive insects are better suited to switch food sources to adapt to their surrounds. Such adaption could lead to variations in endosymbiont composition. Some research suggests that invasive insects can adapt to feed on available food sources, therefore, suggesting that their microflora may be different in those populations that feed on soybean compared to those on kudzu. These differences could, therefore, be an adaptive change in populations and would be apparent in genetic sequence analysis from populations of endosymbionts collected in 2012 compared 2021 collections. The aim of this project was to analyze genetic sequences of endosymbionts for change over time.

P177 - Can we reverse the trend of declining botanical capacity by using high-impact practices in botany education?

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The loss of botany degree programs, retirement of trained botanists, and under-appreciation of plants has reduced botanical capacity of the U.S. at a time when this expertise is critical for human health and ecosystem stability. The botanical community has collectively developed creative approaches to teach plant biology, but it can be difficult to manage an entire plant-based curriculum within a biology department, especially if botany faculty are few. At Old Dominion University, we have embarked on a three-year project to revamp how we teach plant biology, with three primary goals: 1) to make students more aware of plant-associated careers earlier in their undergraduate programs, 2) to provide students with hands-on experiential learning of botanical concepts throughout the curriculum, and 3) to build a botanical community of students, faculty, and other stakeholders on our campus that is diverse, inclusive, and supportive. Here, we detail our experiences in navigating institutional logistics to implement high-impact practices in our courses and to reach students through formal and informal interactions. After one year, we have had some successes, and we also faced challenges. Students participating in the summer research program reported positive gains in problem solving skills, time management, and self-confidence in doing research and a greater desire to find additional opportunities and to explore plant-related careers. While interest in gaining research experience was strong among existing students, we failed to register any freshman for a Learning Community last fall. Not surprisingly, the activities that enabled personal connections between students and with their faculty mentors have proven the most beneficial. In the coming years we will explore a greater diversity of practices to build student networks on and off campus. The implementation of more personalized education may be the key to cultivating diverse plant biologists and reversing the trend of declining botanical capacity. P178 - Life History of an Invasive Genera of Earthworm *Amyntas*

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*Amyntas* is an invasive genus of earthworm native to the Korean peninsula and many of the Japanese islands. It is considered potentially damaging to the native ecosystem via its detrimental effects towards soil quality, the competition with other species of earthworms, and its disturbance to animals which naturally feed on earthworms such as salamanders. To limit the threat that this genera poses it would be best to reveal the life history of the genera in question. This has been done via a survey plot conducted on a weekly basis for two seasons over the course of two full years. The purpose for this study was not only to uncover the unknown ecological damage that this species could be causing, but to hopefully lead to a solution in the future. *Amyntas sp.* are prolific breeders and can achieve high densities within forest soils. A population of them requires large quantities of leaf litter, are known to disturb natural soil content including nutrients, which can damage plant communities. This is especially damaging to juvenile plants as *Amyntas sp.* are capable of consuming large amounts of undergrowth and preventing new trees from growing.

P179 - Analysis of programs to support the success of nontraditional transfer students in a STEM major

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Nontraditional transfer students face unique barriers to degree completion, and this is particularly true for STEM majors. In 2021, Athens State University developed a program to support STEM transfer students, funded by a National Science Foundation S-STEM grant. This program, called the Athens State Scholars in STEM (ASSIST) Program, is designed to alleviate common setbacks to degree completion experienced by nontraditional transfer students. We present a summary of the ASSIST Program and an initial analysis of students' perceptions of the program after the first year of implementation. Overall, we found that support programs focused on minimizing time to degree completion were rated highest by students (faculty mentorship, STEM-focused orientation course), followed by support programs designed to help students in balancing family and work with school commitments (biweekly support emails, loaner laptops, and online tutoring). Those programs designed to prevent switching away from a STEM major by increasing career readiness were rated lowest by students (conference travel funds, research funds, seminar series). These programs generally required an extra time commitment from students beyond their coursework. Students' perceptions of conference travel and research funds may increase as the cohort nears graduation and has more opportunities to participate in these programs. This initial analysis highlights the importance of mentorship practices that provide a clear path to degree completion for STEM transfer students and that help students balance competing commitments.

P180 - Size-dependent Foraging in Dusky Salamanders (genus *Desmognathus*)

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In behavioral ecology of animals, there have been two historically recognized foraging modes, ambush predators and active hunters, depending on the type of available food. We tested the hypothesis that foraging mode in the genus *Desmognathus* depends on salamander size. We chose five species covering the range of body sizes within the genus with two replicate streams for each species. We collected animals at night during their optimal foraging time. We presumed an animal protruding from refuge to be foraging in ambush and one to be outside of a refuge to be actively hunting. We measured snout-vent length (SVL) using a digital caliper and tested for differences in SVL for each species (or complex) with a 2-way ANOVA, setting foraging mode and stream as factors. We further tested for a relationship between the proportion in ambush and maximum SVL using regression. In a twice-replicated laboratory experiment, we put 20 adults and 20 juveniles of the largest species (*D. amphileucus*) and 20 adults of the *D. ocoee* complex in boxes with appropriately sized refuges made from PVC pipe. We placed them on a 14:10 day-night cycle with white light representing day and red light representing night. After one week, we observed them at night to determine their location. From the field data, we found that, for every species, refuged animals were significantly larger than wandering individuals. Moreover, the proportion of ambush predators was significantly dependent on maximum SVL. Contingency analysis of the laboratory data showed no significant difference in inside versus outside of the refuge among the different-sized animals, suggesting this behavior is not innate. We concluded that foraging mode is dependent on body size within and between species. However, this behavior appears to have some link to the natural environment.

P181 - Public Demonstrations of the Cellulose Acetate Peel Technique on Coal Balls as a Means of Promoting Paleobotany

James Mickle

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Fossil fairs at museums and schools are popular public events that usually run 1 – 2 days with displays of fossil collections, children’s activities, movies, and speakers by lay and professional paleontologists. Active demonstration of techniques used to study fossils are not typically included, and animal fossils are usually emphasized, with few fossil plants on display. Coal balls are calcium carbonate permineralizations preserving Carboniferous age coal swamp peat, and that provide an excellent opportunity to showcase both fossil plants and a technique used in the study of fossils. Coal balls are typically studied using the cellulose acetate peel technique. Using a portable lab set-up, the cellulose acetate peel technique provides a unique opportunity to easily and quickly demonstrate a procedure used to study plant fossils in a public setting, and at the same time raise public awareness of the plant fossil record. This technique is simple to do, easy to follow, and gives immediate results that are clearly demonstrated to the public. Properly done, the peel technique is safe and gives visitors dynamic insight into how fossils are studied rather than a static viewing of prepared materials.

P182 - The evaluation of the potential developmental risk associated with two commercial dyes (Methylene Blue and Phloxine B) using the Frog Embryo Teratogenesis Assay Xenopus (FETAX).

Kritika Maharjan<sup>1</sup>, James Rayburn<sup>2</sup>

of MB and Ph B, the Frog Embryo Teratogenesis Assay – *Xenopus*(FETAX) was performed to assess the developmental toxicity. FETAX is a 96hour test that measures the potential of chemicals to induce death, deformity, and growth inhibition in developing embryos using early-stage embryos of the South African clawed frog (*Xenopus laevis*). The objective of this study is to determine the potential developmental toxicity of MB and Ph B. To assess the photoactivity, the Ph B trials were carried out in both light and dark settings. Fertilized embryos were separated for the studies and allowed to develop in test solutions. The LC50 (mortality), EC50 (malformation) for MB and Ph B (in light and dark) were determined using probit analysis. The Teratogenic Index (96-hr LC50 / 96-hr EC50) was calculated to determine the risk to amphibian embryos. Overall, the results indicate these dyes have the potential to cause malformations and risk to amphibian embryos. P183 - Compositae in a Crate: an outreach initiative to promote plant awareness

Erika Moore-Pollard<sup>1,2</sup>, Carolina Siniscalchi<sup>3</sup>, Kathryn Parsley<sup>4</sup>, Jaime Sabel<sup>1</sup>, Jennifer Mandel<sup>1,2</sup>

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Chemical dyes are one of the most commonly utilized chemical substances in our daily life, as they are found in food, textiles, and medications, resulting in high environmental exposure. Methylene blue (MB) and Phloxine B (Ph B), which are both used in textiles, food, and even pharmaceuticals, will be tested for teratogenic effects in this investigation. Methylene blue is most commonly used as a bacteriologic stain and indicator, whereas Ph B is usually utilized as an agar plate stain. Methylene blue has been demonstrated in recent research to produce severe central nervous system toxicity, as well as nitrogen and ammonia poisoning. Ph B is also an effective photosensitizer of cellular membrane damage, meaning skin exposure to the dye and sunshine or artificial light may result in phototoxicity. To better understand the adverse developmental effects

Studies on the perception of animals and plants often report that students prefer to learn more about animals than plants and usually have more difficulty noticing plants in the environment. This could impact conservation programs and initiatives, as animals are often considered more important and may be deemed worthier of conservation efforts. Providing students an opportunity to connect to plants and understand how they affect their lives is one step toward raising awareness around this issue. Compositae, also known as Asteraceae or the sunflower family, is the largest family of flowering plants, accounting for ~10% of flowering plant diversity in the world. They are present in virtually all biomes and environments and fulfill multiple ecological niches. Some members of the family are widely cultivated as crops, such as sunflower, lettuce, artichoke, and chicory; several species are grown as ornamental plants; and others are used in the pharmaceutical and chemical industry. Compositae's presence in so many contexts make the family a good candidate for activities aiming to increase plant awareness, as several Compositae species are already present in our everyday lives. Here we present Compositae in a Crate, a teacher-guided outreach activity aimed at the 4th and 5th school grades. This crate of activities contains four different modules focusing on different aspects of the sunflower family: biodiversity, morphology, society, and genomics. Each module has different learning objectives and can be used independently from each other. A guide and different activities, such as flash cards, puzzles, and 3D models, are provided in each module. The activities will be prepared to meet the national and state of Tennessee education standards for the 4th and 5th grades. The produced crates will be available at different institutions and all materials will be made available online so other interested parties can produce their own crates.

#### P184 - Aquatic habitat resource use of Eastern Box Turtles in northeastern Georgia

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Anthropogenic habitat loss, fragmentation, and degradation lead to decreases in resources, biodiversity, and ecosystem services; thus, understanding how species use specific resources is vital to conservation and restoration efforts. Eastern Box Turtles (*Terrapene carolina carolina*) are a primarily terrestrial species native to the eastern United States that has experienced population declines throughout its native range largely due to habitat loss and alterations. Although the species is considered primarily terrestrial, phylogenetic studies indicate Eastern Box Turtles (EBT) evolved from an aquatic turtle species, suggesting that aquatic habitats may represent an important resource for the species when considering conservation efforts. To investigate this use in northeastern Georgia, we analyzed habitat use of 46 individual EBT (26 M, 20 F) located using radiotelemetry 1-2 times per month for periods of 9-110 months between 2013-2022. Aquatic habitats at our study site included freshwater forested/shrub wetlands created by beaver activity along a permanent creek in addition to multiple, unconnected seepage wetland areas dominated by either Chinese Privet (*Ligustrum sinense*) or native woody and herbaceous vegetation. Preliminary analysis indicates a substantial percentage of radiolocations were recorded in wetland or seepage habitats; however individual use of these aquatic resources varied, with at least 70% of the turtles having locations recorded in these aquatic areas. The high percentage of EBT using aquatic habitats supports the need for consideration of aquatic habitat in conservation efforts.

#### P185 - Assessment of the macroinvertebrate community and water quality in Mill Creek

Bloo Mitchell, Priscilla Dickey

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The Conasauga River and its tributaries are home to a diverse assemblage of fish and invertebrates; however, researchers have noted a decline in local biodiversity over recent decades. These declines occur downstream in areas where agriculture, industry, and residential and commercial development are the primary land uses. Although specific causes for these declines are uncertain, it is hypothesized that high levels of nutrients, pesticide pollution, sedimentation, or a combination of these stressors may be responsible.

A local project, known as the Park Creek Elementary Restoration Project, aims to improve native fish, plant, and pollinator habitats around a portion of Mill Creek and an associated tributary that is the habitat to a federally threatened fish called the trispot darter (*Etheostoma trisella*). To ascertain a baseline of the water quality in this area, measurements of water quality and physical in-stream habitat characteristics were made. Three macroinvertebrate sampling methods were utilized over a four-month period at three sites (two sites adjacent to the restoration area - a Mill Creek site and an ephemeral tributary site - and an upstream reference site).

The results of this study indicate that the downstream site is impaired, and the adjacent tributary is highly impaired, with stark differences in macroinvertebrate diversity between sites. Higher conductivity levels over the length of the study and a sharp reduction in dissolved oxygen in the warmer months was observed at the tributary site. These data will serve to track the progress of the Park Creek Restoration Project in improving water quality.

#### P186 - Diet of larval *Ambystoma opacum* and the effects of *A. opacum* on feeding activity and movement of other ambystomatid larvae.

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A previous study conducted in Weakley County, Tennessee, showed that larval growth rates of Mole Salamanders (*Ambystoma talpoideum*) increased after emergence of Marbled Salamander (*Ambystoma opacum*) metamorphs from ephemeral pools. To help explain the increase in growth rates observed in this study, we suggest that the presence of larval Marbled Salamanders may limit feeding opportunities of other ambystomatid salamander larvae, thereby resulting in slower growth. Our study had two main objectives: (1) describe the diet of larval Marbled Salamanders to

establish that they are potential predators of congener larvae in naturally occurring vernal pools in West Tennessee and (2) to determine if larval Marbled Salamanders influence the feeding success and movement patterns of larvae of other species under laboratory conditions. We determined that small size classes (10 mm – 20 mm SVL) of larval Marbled Salamanders feed primarily on copepods, and ostracods, whereas larger size classes (20.1 mm – 40.0 mm) include larger prey such as cladocerans, amphipods, and isopods. At least one individual contained a suspected larval salamander. We are currently conducting behavioral trials in which chemical cues of larval *A. opacum* are introduced to experimental chambers to assess the effects of predation risk on (1) the number of prey items consumed by larval prey species (*A. maculatum*, *A. talpoideum*, or *A. texanum*) and (2) the likelihood of prey species to move from refugia.

P187 - Gastrointestinal helminths of Eastern Bluebirds dead from the cold in southwestern Kentucky: Were 'buttworms' to blame?

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We examined the intestinal tract of approximately 50 Eastern Bluebirds, *Sialia sialis*, for helminth parasites. These birds were found dead in nest boxes after a winter storm in the Land Between the Lakes area in southwestern Kentucky. We will present a helminth checklist and examine how host sex, age and overall physical condition relates to the parasite community structure.

P188 - Blood lactate levels in diamond-backed terrapins (*Malaclemys terrapin*) captured in crab pots

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Bycatch of diamond-backed terrapins (*Malaclemys terrapin*) and other non-target species in the commercial blue crab (*Callinectes sapidus*) fishery is an issue of concern for fisheries managers and conservationists. Unintentional capture of terrapins in crab pots can result in death by drowning or sublethal effects, such as physiological disruptions due to overexertion or limited access to air. Terrapins that are forcibly submerged in crab pots may resort to anaerobic metabolism to meet energetic needs, resulting in a buildup of lactate in blood and tissues. Lactate accumulation may negatively impact physiological functioning by lowering the animal's internal pH and disrupting molecular and cellular processes. Prolonged periods of enforced submergence may result in high levels of lactate in the blood and tissues, and potentially require a recovery period in order to clear lactate from the system. The primary goal of this study was to investigate the metabolic response of terrapins to enforced submergence in crab pots by documenting blood lactate levels as a measure of reliance on anaerobic metabolic pathways. Blood samples were collected from terrapins captured in crab pots and blood lactate was determined using a commercially available spectrophotometric assay. General linear models will be used to assess the factors (body size, water temperature, capture location) that affect blood lactate levels during entrapment in crab pots. Data analyses are ongoing. Results from this study will enhance our understanding of the sublethal impacts of bycatch on the behavior and survivability of terrapins released from crab pots and potentially contribute to development of protective measures to mitigate terrapin bycatch.

P189 - The effect of larval competition on adult life history traits and the immune response of adult cat fleas (*Ctenocephalides felis*)

Piper Zellner, Lisa Brown

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For many holometabolous insects, conditions experienced during the larval stage can carry over to influence the adult stage. For example, in mosquitoes, reduced larval food and high larval density can increase adult susceptibility to infection. Fleas (Siphonaptera) are holometabolous insects with an adult stage that feeds exclusively on the blood of a vertebrate host, including humans. However, during the larval stage, fleas have chewing mandibles and derive their nutrients from the vertebrate blood proteins excreted in the feces of adult fleas. The unique nutritional requirement of larvae, as well as their limited mobility, greatly restricts the area where development can occur. Thus, larval competition is likely frequent under natural conditions, but the influence of larval density on the subsequent adult stage has not been examined. Using the cat flea (*Ctenocephalides felis*) as our study organism, we tested the effect of larval competition on adult survivorship and adult size, as well as on the adult immune response. Specifically, eggs were collected and divided to create intraspecific larval densities, and hatched larvae from all groups were fed the same amount of adult fecal pellets. Survivorship was measured by tracking the proportion of larvae that reach adulthood, and the length of the abdomen was measured to determine adult size. Additionally, eclosed adults will be fed live bacteria in their bloodmeal for 24 hours, and those individuals will be assessed for infection by plating dissected guts on selective media. Our preliminary results suggest larval density has no effect on adult survivorship, but does influence adult size. Overall, these data yield significant insight into how the ecology of larval fleas impacts the biology of the resultant adults.

P190 - Elderflower extract improves health of a cancer model in *C. elegans*

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Cancer effects one in three people in the United States and costs the American healthcare system over \$21 billion per year. Medicinal plants represent an untapped resource for numerous potential therapeutics. The goal of this research study is to use *Caenorhabditis elegans*, a nematode roundworm, as a model organism to find novel plant therapeutics that can shrink tumors. *Caenorhabditis elegans* is an ideal model organism for studying the progression of tumor growth because of its short life cycle and transparent body. This study exposed *C. elegans* to *Sambucus nigra* (elderflower) extract and measured their overall health by egg laying potential. Both a wild type *C. elegans* (N2) and a mutant strain *C. elegans* (MT10430) were used. The strain MT10430 was selected because this mutation causes tumors in the reproductive area, resulting in fewer eggs being produced. This allowed for the eggs to be used as an effective measurement of their health and tumor size. Four plates, a control wild type (NGA N2), a control mutant strain (NGA MT10430), an experimental wild type (EF N2), and an experimental mutant strain (EF MT10430) were created after age synchronization. The amount of time necessary to lay eggs varied, due to the mutation's effects, but it was typically a couple of days per trial. The EF MT10430 experimental plate showed a drastic increase in egg laying capacity when compared to the control NGA N2 plate, suggesting that the treatment is shrinking the tumors. This study is being expanded upon, awaiting sample submission for RNA sequencing and fluorescent imaging.

#### P191 - Sexual dimorphism in crayfish chelae: a sporadic occurrence or a frequent reality?

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Sexual dimorphism in the crayfish family Cambaridae (Decapoda) is commonly characterized by reproductive phenotypes in males (form I, form II), but reproductive phenotypes have rarely been reported in females. However, recent works have shown that form alteration in females may be more common than researchers think which could have unknown implications for various types of morphological analyses. For example, crayfish taxonomy is traditionally based on form I males, but morphological comparisons used in crayfish taxonomy are commonly used for form II males and females in addition to their form I counterparts. The presence of reproductive phenotypes in females could bias these traditional morphological analyses. Geometric morphometrics allows researchers to characterize shape and how it may vary between individuals or groups. Here, we analyze a number of North American crayfish species using geometric morphometric techniques to assess intra- and inter-sexual dimorphisms of crayfish chelae.

#### P192 - Fisetin inhibits expression of fatty acid synthase and induces metabolic stress leading to apoptotic cell death in LNCaP prostate cancer cells

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Prostate cancer is the most commonly diagnosed cancer and the second leading cause of cancer-related death in men in the United States, making the disease a significant public health burden. Fisetin (3,3',4',7-tetrahydroxyflavone) is a naturally occurring flavonoid found in fruits and vegetables such as strawberries, tomatoes, and onions. Fisetin is known to possess antioxidative and anti-inflammatory effects. Fisetin has also been reported to have antiproliferative effects on various cancer cell types including prostate cancer cells. Fatty acid synthase (FASN) is an enzyme that *de novo* synthesizes fatty acid chains. It is known that FASN is overexpressed in many cancers including prostate cancer. FASN-induced elevated rate of fatty acid synthesis promotes cancer cell growth by providing energy source, membrane lipids, and signaling lipids. Therefore, FASN could be a potential therapeutic target in prostate cancer. In this study, we determined the effect of fisetin on the expression of FASN. In cells treated with fisetin, we observed that the level of FASN was depleted when the concentration of fisetin was increased. AMP-activated protein kinase (AMPK) is a sensor of energy status which is phosphorylated and activated when intracellular energy level is low. Fisetin treatment also lowered the energy level in LNCaP cells which in turn activated AMPK and induced apoptotic cell death. These results suggest that fisetin induces metabolic stress, activates AMPK, and leads to apoptotic cell death in these cells. Taken together, fisetin could be a useful chemotherapeutic agent in treatment of prostate cancer.

#### P193 - Differences in ectoparasite communities of larids from an Avian Botulism outbreak

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Feathers provide niches that support diverse communities of ectoparasites, including feather mites, feather quill mites, feather lice, ticks, and sticktight fleas. As ectoparasites interact with host skin, feathers, and blood, spillback of host microbial infracommunity occurs. *Clostridium botulinum* is a ubiquitous bacterium that, in some conditions, can produce a suite of lethal neurotoxins that is deadly to birds, causing Avian Botulism (AB). AB is considered the causative agent for an avian die-off event on Middleton Island, Alaska, where larid colonies suffered hundreds of deaths in 2021. Here, we compare the ectoparasite communities on those affected birds to conspecific hosts collected in 2016 from other colonies under non-die-off conditions. To evaluate the ectoparasites, we washed individual bird carcasses with soap and strained the 'ectowash' through a 50 micrometer sieve into 90% ethanol. We inspected samples under a stereoscope and sorted arthropod ectoparasites into morphospecies, which we then counted. These species are further confirmed with barcoding of the cytochrome oxidase I (COI) gene. Prior to molecular lumping/splitting, 'healthy' Black-legged Kittiwakes (*Rissa tridactyla*, BLKI) carried larger abundances of ectoparasites ( $p\text{-value}=0.027$ ,  $t\text{-test}$ ), but diseased birds contained more diverse communities ( $p\text{-value}=0.018$ ,  $t\text{-test}$ ), exhibiting higher community evenness. Further, we could differentiate among the ectoparasite communities of diseased vs. healthy birds with  $>92\%$  accuracy ( $R\text{-square}=0.67$ , discriminant analysis). Three of the seven feather mite morphospecies contributed to the successful differentiation among the two groups of BLKI. We continue to refine these assessments with molecular

confirmations for this and other larid species, and intend to evaluate microbial communities of hosts and parasites. This research presents prospects of AB evaluations using ectoparasites, a novel and non-invasive method for collecting samples from seabird colonies. P194 - Identification and Metabolic Analysis of Endophytes in Shining Clubmoss (*Huperzia lucidula*)

Benjamin Crawford, Annkatrin Rose

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The objective of this research project is to extract, analyze, and sequence the endophytic fungi found in the Shining Clubmoss (*Huperzia lucidula*). Endophytes are fungi or bacteria that are found living symbiotically within or near the healthy plant tissue. Almost 300 endophytes have been isolated from the shining clubmoss, with around 20 highlighted to be of special interest through a metabolite analysis. Fungi can produce secondary metabolites of special interest to medicinal research, such as Huperzine A. This secondary metabolite inhibits Acetylcholinesterase (AChE), which has been found to be effective in treating Alzheimer's disease. We tested several strains through HPLC analysis and an enzyme inhibition assay. This proved there is a presence of Huperzine A at varying amounts. Only the highest producing endophytes were processed for identification through DNA sequencing. The process began with a DNA extraction of the tough fungal cells and a polymerase chain reaction (PCR) to amplify the target DNA sequences with ITS primers. The DNA was then cloned into plasmids and sent off to a third-party company for DNA sequencing. The results elicited that we isolated endophytes both from the *Ascomycota* and *Zygomycota* phyla. Results from this study can be utilized in future projects to indicate target endophytes that can be cultured for the production of Huperzine A. Further research is being performed to examine the endophytic profile of the plant using metagenomic sequencing. This will allow for the identification of endophytes unable to grow on medium.

P195 - Morphological Analyses of Spiny Scale Crayfish (*Cambarus jezerinaci*) Lineages in Kentucky and Tennessee

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The spiny scale crayfish (*Cambarus jezerinaci*) inhabits small first and second-order streams in Southeastern Kentucky, Eastern Tennessee, and Lee Co. Virginia. Research conducted by Thoma and Fetzner Jr. in 2008 compared *C. jezerinaci* to the closely related *Cambarus parvoculus* which occurs in Western Tennessee. In this study morphological characteristics were compared between *C. jezerinaci* and *C. parvoculus* with only rostral states found to be significant to species identification. This study also identified a third and genetically distinct lineage which is currently denoted as Group B *C. jezerinaci*. In light of new genetic findings, separation of these three lineages prior to morphological comparison is necessary to better understand the true diversity of morphology in each lineage. Ongoing research will separate genetic lineages of *C. jezerinaci* in order to complete morphological analyses on the distinct lineages and determine if there are statistical significance in characteristics. Determination of morphological differences in unique lineages of *C. jezerinaci* is important to the conservation of these animals and expands upon the understanding of what it means to be a species. Continuing to perform research to understand the differences between species and unique lineages allows a better understanding of biodiversity and speciation. Given the rapidly changing climate, it is imperative to catalog the true diversity of freshwater streams and develop conservation action plans for endemic species such as *C. jezerinaci* before they are lost.

P196 - Use of The Mammalian Eye Lens as a Model of Protein Aging

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*High Point University, High Point, NC*

The lens of the eye focuses light on the retina to facilitate vision. To do so, the lens consists primarily of fiber cells which lack many normal cell components, including nuclei and mitochondria. The result is transparent cells that cannot divide or make new proteins. New fiber cells are generated throughout life and form concentric layers on top of old fiber cells. This forms a "spatiotemporal gradient" in which extremely old proteins, synthesized in the embryo, exist in the center of the lens while newly synthesized proteins exist at its outer surface, with protein of every intermediate age existing between. This makes the lens a unique model for studying protein aging since it can be separated into different layers representative of different ages, much like rings within a tree trunk. As these proteins age, they accumulate different modifications which can alter their function and ultimately cause cataracts to form. To determine which modifications accumulate on older proteins, pig lenses were obtained from a local meat packing site and separated into six layers of equal protein content using a dissolution medium. Proteomic analysis was used to identify and quantify differences in post-translational modifications such as phosphorylation, oxidation, acetylation, formylation, and deamination. Most notably, oxidation and deamidation were found to increase at a steady rate as proteins age while other modification had an initial increase and then remained relatively constant with age. This research provides a new tool to study aging and important insights into how we age at a molecular level.

P197 - A Survey of Crayfishes in the Upper Kentucky and Upper Cumberland River Watersheds

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Information on the crayfishes within Kentucky come from two major surveys: the first being Rhoades in 1944 and the second being Taylor and Schuster in 2004. Therefore, there are large gaps in our understanding of the distribution and conservation status of crayfishes throughout this state,

which warrants updated surveys. The Kentucky (2022) survey aims to provide a modern-day assessment of crayfishes found within the Upper Kentucky and Upper Cumberland River watersheds. New information on abundances of local crayfishes will provide insight on conservation concerns regarding endangered and introduced species. Collection efforts were conducted through a multitude of strategies, such as sein netting, dip netting, rock flipping, and burrow digging. Twelve species, from both the genera *Cambarus* & *Faxonius*, were collected from over 40 sites. Multiple sites were burrowing crayfish focused of which seven contained *Cambarus dubius* with high degrees of phenotypic plasticity. The Kentucky (2022) survey resulted in the discovery of 12 sites for *Cambarus jezerinaci* as well as showing an abundance of *Faxonius cristavarius* consisting of over 200 of the 430 total crayfishes collected. Future study efforts hope to shed light on the current populations and conservation concerns of crayfishes within the Upper Kentucky and Upper Cumberland River watersheds. Further collection data is needed to better understand the abundances of species within these watersheds.

P198 - A Synergistic Approach; Testing the Efficacy of Atovaquone, 5-FU, and Etoposide on Colorectal Cancer Cells

Chris Barton, Chris Barton

*Belmont University, Nashville, TN*

Cancer is a major health concern and there is a huge need for future research into new disease therapeutics. One feasible option is drug repurposing, or the use of currently FDA-approved drugs for additional diseases. Recently, a number of reports have surfaced showing that antimalarial compounds are viable options for cancer therapy. Here, we examine the ability of one antimalarial drug, Atovaquone, to work synergistically with 5-FU and Etoposide, two commonly-used cancer chemotherapeutics. Future experiments are necessary to further examine whether these combination drug regimens will serve as feasible options in the clinic.

P199 - Population genomics among mainland and island chewing lice (*Physconelloides eurysema*) from Common ground doves (*Columbina passerina*).

Paige Brewer, Andrew Sweet

*Arkansas State University, Jonesboro, AR*

Despite the importance of parasites for ecosystems and diseases, relatively little is known about the dispersal patterns of parasite between fragmented habitats, such as oceanic islands. *Physconelloides eurysema*, a body-specific feather louse from Common ground doves (*Columbina passerina*), are an ideal system to compare the influence of island biogeography on parasite evolutionary and ecological patterns. Common ground doves and their lice are present throughout southern North America, South America, and the Caribbean islands. The overarching goal of this study is to highlight *P. eurysema* phylogeography and population genetics among several island and mainland populations. We sampled feather lice from mainland and island populations, including from the U.S. (Texas), Ecuador, Brazil, Mexico, Bahamas, Jamaica, and Cayman Islands. This data was acquired through existing specimens and samples accessible on the National Center for Biotechnology Information (NCBI) SRA database. We extracted DNA from available samples using a Qiagen QIAamp DNA Micro kit and sequenced whole genomes using paired-end Illumina reads. We aligned these reads to an existing reference genome to obtain single nucleotide polymorphisms (SNPs) for population genetic analysis. We then used these SNPs to test for population genetic structure among the samples of lice. We also used a partial Mantel test to test for isolation-by-distance (IBD). We expect to find strong genetic structure between mainland and island populations with an IBD pattern. This is because of the limited parasite dispersal among hosts from populations separated by relatively large oceanic distances. We also expect island populations to have reduced genetic diversity due to the founder's effect. This project will further our understanding of parasite-host relationships, dispersal pathways, and divergence patterns among species and populations of taxa from oceanic islands.

P200 - Loss of HMOX1 Sensitizes Colorectal Cancer Cells to Tafenoquine-Induced Apoptosis

Libby Godo, Chris Barton

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Cancer is a major health concern and there is a huge need for future research into new disease therapeutics. One feasible option is drug repurposing, or the use of currently FDA-approved drugs for additional diseases. Recently, a number of reports have surfaced showing that antimalarial compounds are viable options for cancer therapy. Our group has previously shown that one antimalarial compound, Tafenoquine, is extremely useful in decreasing the viability of multiple cancer types grown in culture, though the mechanism of how this drug is affecting cellular viability is currently unknown. Here, we show that loss of HMOX-1, a gene that serves a protective role against oxidative stress, sensitizes cells to Tafenoquine-induced apoptosis. These data suggest that Tafenoquine may negatively affect cellular viability through the induction of oxidative stress. Further research is underway to continue examining the role of HMOX-1 in cellular protection following Tafenoquine exposure.

P201 - Preliminary assessment of the developmental toxicity of acid fuchsin using the Frog Embryo Teratogenesis Assay-Xenopus (FETAX)

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<sup>1</sup>Jacksonville State University, Jacksonville, AL, <sup>2</sup>Jacksonville State University, Jacksonville, AL

Acid fuchsin is a dye that is used in materials such as textile fabrics, silk, nylon, wool, and leather. These materials may pollute the water, due to synthetic dye pollution. This pollution can be harmful to the animals in these environments. Amphibians are aquatic organisms that are sensitive to harmful pollutants and there has been a large reduction of amphibian populations in the past 40 years. To determine if acid fuchsin can potentially cause developmental effects to amphibians the Frog Embryo Teratogenesis Assay -Xenopus (FETAX) was used. This assay uses early-stage frog embryos exposed for 4 days (96-hrs) in various concentrations of acid fuchsin. During this time they develop from an early-stage cell blastula to a free-living tadpole. Plastic petri dishes of embryos in 8 MLs of test solution was used as the experimental unit. Four controls and 2 replicates for each concentration were used. Every 24 hours dead embryos were removed, and solutions were renewed. At the end of the four days mortality, malformations and embryo length were recorded. Means, Standard error, probit analysis (for LC50 and EC50(malformation)), ANOVA and Bonferroni's post hoc test were calculated using Systat. The teratogenic potential was calculated using the formula 96-hr LC50/96-hr EC50(malformation). The 96-hr LC50 was >1000 and the 96-hr EC50(malformation) was approximately 120 mg/L. At the high concentrations length was significantly affected. Also, there were severe malformations of spine, eye and gut. These preliminary results indicate that acid fuchsin may be teratogenic and further testing is needed.

#### P202 - Regulation of RYBP by MicroRNAs in Glioblastoma Multiforme

Alex Lee, Mason Linker, Michelle Aguilar-Gaspar, Daniel Stovall *Winthrop*

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Glioblastoma multiforme (GBM) is the most common and lethal tumor of the central nervous system that typically arises from malignant astrocytes. Patient prognosis is especially poor, because GBM tumors either do not respond or become resistant to currently available treatments, including chemotherapy, radiotherapy, and surgery. Therefore, understanding the molecular processes driving GBM progression is imperative for the development of new, more effective therapies. The RING-1 and YY1-binding protein (RYBP) is a multifunctional transcription factor that is downregulated in roughly 50% of GBM tumor patients and functions as a tumor suppressor in multiple different cancer types. However, the mechanism of RYBP silencing in GBM remains poorly understood. We investigated the role of microRNAs (miRNAs) in RYBP silencing. MiRNAs are small regulatory molecules that target complementary mRNA transcripts to inhibit their translation or induce their degradation. Using online prediction software (miRDB.org), we identified multiple putative miRNA binding sites in the RYBP mRNA transcript's 3'-untranslated region (UTR), including sites for miR-9-5p, miR-125b-5p, and miR-128-3p. We transfected U-87 glioma cells and U-118 or T98G glioblastoma cell lines with synthetic inhibitors against these oncogenic miRNAs. After 24 hours, inhibition of miR-9-5p and miR-128-3p markedly increased RYBP protein levels compared to a non-targeting control miRNA inhibitor, as detected by SDS-PAGE and Western blot. These results suggest that miR-9-5p and miR-128-3p may contribute to RYBP downregulation in GBM. Future studies should interrogate whether RYBP serves as a functional target of these miRNAs and determine if there is a direct interaction between the miRNA and RYBP transcript.

#### P203 - Using GIS and LiDAR to identify green salamander (*Aneides aeneus*) habitat in Alabama's Cumberland Plateau

John Matthew Erickson

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The green salamander (*Aneides aeneus*) is a habitat specialist found in rock outcroppings in mesophytic forests in the Appalachian Mountains. This extreme habitat specialization in rugged, forested landscapes makes it difficult and time consuming to locate green salamanders and their habitat with conventional field surveys or visual interpretation of aerial imagery. Smith and Mullins (2022) introduced a new LiDAR-based methodology for efficiently and effectively identifying green salamander habitat in the Virginia portion of the Appalachian Mountains. In this study, we apply Smith and Mullins's (2022) methodology in Bankhead National Forest in Alabama, in the Cumberland Plateau physiographic section of the Appalachian Mountains. First, we processed a 1-meter resolution LiDAR-derived digital elevation model (DEM) in ArcGIS Pro to identify areas of greater than 80% slope as potential green salamander rock outcrop habitats. Second, we are performing field verification of the potential rock outcrops and documenting green salamander populations at the outcrops. Field work is ongoing at the time of abstract submission. In this poster, we will present preliminary results from our first round of field work, with additional field campaigns planned for later in 2023.

#### P204 - Evaluation of cytotoxic and apoptotic effects of several regional plant-based extracts on human squamous cancer cell lines

Nicholas Spader

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The south has always been a place where home remedies and scientific-based medicine go hand in hand. Throughout time, natives and other groups have used homemade salves and ointments for what could have potentially been carcinoma or other forms of skin cancers. Through literature study, certain plants of interest have been identified. Plants used in various assays were either grown in the ABAC greenhouse or collected from local areas. Two extracts of interest have been used in the application of cell cytotoxicity assays, cell proliferation assays, and a cellular invasion assay using skin cancer cell lines. The cytotoxicity assay demonstrated cytoprotective properties when applied to cells. The cell proliferation assay confirmed this observation by showing increased cellular proliferation in cells treated with the extracts. The wound healing assay demonstrated increased cellular invasion into an area of artificial damage. Overall, these extracts have been shown to demonstrate increased cell survival, increased

cell proliferation, and increased cellular invasion when used to treat subcutaneous cancer cells in vitro. In summary, the extracts demonstrated a prosurvival phenotype indicative of enhanced wound healing.

P205 - Geographic life-history variation among Southern Appalachian wood frog populations

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Wood frogs, *Rana sylvatica* are a common spring-breeding amphibian found across North America. Their huge geographic range spans 30+ degrees of latitude, from the southern-most Appalachians to the Arctic Circle. Previous analyses across this large geographic range revealed clear evidence for geographic life-history variation: a smaller in body sizes and egg sizes at high latitudes and low temperatures—a signature of geographic lifehistory variation converse to that predicted by Bergman's rule. In this study, we revisited the ability of temperature (and the Bergman's rule) to explain geographic variation in life history in wood frogs. But in this case, we examined variation among 19 wood frog populations distributed along a 1220-meter elevational gradient in the Southern Appalachians of North Carolina and Tennessee. Given our previous results we predicted that geographic variation would again follow a converse Bergman's rule. That is, we expected populations at the lower, warmer end of the gradient would have larger eggs and larger clutch sizes. However, our results showed no evidence for an elevational gradient in clutch or egg size ( $P>0.1$ ) or an effect of temperature ( $P>0.7$ ). Thus, it appears that the geographic patterns expressed across the latitudinal extent of the geographic range does not necessarily manifest along its elevational extent. While not part of Bergman's hypothesis, we did find strong relationships between wintertime precipitation and egg size ( $\beta=0.6$ ,  $F_{1,7}=12.3$ ,  $P=0.01$ ) and clutch size ( $\beta=0.6$ ,  $F_{1,15}=7.4$ ,  $P=0.02$ ). We suspect these relationships reflect life-history adaptation to geographic variation in hydroperiod; however, the significance of these relationships remains unknown. Ultimately, our study shows that patterns of life-history variation emerging along latitudinal gradients do not necessarily mirror those observed across elevational gradients. Our results are a good illustration of the limitations of broad generalization and underscore the need for detailed life-history data, even for common, well-understood species.

P206 - PAR-1 activation interferes with normal axon development in *C. elegans* through the Rho and PLC pathways

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Thrombin, a key coagulation protein, exerts effects on cells outside the vasculature by activating protease activated receptor-1 (PAR-1), a G-protein coupled receptor found on many cells including neurons. This is important since vascular damage following injury/disease leads to thrombin accumulation in the nervous system. Both *in vivo* and *in vitro* studies show that neuronal PAR-1 activation impacts cell survival and axon formation. Of the several G-protein pathways activated following thrombin-PAR-1 binding, the PLC and Rho pathways, that regulate axonal actin filaments, are the most likely candidates to explain these alterations. While the PLC pathway initiates actin depolymerization, the Rho pathway regulates both depolymerization and polymerization. If PAR-1 activation results in abnormally long axons, there should be no difference in neurons exposed to thrombin versus those exposed to a PLC inhibitor; however, if PAR-1 utilizes the bi-directional Rho pathway axonal differences should be observed. This study's purpose was to examine whether either pathway is involved in the observed axonal defects. To examine axon formation, *Caenorhabditis elegans* (*C. elegans*) eggs were exposed to one of six conditions: control, PAR-1 activation (100  $\mu$ M thrombin), PLC inhibition (100  $\mu$ M U73122), Rho inhibition (250  $\mu$ M Y27632), PAR-1 activation+PLC inhibition, PAR-1 activation+Rho Inhibition. A Kruskal-Wallis rank sum test and pairwise Wilcox post-hoc test (95% CI) showed a significant difference in the percent of defects between the controls and all other groups with those groups having more defects. Furthermore, there was a significant difference in the percent of defects between the PAR-1 activated group and all others, except for that which received simultaneous PLC inhibition. Results indicate that PAR-1 activation causes axonal defects through both the PLC and Rho pathways. If thrombin exposure causes axonal defects during development, then exposure to thrombin due to injury or disease could result in axonal damage and/or prevent axon repair.

P207 - Creation of a body condition index (BCI) for Western Hognose Snakes (*Heterodon nasicus*) for use in captivity and conservation.

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Body Condition indexes (BCI) can be used to determine individual fitness for animals in human care, as well as animals observed in the wild. Western Hognose Snakes (*Heterodon nasicus*) currently represent a significant herpetocultural effort, as well as animals worthy of conservation endeavors, especially in the northern portion of their range. While several field studies have focused on determining the population status of imperiled *H. nasicus* populations, little work has been dedicated to determining individual fitness in the field. Herein we discuss our plan to create a BCI for *H. nasicus* using (I.) animals observed in the field as well as (II.) animals held in captivity. Our hope is to use field-observed animals to gather data that would serve as a baseline for the creation of a BCI that then could be utilized to determine BCI for field-observed and captive specimens. *Heterodon nasicus* held in human care often become obese; the creation of a BCI for this species would aid keepers in determining the fitness of their charges, which ultimately could lead to increased welfare for this species.

P208 - Erosion resistance as a function of species-specific interactions between dune grasses and arbuscular mycorrhizal fungi

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Interactions between vegetation and sediment are critical aspects of dune building and erosion. Recent studies demonstrate the importance of plantspecific traits, such as biomass allocation, as determinants of sand accretion and erosion, influencing dune resilience. Root colonization of mycorrhizal fungi confers beneficial stress management characteristics to host plants, including the reduction of sediment loss from extraradical hyphae. Little is known about the colonization of mycorrhizal fungi between dune grass species and whether this may impact sediment loss with increased storm intensity. Our objective was to investigate potential differences in erosion resistance following repeat salinity stress due to differences in plant traits between two native dune grasses, *Ammophila breviligulata* and *Spartina patens*. We hypothesized that *A. breviligulata* would retain the greatest amount of sediment after an overwash event, due to allocation of resources to belowground growth after disturbance and that salinity would increase erosion due to reduced growth and lower mycorrhizal colonization. Greenhouse grown plants were watered with full strength seawater compared to plants watered with freshwater. Erosion resistance was tested in a flume after being subjected to 6 wave events (60 L water total). Sediment loss, multiple plant traits, and mycorrhizal colonization were measured. *Ammophila* experienced more salt stress, contributing to lower levels of photosynthesis and less biomass accumulation in the treatment group relative to *Spartina*. *Ammophila* had a greater percent infection of fungal structures than *Spartina*, and seawater reduced the presence of mycorrhizal fungi in both species. *Ammophila* exhibited less sediment loss than *Spartina*, and both species experienced reduced erosion with salinity treatment. Above- and belowground biomass had the greatest impact on reducing sediment loss. These results will be used to parameterize a barrier island evolution model and allow for future predictions of storm events on dune erosion.

P209 - Effect of Elderflower Extract on a Parkinson's Disease Model in *C. elegans*

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Parkinson's disease is a neurodegenerative disease that causes a multitude of side effects. Some of which are tremors, partial paralysis, cognitive disabilities and more. In an effort to find ways to aid in the reduction of side effects, *C. elegans* are being used for research in this field. For this research study, the worms used to display Parkinson's were DDP1 and JVR-406. They both display different side effects, one being paralysis, and the other tremors. To prevent the side effects, or improve life span, the worms were placed on plates mixed with elderflower and checked daily for signs of improvement. To measure the improvement the worm's neurons were imaged to see if the elderflower affected the growth of alphasynuclein aggregates on the neurons and the aggregates quantified. Finally, to understand the molecular mechanism, we are performing RNA sequencing on worms with or without elderflower treatment. Together, our work shows that this plant may provide a remedy for patients suffering from Parkinson's disease.

P210 - Spatial and phenological variation of flowering plant species along the Blue Ridge Parkway

Brooke Reutinger, Morgan Gill, Annkatrin Rose, Jennifer Geib

Appalachian State University, Boone, NC

Flowering phenology is an essential factor in understanding plant-pollinator relationships and pollinator life histories. As many pollinator species have experienced population declines, inventory and monitoring programs have become a research priority. Including a record of co-occurring flowering species and their phenology in these inventories may provide valuable insight into the impact of climate change on pollinator populations. Following a 2019 pollinator inventory of the Blue Ridge Parkway, we performed a flowering plant survey in partnership with the National Park Service. From May to October 2022, citizen scientists surveyed 59 sites, recording the presence and phenology of flowering plant species using iNaturalist. Survey sites were located along the length of the parkway, 469 miles through Virginia and North Carolina. Our project aims to understand how flowering plant species assemblage and phenology vary between surveyed sites.

Shifts in flowering phenology have been observed in response to climate change; however, these shifts are not uniform across species. Pollinators depend on access to nectar and pollen resources, and their populations respond to the availability of these resources. To evaluate floral resources at the pollinator inventory sites, we will compare flowering phenology between different sites and plant genera, providing important context to the 2019 Blue Ridge Parkway pollinator inventory data. We will also use QGIS to analyze how elevation, climate, and distance from the nearest population center affect the distribution of introduced flowering plant species. We predict that flowering time at lower latitude sites will be earlier, and sites nearest to population centers will have an increased presence of introduced species. Long-term monitoring of both pollinator and plant populations will provide a better understanding of how to protect biodiversity along the Blue Ridge Parkway.

P211 - Confirming the gene model and genomic neighborhood of *Sin1* in *Drosophila obscura* in comparison to *Drosophila melanogaster*.

David McCaulley, Sara Cline

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The Genomics Education Partnership Pathways Project focuses on annotating genes found in well-characterized signaling and metabolic pathways across the *Drosophila* genus. The current focus is on the insulin signaling pathway, which is well-conserved across animals and critical to growth

and metabolic homeostasis. The long-term goal of the Pathways Project is to analyze how the regulatory regions of genes evolve in the context of their positions within a network. To accomplish this, undergraduate researchers in labs around the world work together to confirm relevant gene models and promoter sequences. Here, we confirmed the gene model for Stress-activated map kinase-interacting protein 1 (Sin1) in the fly *Drosophila obscura* compared with *Drosophila melanogaster*. Sin1 is a variable component of mammalian target of rapamycin (mTOR) complex 2 (mTORC2), which activates Akt, a core serine-threonine kinase in the insulin signaling pathway. We demonstrate the presence of a gained intron in *D. obscura*.

#### P212 - Documenting a remnant old-growth forest at America's newest National Park

Tom Saladyga

*Concord University, Athens, WV*

In 1988, the U.S. Forest Service's Eastern Old-Growth Definition Project developed a generic definition of "old-growth" based on tree age and forest structural characteristics. Today, quantifying and identifying old-growth forests remains a priority for scientists and the public for reasons ranging from carbon storage, watershed protection, and species conservation to aesthetics and recreation. In fall 2022, undergraduate students in an upper-level biogeography course at Concord University collected forest structure and dendrochronological data at the Burnwood area of New River Gorge National Park & Preserve in West Virginia, where few pre-industrial forests remain on the landscape. In three 0.07-hectare forest plots, we identified and measured the diameter of all overstory trees, tallied seedlings and saplings by height class, measured coarse woody debris, and collected tree cores from a sub-sample of overstory trees. In addition, we selectively cored trees across the study area that exhibited old-growth morphological traits such as a twisting trunk, balding or flaking bark, and large, gnarled upper branches. Our results indicate a forest composed of multiple overstory tree species, including *Tsuga canadensis*, *Fagus grandifolia*, *Acer rubrum*, *Nyssa sylvatica*, and four *Quercus* species. Large-diameter trees (> 50 cm at breast height) occurred at a relatively high density (85.7 trees/ha), and 28 of the 50 trees that were cored had an inner-ring year that pre-dated 1900. Fifteen of these trees established before 1800, while five had an inner-ring year in the 1670s. Understory vegetation was sparse with *A. rubrum* dominating the 5–15 cm seedling height class, while only six saplings and four individual shrubs were observed in forest plots. Average coarse woody debris volume was 52.0 m<sup>3</sup>/ha and we observed pit and mound microtopography within plots and across the study area. These findings provide evidence for a previously undescribed mixed mesophytic old-growth forest in the Central Appalachians.

#### P213 - Effect of endophytes on *Anethum graveolens* (dill), ability to attract pollinators through volatile organic compounds.

Emma Kinerson, Michelle Barthet

*Coastal Carolina University, Conway, SC*

Volatile organic compounds (VOC's) are emitted from plants to attract various organisms including pollinators. Terpenes are the most widely released plant VOC's, and specifically monoterpenes play a key role in the interactions between plants and insects. In *Anethum graveolens*, methyl eugenol and limonene, are volatile compounds that attract various pollinators to the plant. Endophytes have been discovered to live symbiotically with various plants, but little is known regarding their role in attracting pollinators to the plant. We propose that the presence of endophytes in *Anethum graveolens* (dill) will increase the attraction of pollinators due to improving synthesis of secondary metabolites such as methyl eugenol and limonene. Dill plants will be grown in sterile and unsterile controlled environments. After maturation of the plants, floral organs will be sampled to examine gene expression of enzymes involved in the synthesis of methyl eugenol and limonene as well as ascertain the presence/ absence of endophytes. Molecular data will be combined with behavioral assessment of insect pollinators to discern the influence of endophytes in pollination.

#### P214 - Understanding how common mycorrhizal networks influence northern red oak seedling (*Quercus rubra*, Fagaceae) growth and establishment

Emza Shackelford-Whitten<sup>1</sup>, Thalia Garoufalidis<sup>1</sup>, Jonathan Horton<sup>2</sup>, Alisa Hove<sup>1</sup>

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Nearly all land plants form symbiotic relationships with mycorrhizal fungi. Ectomycorrhizal fungi (EM) encase plant roots in a fungal mantle, exchanging resources such as carbon, phosphorus, and water. Mycorrhizae have been shown to facilitate relationships among plants in communities through the formation of common mycorrhizal networks (CMNs). To examine the role of CMN access in tree seedling establishment and EM community composition, we established manipulative plots with treatments with varying access to CMN and adult roots (+CMN/+Root, +CMN/Root, -CMN/-Root) at two sites in Buncombe County, NC (UNCA and WWC). At each site, northern red oak (*Quercus rubra*) seedlings were planted in early May and harvested in mid September 2021. To consider the possibility that the nature of CMNs varies among plant communities, plots were established in both white pine (*Pinus strobus*)- and hardwood-dominated forest stands. After seedlings were harvested, we recorded root biomass, shoot biomass, and root:shoot biomass ratio, and estimated EM colonization frequency (percentage of EM colonized root tips per seedling). It is difficult to identify EM fungi by viewing vegetative hyphae found on root tips, so DNA barcoding was used to characterize fungal taxonomic diversity. We amplified and sequenced the DNA barcode internal transcribed spacer (ITS) regions 1-4 for a total of 64 root tips from all four

experimental treatments. Root biomass, shoot biomass, root:shoot ratios, and EM colonization frequency did not differ among CMN manipulation treatments. Average colonization frequency was greater at UNCA than WWC but this may be due to differences in sites rather than effects of the treatments. Analyses of fungal taxonomic diversity and comparisons among treatments are ongoing and will be shared at the meeting.

P215 - Determining anti-cancer potential of proprietary compound against triple-negative breast cancer

Ruben Rosado, Shane Olinger, Dr. Lyndsay V. Rhodes, PhD

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Breast cancer is a disease that will affect 1 in 8 U.S. women over their lifetimes. The disease is caused by a growth of abnormal cells that originate in the tissues of the breast. Treatment options include chemotherapy, hormonal medication, radiation and surgery depending on the stage and subtype of breast cancer. The triple-negative breast cancer (TNBC) subtype accounts for 10-20% of those diagnosed in the U.S every year. TNBC is characterized by the lack of expression of hormone receptors Estrogen Receptor (ER) and Progesterone Receptor (PR), and are non-amplified for the growth factor receptor HER2. As a result, current treatments for breast cancer that target these receptors are ineffective in this subtype, leaving TNBC patients reliant on general chemotherapy, radiation, and surgery. A commercially available, proprietary compound (referred to here as compound E) has been identified as having potential anti-cancer activity. However, most of the evidence is anecdotal and somewhat controversial. Using standard, well-controlled in vitro assays, compound E was tested at a range of concentrations to determine its effects on two breast cancer cell lines (triple-negative: MDA-MB-231; ER positive: MCF7). The compound was found to be effective in concentrations as low as two percent across both cell lines, suggesting effectiveness in both triple-negative and estrogen receptor positive breast cancers. Future research will focus on understanding the mechanism of action of this compound, as well as further application against additional cell lines to determine if these effects hold true.

P216 - Dendrochronology of *Pinus palustris* of the Blackwater Ecological Preserve

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<sup>1</sup>*Old Dominion University, Norfolk, VA*, <sup>2</sup>*United States Department of Agriculture, Washington, DC*

The Blackwater Ecological Preserve (BEP), located in southeastern Virginia, holds significant ecological importance as it contains one of the northern most intact population of *Pinus palustris*, the longleaf pine. Longleaf pine populations were once larger; however, populations have been reduced due to extensive logging, habitat degradation, and fire suppression. For this study, I am creating a master chronology composed of BEP longleaf pine. A chronology is a timeline created by analyzing annual growth rings that naturally occur in wood. This is significant as a master chronology of the BEP longleaf does not currently exist. A chronology is necessary for certain future studies, like those related to growth responses in longleaf and reconstructions of fire and disturbance history. Current restoration efforts for this threatened pine are ongoing, but more research is required to understand its natural history and ecology.

P217 - Socioeconomic factors and the development of cervical cancer in states that have adopted the Affordable Care Act through Medicaid Expansion vs. those that have not

Monica Youssif, Zahra Bahrami-Mostafavi

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**Introduction:** Cervical cancer (CC) is a gynecological cancer that is prevalent among women. CC results from disruptions within the cell's internal regulatory system of cervical cells, resulting in uncontrolled cell growth. Of the 2023 estimated 948,000 total cancer cases among females in the U.S., 1.47%, of those cancer cases are CC, with 1.5% of the total estimated 287,740 female cancer deaths in the U.S. resulting from CC. Many factors contribute to the development of CC within populations. This study examines the demographic factors of age, race, annual income level, and health insurance type, and their relationship with cervical cancer rates.

**Methods:** The State Inpatient Data (SID) of over 4 million patients from Florida was examined. The administrative data were collected by the Healthcare Cost and Utilization Project (HCUP) agency. Data being used was collected during 2009 and 2011 from the state of Florida. To compare the health outcome resulted from ACA implementation, the 2009 and 2011 data from California is being used (results not shown).

**Results:** Using Florida data, binary statistical analysis revealed that the odds of CC is increased in women who are ages 46-65 (OR=9.235, 95% C.I.= 6.448–13.227), Medicaid beneficiaries (OR=4.414, 95% C.I.=3.871–5.033), Black (OR=1.651, 95% C.I.=1.470–1.854), and in the first quartile of annual income (\$1–\$38,999) (OR=71.682, 95% C.I.=1.470–1.854). These results suggest that women of lower socioeconomic status (SES) are more greatly affected by cervical cancer.

**Conclusion:** This study demonstrates that CC shares a significant association with variables of lower SES, including health insurance status, age, race, and income. A contributing factor to these observed trends may include ACA Medicaid expansion in Florida (2010), or the lack thereof. These findings highlight the importance of improving access to healthcare for women of all SES to reduce the prevalence of cervical cancer among these populations.

P218 - Understanding Connections Between Diets of Honeydew Insects and Fire Ants in Longleaf Pine Savannas of North Carolina

Hunter Ivey, Lisa Kelly, Kaitlin Campbell

*University of North Carolina at Pembroke, Pembroke, NC*

Fire ant (*Solenopsis invicta*) diets consist of many different foods, including sugary plant juices obtained from honeydew from honeydew insects, facilitating rapid colony growth and possibly invasion of new sites. Longleaf pine savannas are areas of conservation concern in the coastal region of North Carolina and home to unique and protected flora and fauna. Our previous work at these sites in 2017 indicated fire ant invasion, and ryegrass (*Lolium spp.*) was a significant component of fire ant diets, despite ryegrass not appearing in these sites. We hypothesized that mutualistic relationships between fire ants and honeydew insects are allowing fire ants to access ryegrass and to exploit the savanna ecosystems. We used *ITS2* and *rbcLa* DNA barcodes to compare plant diets of fire ants collected in 2017 with plant diets of honeydew insects collected in 2021. The *ITS2* barcode showed ryegrass in the diets of both honeydew insects and fire ants, while the *rbcLa* barcode showed no ryegrass in the diets of either. Some dietary overlap occurred in other plant foods. In the future, we plan to use Sanger and next-generation sequencing to compare the diets of honeydew insects and fire ants collected during the summer of 2022 to determine if there is a strong connection between their diets. Our data have important implications for the role of invasive species in protected areas. We will share our findings with The Nature Conservancy and other organizations to aid in the conservation of these pine savanna ecosystems.

**P219 - Identifying the Role of Insulin Signaling in Thermal Nociception**

DyQuan Kearney, Andrew Bellemer

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Diabetes impacts more than 1.4 million people every year. There are two common types of diabetes. Type 1 diabetes, which is associated with the absence of insulin signaling, and Type 2 diabetes, which is when there is impaired insulin signaling. Insulin signaling is known to regulate glucose, lipid, and energy homeostasis. When the insulin signaling pathway is impacted it can lead to neuron damage. Neuron damage known as diabetic neuropathy is a common side effect associated with diabetes and can lead to pain or numbness in the legs or feet. PI3-Kinase, mammalian target of Rapamycin (mTOR), and AMP-activated protein kinase are some of the key enzymes that regulate metabolism, and function in the insulin signaling pathway. Investigating how the insulin signaling pathway controls sensitivity to noxious stimuli would provide a better understanding of diabetic neuropathy. This study is designed using *Drosophila melanogaster* as the model organism, to further explore the role of insulin signaling pathways in the regulation of nociception behavior. An experiment was conducted using genetic tools to knockdown PI3-Kinase in larval sensory neurons. The larvae behavior responses to noxious thermal stimuli were then tested. The behavioral responses indicate a reduction in thermal nociception. This data suggests that the PI3-Kinase enzyme is required for normal nociceptive behavior. Future experiments will be designed to investigate other components of the insulin signaling pathway, and how they may affect nociception.

**P220 - Lost Savannas, Flora & Ecology of Shortleaf Pine-Oak Savannas of the Black Belt Prairie Ecoregion of Alabama**

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The Black Belt ecoregion of Alabama is known for its prairies but historically it was a patchwork of open ecosystems, including savannas. Most savannas in the region have succumbed to fire suppression and associated succession to closed canopy forest, conversion to pine plantations, land clearing, or invasive species. The Sumter Farm and Stock Company, a privately owned 11,000-acre farm in Sumter County, Alabama, has been burned yearly for at least 75 years and contains several hundred acres of Post Oak-Blackjack Oak-Shortleaf Pine savanna. Our research will focus on: (1) understanding the composition of these savannas to determine if they are remnants of historical savannas; (2) mapping their location and position relative to 1830s Government Land Office (GLO) maps of nearby prairies; and (3) comparing them to other oak-pine savanna ecosystems of the southeastern U.S. We established twenty Carolina Vegetation Survey plots, with each plot surveyed for total vascular plant species and abundance in summer-fall (September-October); spring data will be collected from the same plots in 2023 (April-May). Each species was evaluated for conservative heliophyte status using the provisional Grassland Fidelity Index (GFI) developed by the Southeastern Grasslands Institute (SGI) and North Carolina Botanical Garden. Currently, 388 species are known from the savanna plots. More than a dozen rare species tracked in Alabama were discovered, with notable species including: *Coelochrachis cylindrica*, *Comandra umbellata*, *Nabalus asper*, *Perideridia americana*, *Solidago tortifolia*, and *Veronicastrum virginicum*. *Muhlenbergia glaberriflora* was documented as a state record for Alabama. A cluster analysis will be performed to compare plots across sites and to other savanna ecosystems. The results of our study will be used to inform restoration of more acres of this rare ecosystem at Sumter Farm & Stock Company and elsewhere in the Black Belt ecoregion of Alabama and Mississippi. P221 - Effect of melittin on Hs578t triple negative breast cancer cells

Kevin Suh

*High Point University, High Point, NC*

Honeybees, *Apis mellifera*, produce and secrete venom which contain peptides, amino acids, sugars, and pheromones. Melittin is a 26-amino acid amphipathic polypeptide and a major component of honeybee venom that constitutes 40-60% of its dry mass. Bee venom has been used as a complementary and alternative therapy for thousands of years by injecting the venom into specific points on a body. According to American Cancer Society, it is estimated that nearly 300,000 women will be diagnosed with breast cancer in the US in 2023. Triple negative breast cancer does not

express markers such as estrogen receptor, progesterone receptor, and epidermal growth factor receptor 2. Therefore, common treatments such as hormone therapy do not work for triple negative breast cancer, and these types are considered more aggressive than other types of breast cancer. In the present study, we investigated the role of melittin in Hs578t triple negative breast cancer cells. Using trypan blue and MTS assays, we found that melittin strongly reduces viability in Hs578t cells. When cells are treated with melittin, we also observed decreased expression of proteins involved in cell proliferation and growth pathways. Finally, we performed RNA sequencing analyses, and Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analyses confirmed our findings and revealed novel targets of melittin in these cells.

**P222 - Effect of removing *Pityopsis nervosa* on the soil bacterial microbiome of a longleaf pine ecosystem.**

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*Kennesaw State University, Kennesaw, GA*

The longleaf pine (*Pinus palustris*) is an integral part of a diverse, endangered ecosystem in the Southeastern United States. Longleaf pines are better equipped to survive strong winds and droughts than other native species and are notable for their dependence on fire for successful regeneration and establishment. The herbaceous vegetation within this ecosystem provides suitable conditions for the longleaf pine by supporting a positive feedback loop with fire. The herbaceous community's importance is well known, but the microbial community associated with these key players is currently understudied. The aim of this study is to investigate the association between soil microorganisms and two important herbaceous species in the longleaf pine ecosystem, *Pityopsis nervosa* and *Andropogon* sp. *Pityopsis nervosa* is the most dominant herbaceous species in the montane longleaf pine restoration sites studied; this forb plays a major role in ecosystem maintenance by serving as fuel for low-intensity fires. Two longleaf pine restoration sites in North Georgia were chosen. Six blocks were randomly set up at each site, and each block contained four randomized subplots with one of the following treatments: control, removal of *Andropogon* sp., removal of *Pityopsis nervosa*, and soil disturbance. DNA was extracted using a commercial extraction kit, measured for concentration, and analyzed to identify bacteria inhabiting the soil samples from each subplot. Preliminary results of these analyses indicated that Acidobacteria and Actinobacteria were among the most abundant phyla of bacteria present in each treatment group. Acidobacteria is a keystone taxon known to regulate biochemical cycles and promote plant growth. Actinobacteria are widely distributed in soil and secrete enzymes involved in degrading chitin and chitosan, play roles in carbon cycling, and degrade plant residues. Additional findings will add to the knowledge of key microbial groups inhabiting the ecosystem and interacting with dominant herbaceous species such as *Pityopsis nervosa*.

**P223 - The Cryptic Cardinal Conundrum: Can Mysterious Molt be Demystified?**

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The Northern Cardinal (*Cardinalis cardinalis*) is a passerine unique amongst other songbirds as it is well documented to undergo a complete preformative molt in which all of its juvenile flight and body feathers can be replaced during their first Fall. Due to the Northern Cardinals unique molting strategy, molt limits, or the presence of a juvenile feather located amongst a tract of formative feathers, may not be present despite a bird being only a few months old, resulting in improper aging. This presents an interesting challenge, especially in southeastern states where complete preformative molt is common. In order to study this phenomenon, we set out to photograph, track, and analyze the molt of northern cardinals at 3 locations in Gwinnett County, Georgia. Standard mist netting techniques were used to capture northern cardinals and all birds were subsequently banded, photographed, and had morphometrics taken. In addition to qualitative study, feather samples were collected and analyzed for barb distance and barbule density to investigate differences among known age groups. Our results show that molt of flight feathers generally progresses in a typical sequence (p1-9 and s1-9) with the majority of visible molt limits being present in the tertials, as these were the last juvenile feathers to be replaced. Quantitative analysis showed a statistical difference in barb distance when adult and juvenile feathers were compared, with adult feathers having statistically lower barb distance than juveniles. This supports the general understanding and published literature on feathers as juvenile feathers are known to be looser in appearance than those of adults. This study seeks to help banders better understand the unusual molt of Northern Cardinals and provide insight into the effectiveness of molt limits for aging these birds.

**P224 - Effects of TiO<sub>2</sub> Nanoparticles on the Upper Thermotolerance of Goldfish (*Carassius auratus*)**

Khalil Bohanan

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Titanium dioxide (TiO<sub>2</sub>) nanoparticles can be found in a wide variety of products such as sunscreen, soap, and even foods such as M&Ms. Furthermore, metal oxide nanoparticles are commonly used in medical and agricultural applications where they may be covered in a polyacrylic acid (PAA) polymer capsule. Due to the widespread use, these nanoparticles often find their way to aquatic environments making them a potential threat to aquatic organisms, which has stimulated a surge in research efforts towards understanding the environmental implications of nanotechnology. This current project was inspired by a previous study aimed at understanding the effects of injected TiO<sub>2</sub> on fish's general immune response to stimuli, which found not only was the overall immune response weakened, but also demonstrated presence of TiO<sub>2</sub> NPs in the gills. Our objective was to see if and how TiO<sub>2</sub> inhibited fish's ability to cope with increasing aquatic temperatures as this could present a dual threat when considering global warming heating bodies of water. To perform the experiment, we subjected four groups of fish separated into control, saline injected, PAA capsule injected, and TiO<sub>2</sub> injected treatments to a critical thermal maximum (CTMax) test and sampled them after loss of

equilibrium (LOE). The LOE temperatures for each fish were recorded and an ANOVA test comparing each treatment group to the control group revealed the TiO<sub>2</sub> injected fish demonstrated a significantly reduced thermal maximum compared to control fish. Molecular and histological analysis of the heart, gills, spleen, kidney, and muscle samples from each fish is ongoing. We hope to discover through what mechanisms TiO<sub>2</sub> harms fish by observation of gene expression and morphological changes in the gills.

P225 - Pit-stop ecology: frugivory and seed dispersal by thrushes passing through southern Appalachia

Nicholas Smith

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Bird banding stations all over the world contribute to our understanding of the migratory phase in a songbird's annual cycle. Morphology data collected through the banding process are extremely valuable on their own, but by recording some data that is typically overlooked, researchers can contextualize certain phenological events. This approach helps paint a more complete picture of two often understudied components of this system: fall migration and fruiting phenology. In Fall 2021, we collected fecal samples provided by thrushes (family Turdidae) during migration banding efforts at Blue Ridge Bird Observatory's Big Bald Banding Station (Unicoi County, TN). Members of this family were chosen due to their documented consumption of fleshy fruits and their historical abundance at this site. 12 species of fleshy fruited plants were consumed, and their intact seeds were dispersed by three different thrush species. Swainson's Thrushes were the most frequently captured thrush (n = 363) and consumed/dispersed 11 of the 12 plant species documented. Wood Thrush samples (n = 33) contained only 2 different plant species but had the highest overall proportion with seeds (in ~28% of samples). Gray-cheeked Thrushes were the least abundant (n = 10) and dispersed one plant species. Despite evidence that Swainson's Thrushes which were more than one year old (after hatch year; AHY) passed through this site an average of 4 days earlier than hatch-year (HY) birds, there were no significant differences in rates of general frugivory and seed-dispersal between these groups. There was also no significant relationship between the date of capture and rates of frugivory and seed-dispersal. It appears that the variety of fruit resources lead to nearly continuous availability over the banding season. Our work highlights the nuances of Fall migration and how frugivorous songbirds utilize resources during this critical and understudied phase of their life cycle.

P226 - Species Distribution Modeling for the Endangered Houston Toad *Anaxyrus houstonensis* (Bufonidae)

Travis Rhodes, Kinsey Holbrook, Christopher Randle

*Sam Houston State University, Huntsville, TX*

The Houston toad, *Anaxyrus houstonensis* (previously classified as the species *Bufo houstonensis*) has been classified as endangered since the 1970s, with an worldwide estimated population of 3,000–4,000 adults. *Anaxyrus houstonensis* is native to south-central Texas, with most populations occurring in the Bastrop county. *Anaxyrus houstonensis*' native habitat consists includes loose, deep, sandy soil supporting loblolly pine and/or mixed post oak forest and native bunch grasses. *Anaxyrus houstonensis* requires still or flowing waters for breeding. To predict other areas of highest habitat suitability, we trained three species distribution models using occurrence data from GBIF and iDigBio and environmental factors from the home range. Because of the dearth of occurrence points with GIS data, many occurrence points were geolocated using the GeoLocate platform. Model prediction accuracy was assessed using the Area Under the receiver operating characteristic Curve (AUC), and ensemble predictions were constructed as the AUC weighted average of model predictions. Environmental conditions most important for predicting habitat suitability were identified as well as areas most suitable for introduction and rehabilitation of this endangered species.

P227 - Fish and wading bird diversity in relation to eelgrass loss from eutrophication in Barnegat Bay

Erin Foreman, Marielle Postava-Davignon

*Virginia Wesleyan University, Virginia Beach, VA*

The Barnegat Bay of New Jersey has been heavily impacted by eutrophication, as seen by increased overloading of nutrients and algal blooms. Eelgrass, *Zostera marina*, is known to be affected by eutrophication and has shown decreased populations in this area. The nursery function of *Z. marina* in estuaries benefits brackish and marine fish species, and therefore impacts the availability of this prey for wading birds such as herons and egrets. This study encompassed the broader trophic impacts of eutrophication by comparing the diversity of fish and wading birds at sites in North, Central, and South Barnegat Bay in relation to *Z. marina* meadows using seining and observational methods from June 1 - July 15, 2021. It was anticipated that diversity indices would be higher for both fish and birds where these meadows were present. A higher abundance of both fish and birds, as well as a higher diversity of birds, was found at sites with *Z. marina*. Conclusions cannot be drawn on this evidence, however, since these differences were not statistically significant. Future studies may explore other data analysis methods and collect larger quantities of data to produce more conclusive results that can be used when considering plans to mitigate eutrophication and its effects.

P228 - Using RADseq to determine relationships among longleaf pine (*Pinus palustris* Mill.) populations at the northern edge of its range

Lauren Heyd<sup>1</sup>, Nicholas Flanders<sup>1</sup>, Peter Schafran<sup>2</sup>

nucleotide polymorphism loci. Results from analyses of this data will elucidate how many genetically distinct longleaf pine populations reside at BEP and how these stands may be related to populations native to southeastern VA.

P229 - Shedding some light on artificial illumination's effects on mammal diel activity patterns

Sam Williams, Emily Prince, Lisa McDonald

*Lander University, Greenwood, SC*

Artificial light at night (ALAN) is an important form of anthropogenic pollution in urban habitats. High ALAN affects vigilance and foraging behavior in mammals. However, the effects of ALAN on mammal diel activity patterns are unclear. We hypothesized that ALAN may reduce the distinction between diurnal and nocturnal activity, resulting in a shift to more cathestral activity patterns. To test ALAN's effects on nocturnal and diurnal mammals, we used data collected from a network of 26 motion-activated camera traps at 6 sites across Greenwood, Laurens, and Pickens counties in Upstate South Carolina. A GIS layer for light pollution was placed over a map of the camera traps using QGIS. We measured the radiance value at each camera to categorize each station as low or high ALAN. We compared the number of diurnal and nocturnal observations of raccoons, white-tailed deer, and gray squirrels between sites with low and high ALAN. Raccoons are typically nocturnal, which we observed at the low ALAN stations, but a larger proportion of sightings were during the day at the high ALAN stations. Deer exhibited typical variable activity patterns in low ALAN stations, but the proportion of nighttime sightings was higher at high ALAN stations. Gray squirrels followed the expected diurnal activity pattern at low ALAN stations, however the proportion of sightings in the day was even higher at high ALAN stations. This suggests that our hypothesis was supported for raccoons and deer, but not for gray squirrels. The squirrels may be more active during the day because high ALAN makes them more vulnerable to nocturnal predators. Overall, heightened ALAN levels do shift mammal diel activity patterns, but the direction of the shift is species specific.

P230 - Mind the Gap: A Silvicultural Tool for Hemlock Restoration

Lauren Gonzalez

*North Carolina State University, NC*

The hemlock woolly adelgid (HWA), *Adelges tsugae*, is an invasive and devastating pest of hemlock trees in eastern North America. Both eastern hemlock (*Tsuga canadensis*) and Carolina hemlock (*Tsuga caroliniana*), ecologically important conifer species in the Southern Appalachian Mountains, have experienced great decline and mortality as a result of HWA introduction. State and federal agencies in North Carolina have been attempting to preserve hemlocks in the region through chemical treatments, genetic research, and biological controls since the early 2000s with limited impacts. In recent years studies have shown that silvicultural techniques such as thinning through the creation of gap treatments, that increase light and soil moisture, may bolster existing strategies by improving long-term tree health and survival. Information is still needed on the optimal field placement of hemlock seedlings and the relative benefit of controlling for competing vegetation and HWA in those environments. Evaluating the effects of canopy condition, competition control, and their integration with chemical and biological pest management on the establishment and early growth of both eastern and Carolina hemlock will provide forest managers with an optimal silvicultural approach for replanting hemlocks for restoration.

P231 - Temporal and vertical layering of animals in forested ecosystems in northwest Tennessee

Taylor Underwood, H. Dawn Wilkins

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Increases in vertical layering in ecosystems tends to correlate to higher diversity of animals due to increases in available niches. By placing feeders at different heights in the environment and noting whether the photo was taken during the day or at night, our goal was to observe vertical stratification and temporal layering of animals in forested ecosystems. We mounted motion sensing cameras 1 m and 3 m above the ground. Directly in front of the cameras, we placed suet feeders. We observed some temporal and vertical layering between the animal communities. Birds tend to visit the 3 m feeders during day, probably because birds would be searching for food sources by flying through the environment. Some mammals,

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Restoration of longleaf pine (*Pinus palustris* Mill.) communities at the northern edge of their range in southeastern Virginia (VA) and northeastern North Carolina is limited by the availability of seeds originating from trees native to the region. The provenance of mature stands of longleaf pine at Blackwater Ecological Preserve (BEP) in Isle of Wight Co., VA is unknown. Such knowledge could either expand options for seed collection if the trees are native, or give valuable context for studies of longleaf pine survival and reproduction at BEP if the trees were sourced from elsewhere in the range. In the latter case, BEP studies could contribute to knowledge on the role of provenance in longleaf pine performance at the northern edge of its range. We collected needle tissue samples from 156 longleaf pine individuals at 6 sites across southeastern VA, including BEP, and 3 sites further south. We used RADseq to sequence these samples at a depth of 35 million reads per individual and obtain genotypes at single

such as raccoons and opossums, were more likely to forage on the 1 m feeders during the night, probably because mammals would be searching for food as they are walking through the environment. There was intermediate overlap in species composition between the two communities suggesting some vertical layering is occurring. Low similarity in the species composition of the animal community between the nocturnal and diurnal communities supports temporal layering in resource use between the two groups. To increase our understanding of vertical layering, we would like expand our study by setting up motion sensing cameras at 5 m from the ground.

P232 - Tree Seedling Success at Stream Restoration Planting Sites in Rockingham County, Virginia

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Landowners are increasingly recognizing the importance of restoring riparian land previously degraded by agricultural use. Programs like CREP (Conservation Reserve Enhancement Program), provide an incentive for farmers by offering them tax credit and reimbursement for conservation costs. Incorporating native tree and shrub species that have high value for wildlife is important for long-term ecosystem health and restoration. However, the relative success of many species that are used in these restoration management plans is unknown. We quantified planted woody species growth and survival on two farms, FARM\_19 and FARM\_21, enrolled in CREP for one and four years in Rockingham County, Virginia. After planting, each individual received a unique numbered tag. Height, basal diameter, and survival were initially recorded in the spring and at the end of every growing season to determine the overall success of 24 different species within and between sites. After four years, *Prunus serotina*, *Robinia pseudoacacia*, and *Cornus americana* had the greatest survival (> 88%) and growth rates whereas *Betula nigra* had the lowest survival (< 8%), likely due to heavy deer browse and microclimate conditions. Overall survival of the 539 plantings at FARM\_19 was 67.3% after four years. The overall survival of the 444 plantings at FARM\_21 was 84% after one year. The tallest tree at FARM\_19 was *Robinia pseudoacacia*, which reached 5.2 meters after 4 years while the tallest tree at FARM\_21 was *Diospyros virginiana* at 2.3 meters after 1 year. These results will help inform future restoration projects on which species are most successful in riparian plantings within a range of deer browse pressure.

P233 - Assessing the effect of cattle grazing intensities on mixed-grass prairie arthropod communities.

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Cattle and arthropods are both important herbivores in grassland ecosystems. However, little is known about their interactions and how arthropods are affected by cattle grazing intensity. This is important to investigate considering that there have been widespread declines in arthropod biomass, abundance, and number of species across trophic levels, as well as a rise in cattle grazing intensity with increasing human influence. The effect that cattle grazing has on arthropods may be a result of changes in ecosystem function which can be measured by assessing cattle grazing intensity on plant functional traits (e.g., morphological, physiological, or phenological traits that impact fitness). Therefore, we investigated how cattle grazing intensity alters arthropod communities, and further assessed how community-weighted traits (traits weighted by community abundance) changed across cattle grazing intensities.

We collected arthropod and plant traits in an established grazing experiment in the mixed-grass prairie. Arthropods were collected using a modified leaf-blower and transect-sweep net in three cattle grazing intensities (none, low, high). Following collection, all arthropods were stored in a freezer until identification, then dried and weighed. Plant traits (including specific leaf area: SLA and height) were measured for the top 90% of species based on plant community cover across the site. These data were combined with plant species composition data to calculate community-weighted means of SLA and height to assess how each respond on a community level to cattle grazing treatments. We found that arthropod herbivore richness and abundance decrease as cattle grazing increases. Moreover, community-weighted means of SLA and height shows how cattle grazing may indirectly affect arthropods by changing plant community structure in a way that negatively impacts arthropod communities. Improving our understanding will help to generate science-based adaptive management practices to avoid overgrazing, deterioration of arthropod communities, and deterioration of grassland ecosystems.

P234 - Paving the way for Effective Plant Conservation

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The Southeastern Plant Conservation Alliance (SE PCA) is a diverse partnership that bridges gaps between local and national efforts while collaborating to restore and prevent the loss of plant diversity. This is achieved by building capacity, facilitating novel partnerships, and leveraging shared resources to stimulate collective success in our region. To date, the formation of the SE PCA has allowed partners to leverage funding and conservation actions to address the following actions: Advocating for plants. With other nationally recognized groups, we urged the Biden administration to prioritize the conservation of native plants and ecosystems. We also developed a free-access information sheet on regional conservation needs, goals and activities. Ex Situ Gap Analysis The SE PCA partnered with Botanic Gardens Conservation International - U.S to conduct an ex situ gap analysis to evaluate regional ex-situ collections of priority species and identify gaps needing to be filled to meet conservation needs for priority species. Improving Recovery Outcomes for the Endangered Species Act With funding and collaboration from USFWS, we have defined 13 high-priority federally listed species and are implementing pilot projects for 5 in 2021- 2022. Primary objectives for this innovative project also include on-the-ground conservation action (including research and management), education & outreach to partners and landowners, support for local Plant Conservation Alliances, the promotion of public and private land partnerships, and facilitation of working groups and

workshops. List of Regional Species of Greatest Conservation Need (RSGCN) We are currently working with NatureServe, Terwilliger Consulting, and the Southeast Association of Fish & Wildlife Agencies to create the nation's first Regional Species of Greatest Conservation Need (RSGCN) for plants, which directly enhance data, consistency, capacity, and awareness for plant conservation - both during the development process and as a result of associated research, restoration, regulatory, outreach, and leadership efforts.

#### P235 - The effects of human disturbance on mammal diel activity: Does trophic level matter?

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Globally, humans are the most successful predators of terrestrial mammals. Although human exploitation affects all trophic levels, it is more intense for large predators than for mesopredators or herbivores. To avoid the threat posed by humans, mammals may avoid habitats with high human disturbances, but they may also temporally avoid humans by changing their diel activity patterns. We investigated the effect of human disturbance on the diel activity of mammals of different trophic levels. We hypothesized mammals would be more nocturnal in open habitats because they are more visible to humans. We also hypothesized that mammals would be more nocturnal in sites with human disturbance, but the disturbance would affect large predators the most, herbivores the least, and have an intermediate impact on mesopredators. We tested our hypotheses by deploying camera traps at sites across Upstate South Carolina. Each camera was classified by habitat type based on canopy cover and human-disturbance level based on the frequency of images of human activity. Each picture was labeled with species and time of day. We compared the proportion of nocturnal observations at cameras with different disturbance levels and at cameras in different habitats. As hypothesized, we found more nocturnal observations at cameras in more open habitats for all trophic levels. However, we found that while both large predators and herbivores were more nocturnal in areas with higher human disturbance, mesopredators were more diurnal. Our results suggest that when they are more visible to humans, mammals will be more nocturnal. However, members of different trophic levels show different responses to human disturbance. Although large predators and herbivores temporally avoid humans, mesopredators are more tolerant of human disturbances.

#### P236 - Urban legacies: Exploring how demographics, canopy cover, and land use affect ground arthropod biomass in the city

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Insects are essential for the mediation of ecosystem function through nutrient cycling, pollination, and providing key food sources both in urban and non-urban spaces. Patterns in non-urban systems suggest that insect biomass is positively correlated with tree canopy cover. Additionally, in urban areas several studies demonstrate a link between current tree canopy cover and historical segregationist policies. We will examine the relationships between arthropod biomass, tree canopy cover, land use, and historical demographic patterns in Raleigh, NC. We hypothesize that areas which have been historically disadvantaged will have less tree canopy cover and less insect biomass when compared to areas that have not been disadvantaged. We also expect that land use will play a significant role in predicting insect biomass in both historically disadvantaged and advantaged areas. To test this prediction, we collected insects across 30 locations in Raleigh, NC, in five different land uses (greenways, parks, commercial areas, residential areas, and industrial areas) using pitfall traps in the summer of 2021. We then sorted and weighed the insects and analyzed insect biomass in relation to National Land Cover (NLCD) tree canopy cover data. Demographic data was sourced from the U.S. Census 2020 and 1960 datasets. Preliminary results show that no relationship exists between racial identity and tree canopy cover data both currently or historically in Raleigh. Furthermore, land use type, not canopy cover was most predictive of ground arthropod biomass, such that parks had less arthropod biomass than other land use types in early summer 2021. These results suggest that canopy cover may not be the only factor to consider when designing cities that support a quality of life for both the humans and arthropods that live in them. Ultimately, we hope our work will provide urban planners with new tools to design more just and ecologically resilient cities.

#### P237 - Sociability, Seasonality, and Frequency of Northern Cardinals at a Bird Bath

Olivia Griffin, James Ferrari

*Wesleyan College, Macon, GA*

Northern Cardinals (*Cardinalis cardinalis*) are often the gateway into the bird world due to their colorful plumage as well as their prevalence at feeders, gardens, and bird baths. Taking advantage of this, we measured the frequency of Northern Cardinal visits to an artificial water source and examined their social behavior with each other and other songbirds in Macon, Georgia. A game camera was used to take a photo of a bird bath every minute from sunrise to sunset from September 1, 2021 through April 30, 2022. Bird visits were categorized as either perch, bathe, or drink. The Phi coefficient, a measure of association between cardinals and other bird species, was also calculated for each month. Based on more than 165,000 photographs, Northern Cardinals were the species with the highest number of detections (2,107). Most cardinal visits to water were either solo or with a conspecific (91%), while 0.3% of visits were with both a conspecific and a heterospecific, and 8.7% of visits were with a heterospecific. In terms of hourly visitation patterns, cardinals were more prevalent during the early afternoon to evening compared to the morning. There was a considerable increase in Northern Cardinal visitations during November, followed by a drop in January and then a slow increase in spring. According to the Phi coefficient, cardinals were most sociable with heterospecifics during November and December. Similarly, the rates of solo visitation were lowest in December and January. Northern Cardinals are more likely to be social with birds of other species rather than each other: shared heterospecific visits (i.e., with members of other species) were higher than shared conspecific visits (1.7%). Northern Cardinals may be more sociable in the winter due to the predation risks associated with bathing and drinking.

P238 - Scaling Global Urban Greenspace

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*University of Kentucky, Lexington, KY*

Cities are novel ecosystems. Understanding the complex interactions between urban abiotic and biotic attributes is important for managing future urban expansion. Using urban scaling theory (UST) combined with global datasets raises the prospect of deriving predictive rules for scaling urban greenspace with city size. Aspects of the city, such as greenspace, are influenced by several social and ecological factors that scale with a city's size. Empirical data from European cities showed superlinear scaling, in which greenspace increased disproportionately with city size. However, the comprehension of *global* greenspace scaling still needs to be understood. A macroecological perspective using a scaling approach is needed to understand how urban environments function on broader spatial scales and across more countries. We aim to develop predictive measures of how urban greenspace scales with various attributes by answering the following questions: i) how does urban area scale with population and therefore, density? ii) How does greenspace scale with city size and density globally? iii) Is there a trade-off between green space and density across cities on a global scale? iv) How does urban species richness scale with greenspace using community science data? Our study provides a first look at global greenspace scaling, providing statistical information about how greenspace is changing with city size globally. Our results show that while UST predicts density to increase with city size, this is different for most countries globally. Our next steps are to explain the relationship between urban greenspace, population density, city size, and urban biodiversity. Furthermore, our global macroecological perspective provides novel insight into cities as functional ecosystems. As the number of inhabitants continues to grow globally, this research is imperative to understanding the city as a living ecosystem, acting differently around the world.

P239 - The Effects of Caging to Prevent Herbivory on the Pollination Ecology of the Endangered Plant *Astragalus bibullatus*

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*Astragalus bibullatus* is an endangered perennial herb endemic to the limestone cedar glades of Rutherford County, Tennessee. The species has been out-planted into new sites, but mammal herbivory has been a problem during reintroduction. To prevent herbivory, 1.2-m-tall open-topped metal cages with mesh openings of 0.5 cm<sup>2</sup> (hereafter, small mesh cages) were placed around the out-plants. We observed in 2021 that pollinators would not enter the cages, either through the mesh or from the top. The goal of our study was to test if increasing mesh size would allow pollinators to enter the cage while also preventing herbivore access to the plants. A field experiment was conducted in spring 2022 utilizing three plot types with 10 replicates each: small mesh cages, large mesh cages (38.7 cm<sup>2</sup> openings) and uncaged plants. Each plot contained 1-3 plants of *A. bibullatus*. The size of the mesh in large cages was selected based on the largest insect visiting uncaged plants. Pollinator visitation and fruit and seed production were recorded, and trail cameras captured images of herbivores visiting plants. We recorded eight species of insects visiting flowers and the large mesh size allowed all pollinators to enter the cage. No fruit or seeds were produced from plants in small mesh cages; however plants in large mesh cages and uncaged plants produced similar numbers of fruits and seeds. No signs of herbivory were observed on any caged or uncaged plants despite common herbivores such as white-tailed deer (*Odocoileus virginianus*) and eastern cottontail rabbits (*Sylvilagus floridanus*) being present. The experiment will be repeated in 2023 using sites known to have high rates of herbivory. Based on the effect of caging on pollination, we recommend that large mesh cages be used in the reintroduction of *A. bibullatus* to allow effective pollination and prevent herbivory. P240 - A Short-Term Analysis of Bank Erosion Along Shades Creek in Homewood, Alabama

Anna Martin, Elizabeth Dobbins

*Samford University, Birmingham, AL*

Increasing urbanization within watersheds reduces groundwater recharge, increases stormwater runoff, accelerates and intensifies stream discharge rates, scours banks, and encourages sediment to enter waterways, resulting in intensified flooding. To compensate, early stream management techniques included the deepening and straightening of streams. This channelization of waterways, designed to protect human developments from flooding, resulted in channel incision, bank erosion, riparian area flooding further downstream, and loss of biodiversity. Loss of riparian vegetation, especially trees, can accelerate erosion and bank slumping. Shades Creek, an impaired stream that flows through urban and residential areas of Birmingham, Alabama was channelized over 60 years ago and suffers from intense local erosion. To slow bank loss, localized mitigation efforts, including in-stream modifications (rock vanes and step banks) and revegetation of banks, were performed in 2010. Despite these modifications, we hypothesized the downstream creek bank would exhibit appreciable erosion. We studied two 100-meter stretches of Shades Creek bank along a suburban greenway and characterized the tree communities and bank lip stability from March to September 2022. During this study period there was an average bank lip loss of 2.2 cm/month. Although the mean rainfall (13.9 cm/month) was higher than the mean recorded for previous 30 years (12.0 cm/month), climate change will increase summer rainfall and rain intensity, which suggestss that these erosion rates would continue.

P241 - Effect of removing *Andropogon* sp. on the soil bacterial microbiome of a longleaf pine ecosystem.

Van Par, Dylan Bennett, Isabella Vahle, Olivia Walker, Rylee Shaw, Paula Jackson

*Kennesaw State University, Kennesaw, GA*

The longleaf pine (*Pinus palustris*), native to the Southeastern United States, is part of an endangered, pyrophytic ecosystem. Longleaf pines play a significant role in the environment and are an important economic resource due to timber quality. This ecosystem relies on frequent, low-intensity fires facilitated by the herbaceous community. The importance of these herbaceous species in ecosystem maintenance is understood, but little is known about the microorganisms associated with these species. The aim of this study is to investigate the effect of the removal of two dominant herbaceous species, *Pityopsis nervosa* and *Andropogon* sp., on the bacterial soil microbiome of an area under restoration for the longleaf pine in the Piedmont region of Georgia. *Andropogon* sp., the second-most dominant grass species in the longleaf pine restoration sites studied, is most notable for its ability to contribute to forest fires, which are necessary to the regeneration of the longleaf pine. Two longleaf pine restoration sites were chosen for this study. At each, six blocks were randomly set up containing four randomized subplots with one of the following treatments: control, removal of *Andropogon* sp., removal of *Pityopsis nervosa*, and soil disturbance. Soil samples were taken from each subplot in each block, and DNA was extracted from soil using commercially available kits. Further genetic analysis was performed to identify the bacteria that inhabit the soil microbiome in each treatment. Preliminary results include the presence of Proteobacteria and Planctomycetota. Proteobacteria are well known for their role in carbon, nitrogen, and sulfur cycling in soil. Planctomycetes are a phylum of terrestrial bacteria that inhabit soil microbial communities and are known for their ability to go through anammox in the nitrogen cycle. Additional results will identify bacterial and fungal microorganisms in this ecosystem and the key microbial groups interacting with dominant vegetation such as *Andropogon* sp.

**P242 - The ecological effects of redlining: How is bird biodiversity affected by differences in modern urban development?**

**Jennifer Kovacs**

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Eighty years ago, the federal Home Owners' Loan Corporation created "Residential Security" maps for over 100 major American cities. These maps graded neighborhoods on an A-D scale with "A" neighborhoods being desirable for investment and "D" neighborhoods being deemed hazardous. This redlining practice denied the minority residents in these "hazardous" areas access to capital investment. While outlawed in the 1970s, the long-term impacts of redlining can be seen across urban areas in the US today. For this project, we are asking how differences in urban development due to redlining affect bird biodiversity? We used birdwatching community science data collected by Project eBird to analyze bird counts from locations within urban counties that have been historically redlined. Our analyses focused on major metropolitan cities in the Atlantic Flyway between 1990 and 2020. We used R to download, clean, and analyze raw citizen science data to provide a measure of overall bird biodiversity, measured by species richness and the Shannon's Diversity Index. Using ArcGIS to overlay historical redlining maps, we analyzed the relationship between the redlining grades and the recent patterns in bird diversity. We compiled much of this data during a one-day Science Sprint held in April 2022, in which ASC students learned and used R and GIS to clean up and analyze the bird and redlining data for a single city and year. We then compiled data from all the completed cities to gain a better idea of the large-scale regional effects of redlining on biodiversity. **P243 - Growth and Survival of Salamanders Exposed to Road Salt and Glyphosate-Based Herbicide**

**Jerica Eaton, Jarrett Johnson**

*Western Kentucky University, Bowling Green, KY*

Amphibian populations are threatened by increasing levels of environmental pollution resulting from human activities. Areas with mixed land use that include agriculture and urban development experience increased contamination from surface herbicide and road deicer application. Upon entering water sources, these chemicals have the capacity to disrupt the natural dynamics of aquatic ecosystems. This study aimed to evaluate the effects of these contaminants on amphibian populations. We used the spotted salamander (*Ambystoma maculatum*) as a model, rearing them from hatching to metamorphosis. We subjected each of the treatment groups to varying levels of herbicide and salt concentrations. Length and mass measurements were taken every 10 days. Salt concentrations had a more significant effect on larval lengths at metamorphosis than the herbicide concentrations. Salt concentrations had a stronger effect on larval mass at metamorphosis than the herbicide concentrations. Higher salt concentrations appeared to delay time to metamorphosis while there was a nonsignificant trend towards higher herbicide concentrations leading to earlier metamorphosis. Larval growth was reduced at higher salt levels while there was a nonsignificant trend towards lower growth at elevated herbicide concentrations. Our results confirm previous literature regarding the harmful impact of road salt and glyphosate-based herbicide. However, the effect of herbicide concentration on amphibian populations did not seem to be as strong as the effect of road salt. The results of this study are significant in that they give insight into how anthropogenic land use may be contributing to the global decline in amphibian populations.

**P244 - Fire mediated forest structure's influence on dispersal dynamics of wind dispersed warm season bunchgrasses**

**Andrew Zachman**

*University of North Carolina Chapel Hill, Chapel Hill, NC*

The Longleaf pine (*Pinus palustris*) ecosystem currently occupies just 2.2% of its historical range of 37 million hectares. This loss is primarily attributed to overexploitation, conversion to more lucrative Loblolly pine (*Pinus taeda*), and fire suppression. Land managers across the southeastern United States have approached restoring the longleaf ecosystem through the reintroduction of frequent, low intensity fires. Establishing these fire regimes relies heavily on a robust understory of warm-season bunchgrasses which are the primary fuels responsible for carrying fire. The ability of fire to shape forest structure in longleaf pine systems has been well studied. However, the dynamics of how fire mediated forest structure influences the dispersal capabilities of wind-dispersed bunchgrasses is undocumented. Terrestrial LiDAR scans will be used to derive values of vegetation porosity and height. These values will be related to an in-situ wind dispersal experiment using splitbeard bluestem (*Andropogon ternarius*), a warm season bunchgrass. Results will demonstrate the influence of fire mediated forest structure on the dispersal dynamics of wind dispersed bunchgrasses

essential to frequent low intensity fire regimes. This data can be used by land managers to inform efforts to establish groundcover, particularly in areas with minimal seed bank response that rely on seed dispersal from outside sources. This research also provides a framework for gathering high resolution forest metrics through terrestrial LiDAR which can be adapted to diverse forestry applications.

P245 - The Impact of Non-Native Plants on the Abundance of Native Vegetation

Luke Reed, Jacob Hilgemann, Kiyoshi Sasaki

*Wintrop University, Rock Hill, SC*

Forests in urban areas are especially prone to invasion because of many human activities that modulate the introduction and spread of non-native species. These non-native plants may be detrimental to urban forest plant communities. However, few studies have assessed the effects of nonnative plants on urban forests. Our goal for this study was to determine how non-native plant species impact plant communities. We established 733, 5-m radius plots throughout a 28.3 ha Wintrop Woods located in the city of Rock Hill, SC. In each plot, we identified and quantified the abundance of herbs, lianas, shrubs, and tree species. Non-native species were more abundant than native species in the understory, while non-native species were absent in the overstory. The overall abundance of native species decreased as the abundance of non-native species increased. Also, the abundance of native tree seedlings declined with the increasing abundance of non-native species. Although native trees dominate the current overstory, the reduction of native tree seedlings suggests a potential failure in the regeneration of native canopy trees.

P246 - Arboreal bioindicators: Assessment of canopy lichen and microhabitat conditions along an urbanization gradient

Annalaura Pannill, Joshua Lyon, Diane Styers

*Western Carolina University, Cullowhee, NC*

Forest ecosystems in urban areas are exposed to many sources of localized air pollution due to increased human presence. Atmospheric deposition of pollutants can affect various processes within a forest community, from individual plant to ecosystem levels, and exposure responses are often complex and not well understood. Forest health depends on numerous ecosystem processes that balance tree growth, mortality, and regeneration. When these processes become impaired and the system is stressed, observations of pests, disease, and exotic plant species may increase, while the incidence and abundance of sensitive lichen communities decrease. Therefore, analysis of bioindicators of these and other processes serve as useful tools in forest ecosystem health assessments. A bioindicator is an organism that can be used to indicate an alteration in the environment, and these changes can be physical or chemical. Lichens are a good bioindicator because their anatomy and/or physiology (e.g., high surface area, lack of roots, rapid water absorption) are such that they are sensitive to atmospheric deposition. Since lichens are known to be very sensitive indicators of ecosystem health, in situ tissue samples were collected and analyzed for elemental concentrations. Lichens were collected at tree base, mid-canopy, and upper canopy when accessible. Microhabitat variables were also sampled concurrently including relative humidity, light, and temperature. Analyses are currently in progress, but preliminary data show the most rural site has the highest atmospheric sulfur, and the moderately rural site has the least with the most urban site being in the middle. Next steps will include completing the lichen tissue analysis and evaluating various environmental variables such as light, temperature, aspect, and elevation.

P247 - Exploring the role of UAS in natural resource management

Peggy Mullin

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The use of unoccupied aerial systems (UAS), or drones, to augment natural resource management is an emerging sector of great interest for its efficiency and product quality. The Carolina Drone Lab (CARDNL) within the University of North Carolina at Chapel Hill pursues research, both domestically and internationally, using drones to answer environmental research and management questions. In November 2022, CARDNL flew a UAS at over 14,000 feet in altitude over high-elevation wetland ecosystems, or páramos, in the Andes Mountains of Ecuador. Sensors mounted on the UAS captured high resolution true-color, thermal and multispectral imagery. There is a great amount of local interest in the preservation and restoration of these freshwater ecosystems, which provide water to the surrounding provinces, towns and cities. The data collected as part of this research will be used to refine hydrological modeling of freshwater movement throughout the mountains and the surrounding provinces. Delineation of high-altitude habitat, coupled with a review of the field methodology for uniquely high-elevation UAS operations, will demonstrate suitability of the UAS and associated sensors for high-elevation operations. Ground-truthing the drone data with on-the-ground measurements will provide an assessment of sensors to perform wetland land cover delineation. Demonstrating the use of UAS to map these unique ecosystems contributes to the conversation regarding the role of UAS in augmenting resource management field campaigns.

P248 - Documenting the Fire History of Pilot Mountain State Park

John Guglielmetti, Owen Hunter, Dane Kuppinger

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Fire impacts the biodiversity, structure, and function of ecosystems across the Southeastern United States. While large scale fires are uncommon in the piedmont, they do play a significant role in dry forest communities as evident by the fire-dependent species present. The fire history for this region has not been extensively studied however, which limits our understanding of how these forests have been shaped by fire over time. The adoption of national fire suppression policies from 1930-1980 resulted in decreased fire frequency across the southeastern United States and understanding the impact of this policy is important as land managers had begun reintroducing prescribed fire to these landscapes. This research builds upon a previously published fire history for Pilot Mountain State park; extending that history back to the early 1800's and increasing the spatial extent covered by the scar history. This study added fourteen samples containing sixteen fire scars to the Pilot Mountain fire history record. The combined fire history for the park now documents 38 fires; 18 fires from the pre-suppression period (1826-1929), 11 fires from the suppression period (1930-1980), and 9 fires from the post-suppression period (1981-2022). The Mean Fire Interval (MFI) increased from 3.06 to 4.80 during the suppression period and declined to 3.88 for the post-suppression period. These findings can be used to better understand the effects that shifting management practices have had on fire frequency and provide insights to improve management efforts.

P249 - Reproductive output of closely-related sympatric pitcher plants (*Sarracenia* spp.) and their morphological hybrids

Elizabeth Companion, Jennifer Rhode Ward, Dr. Rebecca Hale, PhD, Caroline Kennedy

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The Appalachian mountain bog ecosystem is home to two closely related pitcher plants: *Sarracenia purpurea* var. *montana*, which is a federal species of concern, and *Sarracenia jonesii*, which is listed as federally endangered. One challenge concerning the conservation of these species is that they readily hybridize when in sympatry, and hybrids do not get the same federal protection as their parental species. Hybrid pitcher plants and their parental species may coexist only if they are able to maintain their population sizes through production of seeds and ramets. During summer 2022, plants at one western North Carolina bog were identified morphologically as *S. purpurea* var. *montana*, *S. jonesii*, or their hybrids and monitored throughout the reproductive season. Flower heads were collected in September and air dried at room temperature in a lab for two weeks. The number of viable seeds produced by each flower was hand counted. ANOVA was used to compare reproductive output among the three plant types. The results of this study may provide a better understanding of the possible consequences of *S. purpurea* x *S. jonesii* hybridization at this site and could be used to help make informed conservation decisions in the future.

P250 - Development and early implementation of a new feral swine population control program in the Yadkin-PeeDee River Basin..

Hannah Harper<sup>1</sup>, Rachel Bentley<sup>1</sup>, Adam Morrison<sup>1</sup>, Andrew Jacobson<sup>1</sup>, Aaron Loucks<sup>2</sup>, Luke Dollar<sup>1</sup>

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Development and early implementation of a new feral swine population control program in the Yadkin-Pee Dee River Basin

Feral swine, *Sus scrofa*, a non-native, invasive species with an estimated population size of 300 million in the Southeastern US, present an ecological, health, and economic threat across North Carolina landscapes. We herein describe the development, scale, implementation, and progress of a collaborative research and management effort between Catawba College and the North Carolina Department of Agriculture and Feral Swine Task Force. With the engagement of collaborative partners ranging from state and federal agencies, non-profit organizations, and private landowners, we seek to expand our knowledge of and address management for feral swine throughout the Yadkin-PeeDee River Basin, using tools ranging from GIS and drone technology to stakeholder engagement, field trapping, biological sampling and analysis. We present techniques implemented, progress to date, and future plans for expansion of this program.

P251 - Modeling species distributions of select obligate metallophytes in response to climate change

Lauren Eberth, Dr. John Quinn, Joe Pollard

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Climate change is threatening ecosystems globally. Species distribution models used to understand the impacts of climate change on plant populations have largely focused on species either of economic importance or with the potential to disperse along a gradient of altered temperature and precipitation. Obligate metallophytes are one group of species that may not be able to disperse in the same way, as the distribution of the soils to which they have specialized will not shift with the climate. Improved understanding of the impact of climate change on these species is needed to minimize the loss of their unique communities. To address this gap, we used MaxEnt species distribution models in R. We referenced a regional database of metallophytes to focus on species which are strict endemics and are categorized as rare, threatened, or endangered. We used current distributions, serpentine soil distribution, elevation maps, and bioclimatic variables as the basis for our models. Preliminary analysis suggests that

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*Clematis morefieldii*, known as Morefield's leather flower, is a federally endangered perennial vine of the buttercup family endemic to the Tennessee River Basin. This species has strict habitat requirements, consisting of shallow rocky soils, relatively open understory, and southwest-facing slopes. Anthropogenic habitat loss has led to the extirpation of some local populations. Little is known about the population status of extant

these species may be lost as a consequence of their narrow niche and the inhospitality of future conditions. This indicates that conservation of metallophytes and similar species may be a more pressing conservation concern than currently recognized.

P252 - Diversity and Dispersal Among Eastern Continental Divide Headwater Stream Fishes in Gwinnett County, Georgia

James E Russell<sup>1</sup>, Molly Botting<sup>2</sup>, Ryan Davenport<sup>1</sup>, Emilee Story<sup>2</sup>

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The Eastern Continental Divide marks the locations across the eastern United States where watersheds diverge to flow into either the Atlantic Ocean or the Gulf of Mexico. In Gwinnett County, the Eastern Continental Divide separates the Ocmulgee and Oconee River watersheds to the east, and the Chattahoochee River watershed to the west. This feature of the Georgia landscape was used as the basis for our hypotheses regarding connectivity, gene flow, and biodiversity of headwater streams. Headwater streams are the initial water source of river networks, and they are vital to the quality of the overall ecosystem despite being small in volume. We sampled three headwater streams across Gwinnett County, two in the Oconee River watershed and one in the Chattahoochee River watershed. Various physical and chemical properties were defined at each stream, as well as measurements of fish inter- and intraspecific diversity. We ran statistical analyses to conclude that each stream habitat was distinguishable based on mean width, depth, and dissolved oxygen. Our preliminary hypotheses correlating fish diversity with stream features were not supported. However, our data suggests that the continental divide acts as a barrier to gene flow based on the genetic diversity of three well-sampled fish species (*Sebastes atromaculatus*, *Notropis lutipinnis*, and *Nothonotus leptocephalus*) that were found on both sides of the divide. Our findings highlight a proposed methodology for headwater stream analysis that combines habitat heterogeneity with community and species-level measures of diversity.

P253 - Seed Germination, Fecundity, and Population Growth of the Endangered Morefield's Leather Flower, *Clematis morefieldii*

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populations due to the lack of long-term, population-level studies. We calculated germination and growth rates for 840 *C. morefieldii* seeds propagated at Huntsville Botanical Gardens. We estimated fecundity by measuring the seed production of 195 *C. morefieldii* vines in wild populations. Using our data along with previously published literature, we built a stage-based demographic matrix model to investigate the population growth rate. We estimated the projected population growth rate as the dominant eigenvalue of the matrix. To determine the life stage that contributed the most to *C. morefieldii* population growth, we conducted an elasticity analysis, which estimated the proportional sensitivity of each stage class to population growth. Our average population growth rate ( $\lambda$ ) indicated a slightly declining population ( $\lambda = 0.94$ ) and was most sensitive to assumptions about the survival of the reproductive life stage. This suggests that management strategies that improve the survival of the reproductive life stage may have the proportionally largest impact on population recovery. Additional research on the survival of the reproductive life stage is necessary to reduce uncertainty in our model and would improve our ability to recommend effective management or restoration strategies.

P254 - Determining Age and Growth Rates of Mussels Using a Simplified Approach

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Freshwater mussels are among the most at-risk fauna in the North America. Limited aspects of life history information are available, but generally many details are lacking for most species including attributes such as growth rates and life expectancy. Aging mussels using accepted protocols involves producing radial thin-sections from shells with a width of about 0.3 mm, a process that is tedious and requires specialized equipment. Our research was designed to create a simplified approach to mussel aging that would maintain a high level of accuracy while utilizing more conventional hardware. We experimented with shell cross-sections using a diamond edged saw blade and wet sanding, focusing on the mussel species *Elliptio pullata* from Gantt Reservoir in southeast Alabama. Our results suggested that a more conventional approach could be used to accurately age *E. pullata* up to at least four years of age, although confidence in results decreased beyond that range, and especially >6 years. Comparing our results to thin-sections would provide more certainty in our analysis as well as allow for comparison beyond the age of exponential growth. Problems associated with the current study included distinguishing disturbance rings and/or intra-annuli from annual growth rings. Further research is needed to confirm our findings, as well as analyzing additional species throughout various habitat types to determine if our methods are adequate.

P255 - The Effects of Coarse Woody Debris on Tree Height/Mortality and Soil Temperature/pH

Ms. Andrea Casallas<sup>1</sup>, Darius Ledbetter<sup>1</sup>, Porché Spence<sup>1</sup>, Amanda Leggett<sup>2</sup>, Ethan Boggs<sup>3</sup>, Zakiya Leggett<sup>1</sup>

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Symbiosis is the close relationship between two or more organisms that can dictate the survival of one another. The cleaning symbiosis between crayfish and annelids that are known as endosymbiotic branchiobdellidan annelids specifically known as *Cambarincola ingens* (*C. ingens*) has been

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Coarse woody debris (CWD) refers to the intact remains of fallen wood greater than 8–10 cm in diameter. Although its presence in forests may be seen as nothing more than dead/dying debris or trees, we know that coarse woody debris is an essential part of a healthy forest ecosystem as it provides nutrients and food to various organisms, allows energy flows, and serves as a habitat for wildlife. Our research evaluates the impact of CWD on tree height, tree mortality, soil temperature, and soil pH. Along with these objectives, this study exposed a group of high school students to the research process while also addressing a research question. We analyzed data collected from twelve plots in an loblolly pine stand at Carl Alwin Schenck Memorial Forest (managed by NCSU). Each plot had varying levels of CWD classified from low to high levels of coverage and size. Our findings showed some trends relating to high CWD and low soil temp, but overall there was no impact of CWD on any of the factors evaluated (tree height, tree mortality, soil temperature or soil pH). In the future, we would like to better understand how/why CWD and other factors affect tree mortality and height.

#### P256 - Symbiosis in Acidic and Microplastic Freshwater

Omar West<sup>1</sup>, Cameron Braswell<sup>2</sup>, Austin Gray<sup>2</sup>, Bryan Brown<sup>2</sup>

John Grosscup

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High gold prices, improved infrastructure, and poorly governed frontiers have led to gold rushes around the globe. The single largest source of anthropogenic mercury emissions worldwide is from artisanal and small-scale gold mining (ASGM), which uses mercury to form an amalgam with gold to facilitate gold extraction from surrounding sediments. However, biologists know relatively little about the long-term effects of gold mining on soil fertility and nutrient cycling. We sought to access the soil fertility by tracing its decomposition rate through time. The *Madre de Dios* Region is an exquisite site of biodiversity and houses several ecological zones; the park encompasses over 6,500 square miles and reaches an elevation of over 4000 meters which contributes to its high levels of biological diversity. To access decomposition, we implemented a novel measurement of soil litter decomposition utilizing green and rooibos tea bags which allows for normal decomposition processes, such as microorganismal breakdown of organic matter, to occur. This model has the potential to explain the sensitivity of nutrient cycling (especially nitrogen) to anthropogenic disturbances. The results show that there was no significant difference in decomposition rate ( $k$ ) between secondary forests and mined sites. Alternatively, a significant difference between the litter stabilization factor ( $S$ ),  $k_{RooibosTea}$  and  $k_{GreenTea}$  values were found based on forest classification. Further studies into macro- and micronutrient content in soils can aid in the investigation into the chrono sequence of gold mined sites and how forests recover over time after mining events.

#### P258 - The effects of dams and an introduced species, *Faxonius virilis*, on the community structure and trophic interactions of stream crayfishes in the Upper Cahaba drainage, Alabama

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Introduced crayfish and dams are two leading threats to freshwater biodiversity. The effects of both stressors can independently have major ecological impacts leading to alterations in community structure and trophic interactions, however, their synergistic effects on native systems are poorly understood. *Faxonius virilis* has been introduced into the Cahaba River drainage where it has become a dominant species in many reaches. With over 117 dams throughout the Cahaba River drainage, the interaction between *F. virilis* and the alterations caused by dams could be significant. By taking a food web approach, we are investigating the combined impacts of these two stressors on the trophic ecology of stream crayfishes and invertebrate structure. Crayfish assemblages, macroinvertebrate assemblages, and stream habitat was quantified using standard techniques on 2

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studied in various stress-induced environments within our lab. In their native environment, crayfish have various abundances observed from fieldwork being 2-4 *C. ingens* inhabiting the crayfish. This relationship is sensitive to environmental changes due to anthropogenic effects pH has varied from the norm which is approximately a pH of 8.2. Research that has been conducted on these organisms showed that the annelids inhabiting the crayfish play a hygienic role for the crayfish that is mutualistic at low and intermediary population densities (1 - 6 *C. ingens*). Previously, the Brown lab has conducted pH experiments with treatments for the experiment pH's of 6,8, and 10, and the *C. ingens* treatments having an abundance of 0, 3, and 6. Each treatment had four replicants that were averaged together over the course of the study. The study's results showed a significant percentage change in blotted wet mass for the crayfish in pH treatments *C. ingens* abundance of 3 and 6. It was concluded that *C. ingens* abundance of 3 and 6 correlate to increase growth percentage in pH's tested. These results warrant questioning how this endosymbiotic relationship would perform in the near future. In fact, it is well known that anthropogenic factors such as microplastic contamination due to freshwater acidity can be toxic to organisms in high concentrations. With this information, we will present procedures for a future experiment that replicates acidic freshwater with varying microplastic concentrations to uncover the significance of *C. ingens* abundance in these circumstances.

#### P257 - Nutrient cycling and decomposition in Amazonian forests following artisanal mining abandonment

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reaches (immediately downstream of dam and ~1km downstream of dam) in 8 impounded streams in the Upper Cahaba drainage. We repeated this sampling strategy in 6 unimpounded streams (14 streams, 28 sites total). We will quantify and compare trophic position and diet of native and introduced crayfishes by using stable isotope analysis and gut content analysis. Based on preliminary data from summer 2022 samples, 6 species were encountered across 20 sites, however assemblages typically were comprised of 1 numerically-dominant species and 1-2 subordinate species. Dominant species included *F. virilis*, *Cambarus coosae*, *C. striatus*, and *Procambarus versutus*. Subordinate species included *F. erichsonianus*, *C. coosae*, *C. striatus*, *P. clarkii*, and *F. virilis*. No range expansion was detected for *F. virilis*, and the species appeared abundant where found. Sites invaded by *F. virilis* often were depleted in native crayfish and there seems to be no strong association of dams on *F. virilis* presence/absence. These results, along with forthcoming stable isotope and gut content analysis, will help elucidate synergistic effects of dams and *F. virilis* on native crayfish assemblages.

#### P259 - Effect of time-since-fire on saw palmetto (*Serenoa repens*) leaf characteristics and potential for photodegradation

Chiara Meredith, Jennifer Schafer

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Photodegradation can increase plant litter decomposition, affecting carbon and nitrogen cycling in ecosystems. Fire may affect photodegradation by altering vegetation structure and plant cover, which can increase exposure of litter to solar radiation. We investigated variation in saw palmetto (*Serenoa repens*) leaf characteristics and the potential for fire to affect photodegradation of saw palmetto litter in Florida scrub ecosystems. Saw palmettos have fan-shaped leaves, which remain attached to the stem and spread outwards without falling to the ground immediately after dying. We measured and counted leaves of six saw palmetto individuals in scrubby flatwoods sites 6 years and 28 years post-fire. We collected sections of four leaves (new live leaf, old live leaf, dead leaf aboveground, dead leaf on the ground) of each individual to measure specific leaf area and carbon and nitrogen concentrations. Vegetation around saw palmettos was taller in sites 28 years post-fire, suggesting that there is less solar radiation reaching dead saw palmetto leaves, and thus, less photodegradation in longer unburned sites. Length, width, area, and number of live saw palmetto leaves did not differ between sites. Dead leaf length was lower in sites 6 years post-fire, which suggests that self-shading of dead leaves is higher in recently burned sites. Specific leaf area varied among leaf types and was greater in sites 28 years than 6 years post-fire, but there was no interaction between leaf type and time-since-fire. Measurements of foliar carbon and nitrogen will allow us to test the prediction that there is a greater decline in nutrient concentrations with leaf age in sites 6 years than 28 years post-fire due to higher solar radiation and photodegradation. Knowledge about the potential role of fire in mediating the impact of photodegradation on plant litter decomposition in fire-prone ecosystems could improve understanding of carbon and nitrogen cycling.

#### P260 - Recovery of the Fish and Crayfish Assemblage in a Southern Appalachian Stream Following a Major Debris Flow

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We followed the process of recovery of the fish and crayfish assemblage in Peeks Creek, a high-gradient second order stream in the Little Tennessee River watershed of North Carolina after a debris flow in 2004 devastated the channel and its riparian zone. *Oncorhynchus mykiss* (Rainbow Trout), *Rhinichthys cataractae* (Longnose Dace), and *Cottus bairdii* (Mottled Sculpin) were the major components of the fish assemblage. Rainbow Trout was the only species to appear in year 1 after the debris flow. Longnose Dace appeared in year 3. Mottled Sculpin did not become established until year 6 and it wasn't until year 9 that sculpins constituted a major component of the fish assemblage. *Cambarus bartonii cavatus* (Appalachian Brook Crayfish) numbers recovered by year 2, though only one individual was found the year following the debris flow. To further assess the recovery of the fish assemblage in Peeks Creek, we used the Horn-Morisita Index to track and compare the fish assemblage in Peeks Creek in the years following the debris flow to fish assemblages in six nearby reference streams that had similar watershed area, elevation, and that had a Jaccard Similarity Index  $\geq 0.4$  as compared to the Peeks Creek fish assemblage in 2019. Based on these comparisons, the fish assemblage in Peeks Creek did not fully recover until year 15, after the channel and riparian zone had stabilized. Unassisted natural recovery occurred after a costly engineered restoration project had been rejected and arguably represents the preferable solution. However, recovery of the fish assemblage may not have been achieved if the stream flowed directly into an impoundment or low gradient river that lacked the source of species for recolonization, or if the stream had been located above a barrier to upstream movement.

#### P261 - Calculating Neighborhood Competition of Trees in Pacific Northwest Coast Range by Using the Dendrochronological Data

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One of the greatest impediments to forestry regarding individual tree competition is that it becomes challenging to devise the optimal competition index. This is because the success of the competition index, which is a numerical expression of the degree of use of the growth potential limited by the genotype of a tree species, will alter depending on stand conditions. The success of a competition index depends on tree species, accessible data, and the structure of the selected model. These competition indices indicate the level of competition for a particular tree. Fundamentally, competition found in forest trees can be categorized into two groups: competition among conspecific individuals, plants of same species, and heterospecific individuals, plants of different species. Literatures focuses on the associations among western hemlock (*Tsuga heterophylla* (Raf.) Sarg), Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) and red alder (*Alnus rubra* Bong.), regarding how competition among them affects their growth. To fully understand this mechanism, specifically for growth among those species, long term studies are needed. In this research, data are collected from

the production plantations of Lewis and Clark Timberland in the Coast Range mountains of northern Oregon and southern Washington. Using tree-ring technology, annual growth of Douglas fir, red alder and western hemlock will be tracked. The first objective is to find the effect of competition on individual-tree diameter growth. Specifically, different competition indices for intraspecific and interspecific competition among those species will be calculated. The second objective is to formulate the best fitted model for the diameter growth rate of the trees by using multiple predictor variables. Diameter growth will be formulated with or without competition index as one of the predictor variables.

Then, regression models will be developed to predict the diameter growth correctly.

**Keywords:** western hemlock, Douglas fir, red alder, competition indices, diameter growth, regression model

P262 - Terrible roommates: Invasive crayfish serve as incompetent surrogate hosts for native symbionts.

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Symbiosis is integral to the life history of many organisms. In the last two decades, there has been considerable research to understand the relationship between crayfish and their symbiotic annelid worms. Symbionts can have a mutualistic effect on their crayfish host; however, at high symbiont densities, they have been shown to damage crayfish gills and alter behavior. We sought to understand how this symbiotic relationship functions under the influence of invasive crayfish species. We hypothesized that the invasive species would possess lower amounts of symbionts. To accomplish this we surveyed 32 sites in an Urban Stream in Montgomery County, Virginia, and obtained 10 crayfish with a carapace length of at least 20 mm in length at each site. Crayfish were then subjected to a 10% MgCl solution to remove all symbionts. We found that the introduced *Faxonius cristavarius* dominated our stream community making up 55% of the observed crayfish, but on average harbored a very low amount of symbionts compared to native species within the creek. *Cambarus appalachensis* and *Cambarus Bartonii* were the only native species found within the creek and contained the highest amount of symbionts. Our hypothesis was supported and we explained this by accounting for *F. cristavarius*' heightened symbiont grooming behavior when compared to native species. The almost total lack of symbionts on *F. cristavarius* is supported by the literature with *F. cristavarius* being observed to groom off symbionts at densities of one, compared to native species which groom at densities of 10. These findings are significant and reinforce laboratory studies demonstrating that *F. cristavarius* may harbor significantly fewer symbionts than native species of crayfish. Further studies should seek to understand if suboptimal hosts are decreasing overall symbiont abundance within a stream community.

P263 - Evaluating the macroinvertebrate community to determine stream flow duration

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True first-order headwater streams are often over-looked for environmental protections even though they are important habitats for macroinvertebrates and are important to the ecology of downstream water bodies. A lack of clarity on formal protections due to changing regulations and frequent exclusion from topographic maps has further allowed for the degradation of already degraded source headwater streams. Macroinvertebrates exhibit varied tolerances and life cycles, evaluating this can allow for a better understanding of stream health but can also be used to determine the stream flow duration. Through this sampling method streams that are not usually seen on topographic maps can attain the necessary legal protections.

We conducted macroinvertebrate surveys on six small true 1<sup>st</sup> order headwater streams near Wheeling, West Virginia. The Alternative Macroinvertebrate Sampling Method for High Gradient, Headwater Step/Pool Dominated Streams as well as The Headwater Macroinvertebrate Field Evaluation Index (HMFEI) were used to determine if streams were perennial, intermittent, or ephemeral. Sites were sampled once in the Spring in 2022 and once again in the Winter of 2022. All macroinvertebrates collected were identified to family level. Spring samplings indicated that four out of the six streams were class III PHWH (Perennial) streams and two out of the six were class II PHWH (Intermittent) streams. In early winter only four out of six streams were able to be sampled with all four of the sampled streams indicating as being Intermittent. This discrepancy in flow duration of our four perennial streams is likely due to life history traits of the macroinvertebrates, where indicator taxa were not yet large enough to be collected.

Additionally, water chemistry at one site was found to be chemically impaired, which may have negatively impact the macroinvertebrate community.

P264 - Relationship between bacterial abundance and watershed elevation in forested streams of northwestern South Carolina

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Anthropogenic land cover, such as urban or agricultural lands, can influence bacterial abundance in freshwater ecosystems. However, natural landscape variations, such as varying topography, might complicate efforts to determine influences of anthropogenic land cover. For example, previous studies in the Great Smoky Mountains have shown that concentrations of fecal bacteria are negatively correlated with elevation in forested watersheds. The goal of our study was to determine how concentrations of suspended bacteria vary with elevation in the Blue Ridge and Piedmont provinces of South Carolina. We hypothesized that concentrations of suspended bacteria would decrease with increasing elevation. During June/August 2022, we sampled 16 second to third order streams that drained from watersheds with  $\geq 80\%$  forest cover and  $< 8\%$  total developed

land cover. The sample locations ranged in elevation from 80 m to 600 m. Each stream was sampled 1-2 times under baseflow conditions. We measured the concentrations of total heterotrophic bacteria, total coliforms, *Escherichia coli*, and *Pseudomonas aeruginosa*. We also measured water temperature, turbidity, and concentrations of major ions and iron. For all bacteria, we found negative correlations between concentrations and sample site elevation. These correlations were statistically significant for all bacteria except *E. coli*. We also found that water temperature, turbidity, and iron correlated negatively with sample site elevation. As a result, concentrations of bacteria correlated positively with water temperature, turbidity, and dissolved iron. Our results suggest that inverse relationships between bacterial concentrations and elevation in streams are not restricted to fecal bacteria. Additional studies are needed to better understand the relationships between bacterial abundance and environmental factors that vary with elevation, such as water temperature, turbidity and dissolved iron. Also, our results suggest that variations in bacterial abundance associated with topography need to be considered in future studies of bacteria/land cover relationships.

P265 - Evaluation of Growth and Mortality in Crayfish Internally Tagged with p-Chips.

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Numerous ecological questions such as movement capacity, population size, growth, and longevity can be answered using uniquely marked individual animals. However, available tagging technology for small-bodied aquatic organisms like crayfish is limited. One popular tagging option for crayfish over the last decade has been passive integrated transponder tags, but their application is limited to individuals greater than ~20 mm carapace length (CL). We tested a new tagging technology called p-Chips which are small (500 µm x 500 µm x 100 µm) microtransponder tags with a unique code read by a light-activated reader. They have previously been used in tagging analysis with ants, honey bees, mice, and fish, but nothing has been published for crayfish. Our objectives were to evaluate the effects of p-Chip tagging on the mortality and growth of crayfish and determine if tags would be expelled or unreadable after molting. We used Woodland Crayfish (*Faxonius hoyi*) ranging in size from 12.2–26.6 mm CL. Crayfish were randomly assigned to a treatment group (n = 38) of either double-tagged (p-Chip + Visible implant elastomer-VIE), singletagged (p-Chips), or control (no tag). Insertions for p-Chips and VIE tags were made on the ventral region of the second or third pleon segment and crayfish were held in individual containers in a laboratory recirculating system. Crayfish were fed daily and checked for mortality and molts. By day 81 of 90 (when this was submitted), we observed low mortality (11%) equally distributed between the three treatment groups and no p-Chips were expelled despite 68% of crayfish molting at least once. Of the 76 p-Chips implanted, four did not work after insertion and four were unreadable after molting. Our results suggest p-Chips are a viable option for the application of individually identifiable marks on crayfish as small as 12 mm CL.

P266 - Oyster spat (*Crassostrea virginica*) and barnacle (*Chthamalus fragilis*) settlement in relation to microsite characteristics of two southern Georgia salt marshes

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Much of the United States' East Coast has experienced a decline in eastern oyster (*Crassostrea virginica*) landings since the early 1900s. In Georgia, this decline has been linked to overharvesting, disease, storms, and alterations in both water quality and natural flow regimes. Although past research has provided important information about the distribution of Georgia oyster reefs, spat settlement differences in relation to microsite characteristics, especially of the southern Georgia coast, have not been adequately addressed. A study was begun in March 2020 to investigate correlations between environmental parameters and oyster spat settlement. Three sites were selected on Sapelo Island and three sites on Jekyll Island in relation to the upland-estuary-sound gradient. Oyster spat settlement racks were established at each site. Tiles were collected every six weeks and number of oyster spat along with other biofouling organisms such as barnacles (*Chthamalus fragilis*) were determined. Additionally, water quality and sediment composition were measured during each collection period. The highest oyster spat settlement occurred in open sites with relatively high salinity, low turbidity, and high nitrogen levels. The highest barnacle settlement occurred in upper tidal creeks with relatively low salinity, high turbidity, and low nitrogen levels. Implications of this preliminary analysis are discussed as well as future in-field manipulation experiments. This study is an initial attempt to develop a model for identification of potential oyster reef restoration sites.

P267 - Patterns in metabolic activity in a small, human-constructed pond.

Lucy Ellis, Kenneth Fortino

*Longwood University, Farmville, VA*

Small, human-constructed ponds are abundant aquatic habitats that are sites of high primary production, organic matter storage, and dynamic hydrology. Because of this, small human-constructed ponds serve as important control points for ecosystem function, which can have substantial impacts on watershed biogeochemistry. We quantified the variation in dissolved oxygen, temperature, and light in Chalgrove Lake, a 1.2 ha. humanconstructed pond over the course of a year. Environmental conditions (temperature, dissolved oxygen, pH, and conductivity) near the surface of the pond were continually measured using a YSI EXO3 sonde. Depth profiles of temperature, dissolved oxygen, and light were also collected approximately monthly. The depth profiles of temperature show that the pond did thermally stratify down to approximately 2.5 m during the summer before turning over in November and remaining fully mixed throughout the winter. During the summer stratified period, the surface waters of the

lake were supersaturated in oxygen but oxygen levels declined with depth, even above the thermocline. These patterns suggest high levels of water column primary production but also high levels heterotrophic oxygen consumption, which is likely from the sediments. Following fall turnover and throughout the winter, the lake remained undersaturated in oxygen but there was still a decline in oxygen concentration with depth, suggesting netheterotrophic conditions. Although light was rapidly attenuated with depth in all seasons ( $k$  ranged  $1.4 - 3.3 \text{ m}^{-1}$ ), sufficient light for photosynthesis ( $> 1\%$  surface irradiance) reached the bottom of the lake, suggesting that primary production was not light limited. Overall our results suggest that Chalgrove Lake is a metabolically active system that both accumulates and processes considerable organic matter throughout the year and is likely a control point for organic matter processing within the watershed.

P268 - The presence and consistency of microplastics from sediment and sand samples from Dauphin Island Sea Lab, Alabama.

James Rayburn, Sarah Yarborough, Leandra Custodio

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Microplastics are widespread contaminants that are present in virtually all environmental areas. Microplastics are small pieces of plastic  $< 5 \text{ mm}$  that can be either intentionally produced or produced by abrasive action of larger pieces of plastic to create the smaller pieces. Microplastics are present in marine environments such as the areas around Mobile, Alabama and the Dauphin Island Sea Lab. These microplastics are highly persistent contaminants and potentially harmful to organisms and ecosystems. An attempt to detect the presence of and quantify the consistency of microplastics from Dauphin Island Sea Lab Alabama, samples were taken both from sediments from the bay and channel and from the sand from the shore. We hypothesized that there would be more microplastics present in the sand samples taken from shore and less from the sediments. We based this on microplastics being produced and shed near people. A  $> 1 \text{ Kg}$  sample was taken from several sites around Dauphin Island Sea Lab Alabama. Subsamples of 200 grams were mixed with a hypersaline solution for 10 minutes. The supernatant was then filtered, and the filter paper analyzed under a stereo microscope for the presence of microfibers. Three replicates from each sample were analyzed to determine the consistency an amount of plastics present. Means, standard deviation, standard error of the mean, ANOVA and Fisher's LSD post hoc test was performed. These results show the continued need for understanding the amounts and types of microplastics that are present in our environments.

P269 - Using Environmental DNA to Locate and Identify Bacterial Species in Polluted and Non-polluted Freshwater Ecosystems in South Florida.

Analuz Nieves<sup>1</sup>, Jacqueline Valiente<sup>1</sup>, Tammy Laberge<sup>2</sup>

results were complicated when we had some contamination in our PCR results. Therefore, we repeated the PCR reactions multiple times. The significance of this research is to highlight the differences in bacterial communities in polluted and unpolluted waters. Using eDNA is an ideal way to determine the types of bacteria that may inhabit both polluted and clean aquatic environments. In the future, we will create a database of the bacteria that inhabit these freshwater environments using the DNA sequences we obtain from our study. This information will allow more precise monitoring of the types of bacteria found in our environment.

P270 - Variation Among Individual Plants of Golden Ragwort (*Packera aurea*) with Regards to the Presence of Abaxial Anthocyanins in Overwintering Basal Leaves

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The herbaceous perennial, *Packera aurea*, is widespread across the southeastern United States. Commonly found growing in the understory of temperate forests, it has been observed to feature anthocyanins on the abaxial side of its leaves, particularly in winter and early spring prior to canopy leaf out. Within a community some individuals have deep purple abaxial coloration, a few show anthocyanins on both leaf surfaces, while others have no purple coloration at all, even though they are growing adjacent to each other. This population variation brings into question what stimulates the production of anthocyanins and what benefit such pigmentation can bring to the physiological functioning of the plant. The currently accepted hypothesis is that abaxial anthocyanins function in photoprotection from periods of high light, i.e., sun flecks, but this does not explain why some individuals can survive without anthocyanins. We measured pigment concentrations in these morphs and in December, purple morphs,

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We collected environmental DNA (eDNA) to examine the diversity of bacterial species in freshwater environments in the areas surrounding MiamiDade College (North). Environmental DNA arises as a result of DNA being shed into the environment by organisms that are present in the area; it can be used to identify species present without having to locate or culture these organisms. Our goal is to identify and compare bacterial species in previously polluted (restored Superfund sites) and clean freshwater sites. We plan to identify the bacterial species present using DNA metabarcoding. Numerous eDNA extractions were completed from the following sites: Amelia Earhart Park in Miami, FL, Optimist Park in Miami Lakes, FL, and the MDC Pond at North Campus, Miami, FL, and Spring Garden Point Park, Miami, FL. Water was collected from these sites, and DNA was filtered from these samples. We amplified a portion of the 16S gene by polymerase chain reactions (PCR) and visualized our results by gel electrophoresis to identify samples that had enough DNA for sequencing. As a result, we obtained about 30 samples to be sequenced. Our

as expected, had higher anthocyanin contents, but the two morphs did not differ in their chlorophyll concentrations. We plan to make gas exchange measurements of basal leaves of each morph during the leafless winter season coupled with fluorescence measurements (F<sub>v</sub>/F<sub>m</sub>) to determine whether individuals differ in rates of gas exchange and cold/high light stress. Measurements will continue throughout the spring and summer to determine ontogenetic changes in pigment concentrations and how this is related to photosynthesis and protection against stress. The results of this study should help to explain what causes plants to produce abaxial anthocyanins and what evolutionary benefit such pigments provide to the success of a plant's photosynthetic functioning.

P271 - The Effects of Varying Microplastic Morphologies on Crayfish-Annelid Symbiosis

Cameron Braswell, Tyler Allen, Bryan Brown, Austin Gray

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Annually within the United States, an estimated 1.13 to 2.24 metric tons of plastic waste is mismanaged or littered, ultimately, evading waste management practices. Once scattered across our environment, this plastic debris undergoes natural degradation processes causing exponential degradation and fragmentation. In fact, microplastic pollution in marine and freshwater systems can vary with concentrations ranging between (4137 to 12,000 items/m<sup>3</sup>). This poses the question - what impact does microplastic pollution have on freshwater ecosystems? Through the use of exposure-response assays, we investigated how plastic fibers, tire wear particles, and microbeads (15 mg/L) impact the symbiotic relationship between crayfish and annelids. Following a 21-day chronic exposure, we observed crayfish mortality, growth, and plastic particle bioaccumulation. At the conclusion of the exposure, crayfish demonstrated resilience to microplastic exposure with mortality ranging between (0 to 5%). Furthermore, there were no differences observed in growth rate between control and the three plastic morphologies ( $p > 0.05$ ). Average percent change for crayfish mass and length were ranged (Mean  $\pm$  SE) (0.94%  $\pm$  1.79) and (0.43%  $\pm$  0.69), respectively. Although no toxicological effects were observed across these three plastic morphologies, there was an inherent plastic particle bioaccumulation that potentially explains this result. Given the inherent bioaccumulation of plastic particulates from their habitat, crayfish may act as a prey-item introducing microplastics to the freshwater food web.

P273 - The effects of the riparian zone and stream wood abundance on fish populations in upstate South Carolina streams

Marshall Trout, Dennis Haney

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Riparian zones serve a variety of functions in both the terrestrial and aquatic environments. They provide shade and habitat for both terrestrial and aquatic animals, are a major supplier of energy to streams in the form of allochthonous input and provide a buffer slowing down the movement of chemical and sediment runoff into streams. Because of this the presence of a robust riparian zone is likely very important to the distribution of stream fishes as well. However, riparian zones are threatened by human use of the land, whether this is by removing the riparian zone for agricultural use, or by channelizing streams for irrigation. The primary purpose of this study was to see if there are relationships between the quality of the riparian zone, abundance of wood in streams serving stream functions, and fish assemblages. We were also interested in relationships between these microhabitat level measurements with watershed-scale land covers and measures of habitat and sediment quality. These different factors were sampled in 15 different streams in Upstate South Carolina varying in the amount of urban and forested land covers. We found that there was a negative correlation between riparian score and development of land around the stream, as well as a negative correlation between total abundance of fish and development. Greater cover provided by the riparian zone was positively correlated with benthic fish abundance, but with fewer pioneer fish, suggesting the importance of riparian habitat for fishes less able to acclimate to human-induced changes in stream conditions. We also found that even when the land around the stream has abundant cropland or grassland there is still a positive correlation with stream structure and fish distribution and the quality of the riparian zone.

P274 - Evaluating bat diversity and activity in developed and undeveloped locations of West Virginia's northern panhandle.

Julia Purks, Diana Barber

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Bat populations are declining due to habitat loss and the spread of the deadly fungal pathogen, white-nose syndrome. Increased studies of bat populations and their diversity are necessary to ensure their survival. Bat diversity and activity levels are understudied in the northern panhandle of West Virginia. For our summer 2021 study, sixteen sites were selected (four in each county: Ohio, Brooke, Hancock, and Marshall). Using mobile ultrasonic bat detectors, bat calls were detected from different species at each location while walking a 1-kilometer transect after sunset. Our null hypothesis suggests that sites will not show any statistical difference of bat diversity in developed or undeveloped locations of West Virginia's northern panhandle. Our alternative hypothesis formulates that bat diversity will be statistically different between developed and undeveloped transects. Results from the summer transects show a significant difference between activity in developed and undeveloped sites ( $p = 0.008433$ ). During the spring of 2022, a study was started to assess temporal changes in bat activity and diversity. Two sites were selected (Bethany College Woods and West Liberty University Woods) to place stationary recorders starting at the end of February. Sites were, and still are, being evaluated based on the number of species recorded at each site.

P275 - Determining the variation in anthropogenic microfiber pollution in the Richland Creek watershed in Nashville, TN

Asher Amundson, Callie Swallows, Matt Heard

*Belmont University, Nashville, TN*

Anthropogenic microfibers including microplastics have been suggested to pose a threat to both wildlife and humans when they are present in freshwater ecosystems. However, there have been relatively few studies that have examined how these microfibers vary in presence and abundance in most streams and tributaries. In this study, we examined how the presence and abundance of anthropogenic microfibers varied in six tributaries of Richland Creek, which is an important body of water that runs into the Cumberland River in Nashville, TN. To do this we collected grab samples of surface water at four time points from six locations. Overall, we found that all of our sites had microfibers present, but that there was significant variation in the amounts over time. Collectively, we hope that our findings represent an important first step in documenting this form of pollution and help us to begin to source where these microfibers are coming from.

**P277 - Photosynthetic Measurements of Several Common Moss Species of the Southern Appalachian Mountains**

Leigha Henson, Howard Neufeld

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Mosses function as keystone species and bioindicators of forest integrity. They are sensitive to changes in atmospheric conditions and may influence ecosystem functioning out of proportion to their biomass. Yet despite their importance, there have been no studies of their ecophysiology in the Southern Appalachian Mountains (SAM). We therefore have begun a study of SAM moss ecophysiology and are investigating photosynthetic responses of four SAM moss species to varying levels of PAR, using two species from open habitats and two from forest understories. We used a custom-built cuvette attached to a Li-6800 gas exchange system to better control RH. We calculated the following parameters: maximum photosynthetic rate ( $A_{max}$ ) at light saturation (LSP), dark respiration rate ( $R_d$ ), light compensation point (LCP), and apparent quantum efficiency (AQE) for each species, as well as chlorophyll contents of each species. We hypothesized that mosses in understory habitats would have more chlorophyll and lower a:b ratios than open-habitat mosses. We also hypothesized that open habitat mosses would exhibit higher LCP, LSP,  $A_{max}$  and  $R_d$ , but lower AQE compared to understory mosses. *Polytrichum juniperinum* and *Ceratodon purpureus*, open habitat mosses, had higher LSPs (1825 and 1050  $\mu\text{mol m}^{-2} \text{ s}^{-1}$ , respectively), than the understory species (*Hypnum imponens* and *Thuidium delicatulum* at 805 and 565  $\mu\text{mol m}^{-2} \text{ s}^{-1}$ , respectively). Understory mosses had the lowest  $A_{max}$  at 1.90 and 1.1  $\mu\text{mol m}^{-2} \text{ s}^{-1}$  for *H. imponens* and *T. delicatulum*, while open habitat mosses, *C. purpureus* and *P. juniperinum*, were higher at 4.6 and 17.0  $\mu\text{mol m}^{-2} \text{ s}^{-1}$ , respectively. *Polytrichum juniperinum* had the highest total chlorophyll at 53.5  $\mu\text{g/mL}$  while *H. imponens* and *T. delicatulum* were lower 11.9 and 10.7  $\mu\text{g/mL}$  respectively. The results of this research should provide a basis for understanding how SAM mosses are adapted to their habitats and may respond to future climate change.

**P278 - Molecular survey of methane-cycling archaea in methane-soaked subsurface sediments (Guaymas Basin, Gulf of California)**

John Hinkle<sup>1</sup>, David Geller-McGrath<sup>2</sup>, Paraskevi Mara<sup>2</sup>, Virginia Edgcomb<sup>2</sup>, Andreas Teske<sup>1</sup>

known methanogenic lineage. Methane-sulfate interfaces, which extended into intermediate depths yielded numerous ANME-1 sequences from each examined site. A general ANME-1 phylogeny was constructed using the five most abundant ANME-1 *mcrA* gene sequences from seven of eight drilling sites. Reference sequences from NCBI GenBank were added to add phylogenetic context to the tree and allow for cluster labeling. We also constructed two site-specific (1548B, 1552B) trees using all *mcrA* gene sequences with >100 clones. Site 1548B is a hydrothermally hot site, while site 1552B is a cold seep site. Numerous ANME-1 phylotypes were closely related to mud volcano reference sequences, as well as reference sequences from Guaymas Basin and Gulf of Mexico subsurface sediments. Our work allows for a greater understanding of the diversity of methane-cycling archaea in the Guaymas Basin.

**P279 - Gut's going on? Seasonal and sex-specific changes in the gastrointestinal tracts of *Peromyscus maniculatus***

Olivia Chapman, Bryan McLean

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The Guaymas Basin is a young marginal rift basin in the Gulf of California characterized by active seafloor spreading, rapid deposition of organicrich sediments, and steep geothermal gradients. Methane is abundant in the Guaymas Basin hydrothermal sediments and previous studies of surficial Guaymas sediments have yielded a diverse array of methane-cycling archaea, yet their diversity in deep subsurface sediments is still unknown. Subsurface sediment samples were selected from eight drilling sites to explore the diversity and range of methane-cycling archaea in the Guaymas Basin subsurface. DNA was extracted from near-surface, intermediate, and deeper depths using commercial DNA extraction kits and was PCR amplified using primer pairs that target the *mcrA* gene and ANME-1-related archaeal groups. *mcrA* was utilized as an alternative marker gene to 16S rRNA for the phylogenetic analyses. Three of the eight sites yielded the *mcrA* genes related to previously established methane-cycling lineages. Uncultured *mcrA* groups included *mcrA* group e (presumably anaerobic methane oxidizers) and a new uncultured *mcrA* lineage unrelated to any

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Functional traits are phenotypic characteristics that contribute to fitness of individuals in dynamic and changing environments. In mammals, both categorical and continuous (e.g., quantitative) functional traits have been extensively utilized as proxies for diet, locomotion, and other aspects of species ecology, but there has been less focus on soft tissue form and function. This is particularly true for the digestive system, which varies in size and complexity across Class Mammalia and plays a major role in the energetics of species. To guide more effective utilization of gastrointestinal (GI) morphology as a functional trait in small mammals, we examined how GI tracts (lengths and masses of four GI sections) varied within a population of deer mice (*Peromyscus maniculatus*) in the Southern Appalachian Mountains of North Carolina, USA. We collected samples of adult *P. maniculatus* monthly for one year and measured these GI traits to quantify variation with seasonality and diet, providing insight into temporal plasticity in this soft tissue trait over time. We found that both season and reproductive activity had a significant effect on the total length and wet mass of the GI tract. January mice had the longest GI tracts and lengths decreased into the summer, before rising again in the fall and the beginning of winter. Males and females responded in sex-specific ways to demands of reproduction, with reproductively active males having significantly shorter and lighter GI tracts than non-reproductively active males. Our study provides proof-of-concept for understanding population-level plasticity in a rarely-collected soft tissue trait, which may also be complimentary to standard craniodental measurements as a functional dietary proxy to understand mammalian ecology and community assembly.

P280 - Investigations of Nucleotide Modifications in Winthrop's Bacteriophage Collection

Gabrielle Walker, Victoria Frost

*Winthrop University, Rock Hill, SC*

Bacteriophages (phages) are viruses that replicate in bacteria and have co-existed with their host in a complicated, evolutionary arms race for approximately three billion years. Both partners continuously evolve attack and defense mechanisms to ensure their own survival. Bacteria employ an array of defense mechanisms, including a Restriction-Modification System that utilizes restriction endonucleases (REs) to destroy infecting phage genomes. Methylation of the DNA bases at RE recognition sites is often used by bacteria to protect their own genomes. Phages can utilize methylation, or as appears to be more common, using noncanonical nucleobase substitutions. Purine and pyrimidine precursor molecules are modified by phage encoded genes. They are then incorporated into phage genomes during replication, and effectively "camouflage" their RE sites. The objective of this study was to reveal possible phage DNA modifications by comparing patterns of endonuclease cleavage of Winthrop's collection of phage genomes, to virtual profiles of predicted endonuclease activity. Our protocols utilized methylation-insensitive enzymes, to reveal possible RE site protection in the phage genomes. In parallel, an *in-silico* tool, available at New England Biolabs (NEB), was used to generate virtual digestion patterns using the same suite of enzymes. Our results highlighted that several of our phages' DNA was blocked from digestion, especially those grouped in the EA cluster. These EA phages show resistance to similar REs, suggesting that phages within the same cluster may display similar nucleobase substitutions. Further investigations are underway to identify modification similarities between phages within related or unrelated clusters. Biochemical analysis to reveal the presence and molecular structure of noncanonical nucleobase substitutions is a future goal of these investigations. Detailed knowledge of phage genome protective mechanisms is important, especially as it may contribute a competitive advantage to the phage when used in a therapeutic setting.

P281 - Growth and Development of *Ambystoma maculatum* Larvae in Relation to Vernal Pool Size and Hydroperiod

Matthew Hanbury, Dr. Rebecca Hale, PhD

*University of North Carolina Asheville, Asheville, NC*

Vernal pools are seasonal wetlands that provide important habitat for regional biodiversity and are an essential reproductive environment for many amphibians. Importantly, vernal pools are susceptible to increasing drought and flooding events, which may be driven by the hydrological impacts of urbanization and climate change. Little is known about how fluctuation in the hydroperiod of these pools could affect the development and survival rate of the species that depend on this habitat. Vernal pools typically dry up in mid-summer and are usually refilled by late winter; however, if these habitats dry up too early, during the aquatic phase of developing amphibian larvae, this could impact the developmental rate and potential survival of larvae. In this study we investigated how the fluctuation of hydroperiod can affect the rate of metamorphic development and survival of larval stage salamanders. We selected four isolated vernal pools with breeding salamander populations, notably differing in size to measure hydroperiod. We measured the wet length, wet width, and maximum depth of each pool in weekly intervals. From each pool we collected three random samples of ten dip net sweeps and measured the number of individuals, as well as the developmental stage and size of the salamander larvae collected. We used these data to determine if the developmental rate of larvae was increasing in the smaller pools, due to the increasing pressure of decline in hydroperiod, compared to those sampled in the larger pools. The methods used and data collected here, coupled with a regional expansion of similar vernal pool monitoring, could further contribute to our understanding of the importance of this vital habitat and future amphibian conservation efforts.

P282 - Kanamycin-Resistant Bacteria in Earthworm Digestive Tracts and Soil from Residential Compost Bins

Samantha Dooyema, Abby Morse, Chris Barton, Darlene Panvini

*Belmont University, Nashville, TN*

Antibiotic-resistant bacteria are becoming more prevalent in the soil as humans have increased impacts on the environment. Previous research proposed that earthworms could affect bacteria distribution as the soil is ingested and filtered through their bodies. This research aimed to determine if kanamycin-resistance bacteria are the same in earthworm digestive tracts as in the composted soil the earthworms inhabit. We expected that the gut contents and the composted soil would vary in the occurrence of kanamycin-resistant bacteria. Earthworms and composted soil samples were collected from compost bins at a residential home in Nashville, Tennessee. Digestive tracts were dissected from the earthworms and homogenized. Kanamycin-resistant bacteria were isolated from the gut contents and soil samples, then identified to genera using DNA barcoding and Sanger sequencing of 16S rDNA. Kanamycin-resistant bacteria were found in both composted soil and earthworm guts. More types of kanamycin-resistant bacteria were found in the composted soil (8 genera) compared to the earthworm digestive tracts (3 genera). *Cellulosimicrobium* was found in both soil and gut samples and was kanamycin-resistant, indicating that some bacteria are shared between the two samples. *Krasilnikoviella* and *Cellulomonas* were two kanamycin-resistant bacteria found only in the worm samples and are typically found in the normal gut microbiome of earthworms. These findings indicated that there are differences in the occurrence of kanamycin-resistant bacteria found in earthworms and soil from compost bins and give insight into the role that earthworms may play in transferring microbes among locations.

#### P283 - Population Demographic Modeling of American Ginseng (*Panax quinquefolius* L.) Populations in Western North Carolina

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American ginseng (*Panax quinquefolius* L.) is an herbaceous perennial understory plant distributed throughout deciduous forests in eastern North America. Ginseng is widely sought-after for the medicinal compounds (ginsenosides) in its roots. A majority of harvested ginseng is sold for use in traditional Asian medicine. Overharvest and scarcity of Asian ginseng (*Panax ginseng* L.) led to mass exports of American ginseng to Asia starting in the early 1700s. American ginseng has since been overexploited and has experienced major population declines. Wild ginseng harvest is regulated in many states to conserve and protect populations of the species, yet illegal poaching still occurs. Ginseng is slow growing and typically reemerges each year from its roots as leafy stems in different size classes, which are often categorized by the number of leaves present. Their aboveground vegetative growth year-to-year can increase, stay the same size class, or revert back to smaller size classes (typically in response to stress). Using several years of demographic data collected between 2011 and 2022 from six western North Carolina populations, we created demographic models to understand patterns, demographic change, and the long-term population viability of these populations. The demographic monitoring of these populations did not include extensive fertility data (seed production and seed fate), so we are using published fertility data from studies in West Virginia, Missouri, and Quebec, Canada to see which best fits existing demographic data. Through this research, a better understanding of the population dynamics of wild ginseng in western North Carolina will be attained. Once defined, models can also be used to simulate the effects of different harvesting intensities, which can aid in developing sustainable harvesting protocols. This work will also highlight demographic monitoring work, such as collecting regionally specific fertility data, that is needed to better model wild ginseng populations in western North Carolina.

#### P284 - Survival of clinically relevant bacterial species on the surface of different types of plastics used in healthcare

Ashely George<sup>1</sup>, David Levine<sup>2</sup>, Henry Spratt<sup>3</sup>

#### P285 - Determining Ecological Parameters for the Trapdoor Spider *Antrodiaetus unicolor*

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A factor contributing to higher patient morbidity and mortality rates continues to be healthcare-associated infections (HAI). Although patient endogenous bacteria may be the primary cause of many HAIs, 20 to 40% of these infections are estimated to be caused by cross-contamination. Contact with contaminated surfaces in medical facilities may result in infection of a patient causing an HAI. Many Gram-negative bacteria and Gram-positive bacteria can contaminate surfaces and cause HAIs. Previous studies have found that different species of bacteria survive differentially on different types of plastics found in healthcare facilities. For this study survival of *Staphylococcus aureus*, a gram-positive cocci and *E. coli*, a Gram-negative rod were tested on four different types of plastics (polyethylene, polyurethane, PVC, latex) typically found on reusable healthcare surfaces (e.g., walker handles, exercise balls, gloves) often shared by multiple patients. Known quantities of bacteria were aseptically placed on disinfected plastic surfaces in triplicate, air dried, and then incubated at room temperature for 30 minutes, 20 hours, and 40 hours. After incubation, using sterile transport swabs, samples were collected from the plastic surfaces, transferred into a tube of sterile saline, vortex mixed, and serially diluted before being plated on tryptic soy agar for plate counts. Overall, both species of bacteria survived on the plastic surfaces tested for at least 30 minutes. Bacterial survival on polyethylene/polyurethane was greater compared to PVC. Bacterial survival on latex was lower than for all the other plastics. The relevance of these findings is that depending on the plastic used in health care facilities, there may be better choices of plastics to suppress the growth of unwanted environmental bacteria. In addition, the potential for the addition of antimicrobial chemicals (e.g., Zinc, Silver, Microban) to plastics to help suppress microbial numbers on these surfaces may help reduce HAIs linked to environmental exposures.

*Antrodiaetus unicolor* is a trapdoor spider species endemic to the eastern U.S., specifically the central and southern Appalachian regions. Fringe populations are found as far west as the Ozarks and down south as far as the Gulf coast. Characteristically, *A. unicolor* excavate burrows are lined with silk that are hidden by collapsible or foldable “doors”. The doors are made of a combination of silk, soil, and dead plant material. Burrow aggregations are found in cool, humid, and loamy soils in densely forested areas. Burrows are often located on inclined surfaces such as ravines, stream banks, roadsides, and hiking trails. Despite the broad distribution of *A. unicolor*, there have been no extensive formal studies on the ecology or life history of *A. unicolor*. This study aims to establish methods for studying *A. unicolor* that could be later used for more comprehensive ecological studies of this and other mygalomorph spiders. Proposed future studies include *A. unicolor* growth rates, life spans, recruitment, gene flow, and colony delineation. This study was focused on a single site for observation and data collection. The entire site was 31.0 m<sup>2</sup> and was located on a slope leading down into a creek in a deciduous forested area. The site was visited between August 30th, and October 7th, 2021, for approximately 1.5 hours weekly/biweekly. This time was spent locating, tagging, and measuring the diameter of *A. unicolor* burrows. Notes on door status were taken for each burrow opening to track activity of each burrow’s occupant. Primary findings from this project include measurements of average burrow size, burrow density, and activity trends.

P286 - Characterization and Analysis of Mycobacteriophage Cain's Genes

Laela Walker, Jessica Morgan, Dallas Nivens, Gabrielle Walker, Kathryn Kohl, Victoria Frost *Winthrop University, Rock Hill, SC*

The majority of bacteriophage genes have no predicted function, which provides a wealth of opportunity for investigation, characterization and potential downstream applications. The work described here includes, and extends, the research initiated by Winthrop University students as part of the SEA (Science Education Alliance) GENES (Gene-function Exploration by a Network of Emerging Scientists) program, sponsored by the Howard Hughes Medical Institute (HHMI). Our phage of interest is a K6 sub-cluster mycobacteriophage; Cain, which has a genome consisting of 100 diverse genes. To gain clues to their functions, the workflow began by cloning each of Cain’s genes individually using a shuttle vector (pExTra) and amplifying in *Escherichia coli*. Each pExTra + gene was then transformed into its bacterial host *Mycobacterium smegmatis*. A phenotypic assay then enabled the observation of host growth inhibition after inducing the expression of each phage gene individually. As a result of our work, 17 of Cain’s genes have demonstrated this cytotoxic trait which infers some sort of interaction with the host’s proteome. All cloned genes were concurrently sequence verified by comparing against the published Cain genome using *in silico* NCBI sequence alignment tools and SnapGene Viewer software. Truncating variants of individual cytotoxic genes are currently being created, in an attempt to elucidate whether, and which, protein domains influence the gene product’s activity. Our group is also performing Bacterial-2-Hybrid (B2H) assays to investigate the interactions occurring between a phage gene product and its host’s proteome, in more detail. Elucidating specific interactions at the phage-host interface may provide opportunities to exploit previously unknown proteins or manipulate the outcome of the microbial co-existence.

P287 - Comparing divergent plastid genomes in *Campanula americana* at a structural level using Minion long read sequencing

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Genetic incompatibilities are thought to be one of the first reproductive barriers to arise in speciation. Although genetic incompatibility is often assumed to occur between nuclear genes, it can also occur between nuclear and cytoplasmic genes, possibly driven by cytonuclear coevolution. Typically genetic incompatibility has been studied in relation to sequence level divergence (SNPs). However, structural variants could also play a role. To examine the role structural variation may play in genetic incompatibility we characterized plastid genome structural variation in a plant species that exhibits strong cytonuclear incompatibility, *Campanula americana*. *C. americana* has three genetic plastid lineages, Western, Appalachian and Eastern, which when crossed with each other exhibit a range of cytonuclear incompatibility, with the Appalachian and Western lineages being most incompatible. While SNP level divergence between these lineage’s plastid genomes has been well documented, and correlates with strength of incompatibility, the extent of structural variation remains unknown. Across most land plants the plastid genome is typically conserved, but *C. americana* is part of the Campanulaceae family, which is known for its highly complicated plastid genomes containing large amounts of repetitive DNA, which may facilitate rearrangements. We sequenced and assembled multiple Western and Appalachian populations using a combination of short (Illumina) and long-read (Minion) sequencing. Structural variation was then assessed by comparing gene order and content as well as identifying insertions/deletions and repeat expansions/contractions.

P288 - Introduction of the Bacterial-2-Hybrid Assay into the SEA-GENES workflow.

Jessica Morgan, Dallas Nivens, Laela Walker, Gabrielle Walker, Victoria Frost  
*Winthrop University, Rock Hill, SC*

Increasing our understanding of bacteriophage-host interactions is crucial, especially as advances in the use of phages therapeutically looks likely. As a part of the SEA (Science Education Alliance) GENES (Gene-function Exploration by Network of Emerging Scientists) program, we have identified 17 genes in *Mycobacterium* phage Cain that cause the inhibition of growth of its bacterial host; *Mycobacterium smegmatis*. In order to explore this interactive effect further, a Bacterial-2-Hybrid (B2H) assay has been implemented to help characterize protein-protein interaction (PPIs) between the phage and host’s expressed genes. Each of the previously cloned phage genes were transferred from pExTra, into a new vector; p2Hα. Using p2Hα enabled the expression of our gene of interest as a protein fused to a subunit of RNA polymerase. The p2Hα + gene was transformed into a specially engineered strain of *Escherichia coli*, called B2H SELECT (designed by the lead scientists of the SEA-GENES program at HHMI).

This cell line contains a library of random fragments of the *M. smegmatis* genome, housed in another plasmid; pCI. As the *M. smegmatis* gene fragments are expressed, they will be fused to the repressor protein CI. Following induction, both our phage gene of interest and the *M. smegmatis* gene fragments, are expressed as independent fusion proteins. Phenotypic indicators monitor pairwise interactions between the proteins using an engineered test promoter, reporters and genetic selectors. In this way we can assess PPIs between an individual phage gene product and up to a million different host cell protein fragments. pCI plasmids are sequenced to reveal the host's gene product (s) that is associated with our phage gene product. Consequently, the interactive partnerships between a bacterial host and its viral parasite reveals clues to phage gene function, and may lead to a number of beneficial applications in science.

P289 - Microsatellite marker development in Bloodroot (*Sanguinaria canadensis*)

Megan Clements, Matt C. Estep

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*Sanguinaria canadensis* L. (Papaveraceae) is an herbaceous perennial that produces a brilliant white flower, has a distinct leaf shape, and a redorange rhizome that is the basis for its common name - Bloodroot. It has been used by Native Americans to treat a range of ailments, although claims to its usefulness in current medicinal and medical practice have been debated. This potential to possess powerful medicinal alkaloids could encourage overharvesting or exploitation of wild populations before we fully understand *Sanguinaria*'s role in the understory environment as well as its genetic diversity and population structure. The first steps in marker development include low pass sequencing of the genome. Followed by identification of microsatellite loci within the sequence data. Primers can then be designed to amplify these loci. These steps are then followed by screening a diverse selection of individuals to identify polymorphic loci that can be useful in diversity studies. Effective conservation strategies rely not only on knowledge of endangered or rare species but of thriving species as well. A better understanding of common Appalachian natives can provide a more effective framework for making conservation decisions, especially for those species that possess potential medicinal properties.

P290 - Inhibitive effects of Tyr-15 and Tyr-45 on biofilm in *B. subtilis*

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Biofilm production by pathogenic bacteria generated downstream of quorum sensing has been shown to cause multiple health risks in humans. We generated six potential quorum sensing compounds (Gly-15, Phg-15, Pro-15, Trp-37, Tyr-15, and Tyr-45) with the goal of reducing biofilm production and avoiding bacterial resistance to antibiotics. A common trend among the inhibitors was the presence of a benzene ring or a strong electronegative molecule. We also included non-polar amino acid and carboxylic acid groups. Each drug was tested for its ability to decrease biofilm production in *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus mutans* using crystal violet staining. Furthermore, secondary assays assessed bacteriostatic, and bactericidal effects of the novel compounds. A 63.2% inhibition was calculated for Tyr-15 in *Bacillus subtilis* while Tyr-45 yielded a 91.3% and 44.5% percent inhibition in *Bacillus subtilis* and *Staphylococcus aureus*, respectively. Retesting and repetition of crystal violet staining, and some secondary assays are required to assert Tyr-15 and Tyr-45's viability as good biofilm inhibitors. Furthermore, based on this data, we are conducting further studies on Try-15 and Try-45 to assess the importance of stereochemistry of the amino acid and whether the drugs elicit cytotoxic effects in mammalian cell culture.

P291 - Prediction of temperature-sensitive regions of hemagglutinin protein from avian influenza using residue interaction network methods.

Soul Atkins, Jeddidiah Griffin

*Mars Hill University, Mars hill, NC*

Avian influenza has killed millions of chickens and has had serious economic impact. One of the strains of avian influenza, H5N1, is of particular concern. This study investigated the structural biology of the hemagglutinin 5 (H5) protein using a residue interaction network approach. Previous studies have shown temperature sensitivity of the virus, and our aim was to uncover potentially temperature-sensitive regions of the H5 protein. The B-factor (BF) of amino acid residues is associated the flexibility of a region, indicated regions that are likely to be more temperature-sensitive. Betweenness is a network metric that identifies residues that are important for a residue interaction network, indicating structural importance. If a residue region has a significantly high BF and a significantly high betweenness metric, then this indicates protein regions that are both temperaturesensitive and important for maintaining protein structure. Several structures had a significantly high BF in the area of residues 485-510. Preliminary evidence suggests that structures may also have high betweenness values for these regions. This study tentatively concludes that residues 485-510 of the H5 protein may contribute to the temperature sensitivity of the virus.

P292 - Mitochondrial Genome Sequencing via Genome Skimming in Crayfish

Paul Cabe

*Department of Biology, Washington & Lee University, Lexington, VA*

Mitochondrial gene sequences have been used extensively for taxonomic and phylogenetic research. In particular, complete or partial cytochrome oxidase I, large (16S) and small (12S) ribosomal RNA genes and cytochrome B have all been widely used. Notably, a portion of cytochrome oxidase I has been used in DNA barcoding studies in animals. Many studies use multiple mitochondrial markers to provide increased resolution. A logical endpoint would be to use entire mitochondrial chromosome sequences, but traditional approaches (PCR amplification of overlapping sections coupled with Sanger sequencing) are time consuming and costly. With the precipitous drop in sequencing cost on the Illumina platform, genome skimming offers an attractive and cost-effective alternative. In this approach, low coverage whole genome shotgun sequencing is performed, and a variety of bioinformatics approaches allow reconstruction of complete or nearly complete mitochondrial genomes. We performed a proof-of-concept trial of this approach with North American crayfish (Cambaridae), using one individual from two different genera (*Cambarus*, *Creaserinus*). Sequencing produced between 25-30 million paired end reads of 150 bp, providing ample coverage to confidently reconstruct all coding regions of the mitochondrial genome (the more variable control regions were reconstructed with lower confidence). As a bonus, we were also able to reconstruct the nuclear ribosomal RNA gene region, including the 28S, 5S, 5.8S sequences plus internal transcribed spacer regions and several histone gene sequences. These sequences have also been successfully used in taxonomic and systematic studies, providing a wealth of useful sequence data easily, quickly, and at reasonable cost.

#### P293 - Analysis of Thor gene structure in multiple *Drosophila* species

Noah Van Heusden

*Western Carolina University, Cullowhee, NC*

The Thor gene encodes the eukaryotic translation initiation factor 4E binding protein (EIF4E-BP) and is located downstream of Tor in the insulinlike signaling pathway. This protein functions by regulating cell growth, responses to environmental stress, and 5' cap mRNA translation by inhibiting EIF4E. In this project, we are studying the Thor gene structure in multiple *Drosophila* species to improve existing annotation and understand its genomic evolution. Using BLAST alignments, RNA-Seq data, Top Hat splice junction analysis, etc., we investigated the changes in exon-intron boundaries, sequence homology, isoform variance, and syntenic structure across *Drosophila* species. While researching the synteny of the Thor gene, we observed that in *D. melanogaster* it is found in a single copy between the Pgant4 and CG15414 genes. By contrast, within in the *D. virilis* clade, which includes species from *D. busckii* to *D. grimshawi*, there are two tandem functional copies of the Thor gene, with the genes Pi3K21B and Stip1 located downstream. In *D. willistoni*, there is the presence of only one copy of the Thor gene and the same downstream changes as in the *D. virilis* clade. This suggests an evolutionary path of gradual changes in the Thor gene neighborhood. The Thor gene in *D. melanogaster* spans a genomic length of 1179bp and has two isoforms with two exons each, differing only in the 5'UTR. We have found that this structure of the Thor gene is primarily conserved among the different *Drosophila* species studied so far. However, in the more distant species, there appears to be a three amino acid insertion within the first coding exon. We would like to determine whether this insertion has any effect on the overall Thor protein structure. In the future, we would like to identify the transcription start sites and motifs in the promoter region of the isoforms. P295 - Exploring genetic diversity in yaupon holly (*Ilex vomitoria*), an American native beverage plant

Ben Long, Jeffery Bennetzen, James Leebens-Mack

*University of Georgia, Athens, GA*

Despite their unique physiology and ubiquity in human culture, beverage plants have received little attention from the genetics community. Yaupon holly (*Ilex vomitoria*) is a southeastern American native plant that has been used by local Indigenous peoples to brew a caffeine-containing infusion for thousands of years. Its unique phytochemistry, wide natural range, and observed variation in secondary metabolite profiles would make it an excellent system to study how the production of beverage-relevant chemicals is shaped by evolution, but the genetic architecture of these traits remains unknown. To address this gap, I am genotyping 300 individuals sampled throughout the species' native range and propagating cuttings from each to construct the first duplicated diversity panel for yaupon holly. A forthcoming chromosomal genome assembly for *I. vomitoria*, together with metabolomics data, will enable genome-wide association studies and quantitative trait locus mapping for secondary metabolite production. Ultimately I will characterize diversity in caffeine biosynthesis pathway genes, as well as genetic and genomic components of variation in caffeine production and other important horticultural traits. Completion of these objectives will bring us a step closer to unraveling the complex relationship between ecology, genetics, and plant secondary metabolite diversity.

#### P297 - Adding populations to our understanding of Genetic Diversity in *Liatris helleri* Porter (Asteraceae)

Jennifer Belk, Matt C. Estep

*Appalachian State University, Boone, NC*

*Liatris helleri* Porter (Asteraceae) is a federally listed rare Southern Appalachian perennial herb. This species is restricted to high elevation rock outcrop communities in Western North Carolina. An understanding of the genetic diversity and population structure of *L. helleri* will provide valuable information for future management activities. Building on past research, two further populations were sampled and genotyped utilizing 12 microsatellite markers. These populations include the type locality as well as an A-ranked population from the north western portion of the species range. Genetic diversity estimates for these two populations will be presented. Implications of this data on land management and comparisons to populations with existing diversity estimates will also be discussed.

#### P298 - Isolating and Characterizing the Host Range of Bacteriophage Found in Davidson, NC

Emma Heiderscheit, Catherine DuPuy, Virginia Heiser, Dave Wessner

*Davidson College, Davidson, NC*

As fears continue to grow around the rise of antibiotic-resistant bacteria and “superbugs,” we have investigated bacteriophage as a viable and potentially more effective solution to cure infection. Although the therapeutic use of bacteriophage was explored extensively during the 1950s in Soviet-era Russia, it has most recently gained Western popularity as a potential treatment option. Though bacteriophage’s specificity of hosts often allows them to kill one malevolent bacterial target without further detriment to the microbiota, this same specificity makes it extremely difficult to achieve phage therapy on the same scale as antibiotic treatment, which tends to indiscriminately kill bacteria. Many bacteriophages are known to only infect a few strains of bacteria, permitting a limited range of treatment, and lessening its practical application. The discovery of phage with broad host ranges can allow for more efficient, widespread application of phage therapy. Currently, we have isolated several bacteriophage strains that appear to infect multiple natural (environmental) isolates of *E. coli*. Thus far, we have successfully isolated several bacteriophages, characterized their host range, and examined their efficiency of plating.

P299 - Genetic assessment of Familial Hemiplegic Migraine Type 1-associated CACNA1A variants of uncertain significance in *C. elegans*

JoAnna LaPoint, Ashley Turner

*Jacksonville State University, Jacksonville, AL*

General migraine disorder is one of the most common neurologic disorders, affecting more than 1 billion individuals each year across the world. The exact cause of the underlying migraine can be complex and multifactorial with both genetic and environmental factors at play. Familial hemiplegic migraine type 1 (FHM1) is a rare but very severe form of migraine with patient symptoms including photophobia, hyperacusis, unilateral weakness, mental retardation, epilepsy, and cerebellar degeneration. FHM1 is caused by pathogenic mutations in the CACNA1A gene. The CACNA1A gene encodes for the alpha protein subunit of voltage-gated calcium channels. We examined evolutionary conservation analysis of FHM1-associated missense variants of uncertain significance (VUS) in human (CACNA1A) and nematode, *Caenorhabditis elegans* (unc-2). Using Benchling, 12 VUS obtained from ClinVar were examined through multiple sequence alignments and 2 were identified to be conserved across all species. Poly-Phen 2 analysis also revealed I1707T to be likely pathogenic (HumDiv score 0.988). Primers were designed to amplify the VUS region in *C. elegans* unc-2. Future experimentation includes polymerase chain reaction (PCR) experiments to optimize the primer pair, CRISPR RNA guide design to target unc-2, CRISPR-Cas9 microinjection to generate the VUS-unc-2 *C. elegans* model, and phenotyping the VUS model. If the VUS does impact function of unc-2, then it is expected to impact nematode movement and sensitivity to light and sound. This study will provide *in vivo* assessment of this FHM1-associated VUS shedding light on its potential clinical significance for humans.

P300 - Occurrence of Kanamycin-Resistant Bacteria is Greater in Commercial and Residential Sections of Richland Creek in Nashville, Tennessee

Jolene Mach, Brandon Ramirez, Chris Barton, Darlene Panvini

*Belmont University, Nashville, TN*

The overuse of antibiotics has caused an increase in antibiotic resistant (AR) bacteria, which is a serious public health concern. Previous studies showed a significant correlation between anthropogenic pollution and AR bacteria. This project identified AR bacteria in Richland Creek relative to local anthropogenic pollution. Water samples were collected at four locations along Richland Creek in Nashville, Tennessee. The four locations represented a neighborhood, commercial area, greenway, and park. iTree Canopy was used to assess levels of impervious substrate surrounding each location. Bacteria resistant to the antibiotic kanamycin were isolated from the water samples, identified to genera using DNA barcoding, and compared among the sites. Kanamycin is an antibiotic used on patients that may suffer from a severe bacterial infection and is usually a last line of defense antibiotic. Fourteen genera of kanamycin resistant bacteria were identified with only one genus, *Aeromonas*, found at all four locations. There was greater abundance and diversity of kanamycin-resistant bacteria at the head of the creek, most likely due to higher levels of human activity in the surrounding commercial and residential areas. This research contributes to the knowledge of the occurrence and diversity of AR bacteria species present in the stream in different areas of Nashville and has public health consequences if the disparities in the distribution of AR bacteria correlates to human activity and/or socioeconomic differences along the stream.

P302 - DNA Barcoding for Identification of Kanamycin-Resistant Bacteria from Playgrounds of Varying Socioeconomic Status in Nashville, Tennessee

Remah Hatem, Chris Barton, Darlene Panvini

*Belmont University, Nashville, TN*

Antibiotic resistance is a universal public health issue associated with infections that are harder to treat, prolonged hospital stays, higher medical costs, and increased mortality. Previous studies suggest that antibiotic resistance in some locations is linked to the socioeconomic status of the area. In this study, three Nashville metro playgrounds from areas with lower socioeconomic status were compared to three playgrounds from more affluent areas. Swabs were collected from playground equipment benches, handlebars, and slides at each site. The bacteria collected were cultured

in LB+ Kanamycin media at 37°C to isolate kanamycin-resistant bacteria. Then the 16s rDNA region was amplified using Polymerase Chain Reaction (PCR) to prepare samples for sequencing and species identification using BLASTN. Results showed kanamycin-resistant bacteria were more prevalent in less socioeconomically inclined areas than in more affluent areas, as supported by previous studies. There was also a significant growth of fungi, with a similar pattern between socio-economic status as observed with the bacteria. Sequencing results showed more bacteria were common between the economic statuses than were different. People living in areas of lower socioeconomic status may experience greater negative health effects due to poor sanitation, limited healthcare access, and/or higher rates of antibiotic misuse.

P303 - Evaluating the Effects of Antimalarials on Gene Expressions in HCT-116 Colorectal Cancer Cells

Lydia Heron-Goar, Chris Barton

*Belmont University, Nashville, TN*

Evaluating the Effects of Antimalarials on Gene Expressions in HCT-116 Colorectal Cancer Cells

P304 - Screening for Antimicrobial Activity of Metabolites Produced by Entomopathogenic Fungi Isolated from North Carolina Soil

Catalina Rengifo Duque, Hannah Walsh, Michelle Thomas

*Campbell University, Buies Creek, NC*

Entomopathogenic fungi are used worldwide as alternatives to chemical pesticides in agriculture and produce secondary metabolites, including Destruins (Dtx). Dtx produced by *Metarhizium anisopliae* are known for their insecticidal, phytotoxic, and anticancer activities. The focus of this study was to identify isolated fungi from blueberry shrub soil at Hannah Forest Farms (34°47'31.388" N, 78°12'32.955" W), North Carolina, and to characterize their entomopathogenic and antimicrobial activity. Isolation of entomopathogenic fungi was performed through the insect bait method using *Galleria mellonella* followed by pathogenicity assays. *Galleria mellonella* were introduced onto 3-day old fungal plates and upon death were surface sterilized with 1% Na-Hypochlorite. The time course for death and emergence of fungi were followed. Spore arrangements suggested that the isolates belong to the genera *Nomuraea*, *Gibellula*, and *Metarhizium*. As a control, identities of *Beauveria bassiana* and *Metarhizium anisopliae* (MetMaster) were confirmed. Antimicrobial activity was investigated by overlaying fungal colonies onto 1.5x108 CFU/mL of *E. coli*, *P. putida*, *B. subtilis*, *E. aerogenes*, *E. raffinosus*, *P. aeruginosa*, and *A. baylyi*. Antimicrobial activity of secondary metabolites appeared to vary between media type and organisms present on the plate. To optimize fungal secondary metabolite production temperature and incubation time were varied and assayed through antimicrobial activity. Secondary metabolites were obtained through ethyl acetate extractions and characterized by 1H NMR and mass spectroscopy providing evidence for Dtx presence. This work furthers our understanding of the antimicrobial properties of metabolites from these entomopathogenic fungi and will allow for future screening of the secondary metabolites for anticancer activity.

P305 - Investigating shade tolerance and phenotypic plasticity of Virginia spiraea (*Spiraea virginiana* Britton), a federally threatened shrub

Rose Dickson

*University of North Carolina at Asheville, Asheville, NC*

*Virginia spiraea* (*Spiraea virginiana* Britton, Rosaceae) is a shade-intolerant, disturbance-adapted, riparian shrub species native to the southern Appalachian Mountains. This species was listed as threatened in 1990, and a recovery plan was developed in 1992. Current reassessment of the recovery plan includes proposals for propagation and outplanting to supplement and restore wild populations. Without flooding disturbance, competing vegetation needs to be actively managed to reduce shade for spiraea to thrive. Genotypes with greater shade acclimation would likely have higher survival in natural populations, would require less frequent shade-reduction management, and could be integrated into populations that need restoration. In summer 2022, we examined photosynthetic characteristics (maximum light-saturated photosynthetic, dark respiration rates, quantum yield, light compensation point, and pigment concentrations) and the ability to respond to sunflecks (photosynthetic induction and loss) of cloned propagules from five different source populations along an artificial light gradient (100%, 75%, 50%, and 20% of full sun) in a common garden. Data were compared among light treatments and source populations using ANOVA or non-parametric tests. Light treatment had significant effects of maximum photosynthesis, dark respiration, specific leaf mass, and light compensation, but not quantum yield, pigment concentrations, or sunfleck utilization. Source population did not have a significant effect on any parameter. The five source populations were all from the same river drainage (New River, Ashe County, NC), and subsequent studies have shown little genetic difference among individuals within the same drainage. Higher genetic variability has been shown between drainages. Future studies should examine photosynthetic characteristics of individuals from different river drainages in order to attain measurements from genetically contrasting source populations. More work needs to be done to understand the species' plasticity and acclimation potential under a wider range of environmental conditions to help develop a plan for successful recovery of *Virginia spiraea* in wild populations.

P306 - Using small herbaria and floristic surveys to identify conservation needs for rare, threatened, and endangered plant species in western Maryland.

Kayla Connor, Clara Thiel

*Frostburg State University, Frostburg, MD*

Small herbaria are of great importance because they contribute to knowledge regarding plant distributions and occurrences. Specimens housed within Frostburg State University's Herbarium can also highlight unknown or forgotten collections of rare or at-risk plant species. These collections may provide useful information in the prioritization of resurvey efforts of previously unknown populations of rare, threatened, or endangered species. Thus, our objective is to utilize herbarium specimens to identify previously undocumented occurrences of these species in western Maryland by resurvey efforts. These resurveys will help fill knowledge gaps surrounding the unknown presence of ecologically significant and rare flora in western Maryland. Western Maryland is a significant geographical survey area due to being settled within the biodiverse mountains of Appalachia. Our findings will help inform on conservation and restoration efforts to preserve rare flora populations as well as continue long term monitoring projects.

P307 - Growth of *Platanthera dilatata* Orchid Seeds with Different Nitrogen Sources

Sofia Baez, Lisa Wallace

*Old Dominion University, Norfolk, VA*

Orchids use their relationships with mycorrhizae in the soil around them to facilitate germination in the wild. In the lab, the seeds can be germinated in petri plates using an agar-based medium supplemented with elements like carbohydrates, nitrogen, potassium, and phosphorus to replicate the nutrients they receive in the wild. In this experiment, the germination of seeds of a temperate terrestrial orchid, *Platanthera dilatata*, was measured over time to evaluate the effect of different nitrogen sources on their growth. Varying nitrogen sources were tested because although most orchid species respond to environmental factors at the germination stage, few of these species have been studied in-depth to identify their specific responses to these factors. Two sources of nitrogen, sodium nitrate and ammonium nitrate, in two concentrations were added to BIM-1 medium, and seeds were plated. Additionally, seeds sampled from four populations were tested to look for intraspecific variation in germination response. Early germination results throughout all populations indicate contamination from seed or airborne sources is a persistent problem with asymbiotic germination, but the plates with ammonium nitrate seem to have far less contamination than the plates with sodium nitrate. Furthermore, germination seems to advance at a faster rate in certain populations than others, and in some populations, rate of germination over time seems slower because of the increased number of empty testas. This work is important because it adds to our understanding of orchid seed germination and how species respond at different life stages to a constantly changing environment.

P308 - Transcription and Digitization Initiatives at the Marshall University Herbarium (MUHW)

Erica Edinger, Jacob Webb, Pamela Puppo

*Marshall University, Huntington, WV*

The Marshall University Herbarium (MUHW) houses the second largest plant collection in West Virginia, consisting of approximately 52,000 specimens. From these, 99% of the vascular plants were digitized as part of the SERNEC (Southeast Regional Network of Expertise and Collection) initiative and are available at the online SERNEC portal, which presently hosts 233 herbaria from 14 southeastern states. To allow researchers and the public to search and view information about the specimens however, it is necessary to transcribe the specimens' labels from the images into the database. Currently, only 40% of MUHW specimens present in SERNEC have been transcribed. Additionally, some of the specimen images require re-digitization, as in some cases the image might have been cropped, resulting in the online page not showing the entirety of the sheet and cutting off portions of the label that are necessary to provide complete transcriptions. During the fall of 2022, the authors transcribed a total of 1,261 specimen labels and re-imaged 250 specimens from the vascular plant collection present at MUHW. The work done last fall exemplifies how undergraduate students can significantly contribute to the curation and accessibility of natural history collections.

P309 - Taxonomic trends in anatomical distribution of anthocyanins within autumn leaves

Alyson Acquard<sup>1</sup>, David Lee<sup>2</sup>, Mason Heberling<sup>3</sup>, Howard Neufeld<sup>4</sup>, Cam Webb<sup>5</sup>, Nicole Hughes<sup>1</sup>

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Red anthocyanin pigments are produced by many species during autumn in the temperate zone. However, it remains unclear why certain species exhibit anthocyanins and others do not. Furthermore, very few researchers have surveyed the anatomical location of anthocyanins in autumn leaves. This research is part of a larger, collaborative project aimed at understanding the taxonomic and anatomical patterns of anthocyanin distribution within autumn leaves. Red leaves of nearly 300 species of herbs, shrubs, and trees were sampled at 13 field sites spanning Alaska to Florida throughout autumn. Fresh leaves were sectioned and viewed with a light microscope, and anthocyanin anatomical location (i.e., upper epidermis, palisade mesophyll, spongy mesophyll, and/or lower epidermis) was scored. We present here examples of taxonomic trends in anthocyanin

Agnes Scott College, Decatur, GA

At Agnes Scott College, we are currently in the process of digitizing our herbarium. We have two herbarium cabinets of specimens. Currently, 400 specimens have been digitized using a Canon EOS R camera with UHD 4K resolution. We estimate that there are around 2,000 specimens total in our collection. The oldest specimens we have digitized thus far are 100 years old, from 1923. Some specimens have more data associated with them than others, but an ideal collection would have the collector's name, date collected, geospatial data (often including road, city, or county information), GPS coordinates, elevation, a site description, a specimen description, the family, genus, and species. So far, we only have GPS coordinates for 5 specimens that were collected in 2003. We do not expect to have many specimens with GPS data because a lot of the collection is older than easily portable GPS technology. Our setup for digitization includes a Canon camera mounted above a flat surface with lights on either side. Each photo includes a small ruler and color swatches for scale. Our goal once everything has been digitized is to publicize this data via the Southeast Regional Network of Expertise and Collections (SERNEC) allowing the scientific community to more easily access our collection.

P311 - Exploring the relationship between nitrogen limitation and leaf reddening during fall in *Quercus phellos*

Sarah Smith, Samantha Ethier, Corinne Gumpman, Nicole Hughes

*High Point University, High Point, NC*

The functional significance of autumn leaf coloration is an issue currently debated by plant physiologists and evolutionary biologists. Plant physiologists argue that red anthocyanin pigments evolved in trees adapted to cold and bright conditions in autumn, and function in photoprotection, while evolutionary biologists argue that red coloration coevolved as a signal to insects choosing host trees during autumn (e.g. aphids) that the tree is poor in quality. In a recent paper we argued that N-deficiency (known to induce leaf reddening in many species) should simultaneously increase the need for photoprotection *and* render leaves low-quality to insects, making both hypotheses simultaneously viable. However, so far the inverse relationship between leaf N and autumn reddening has only been demonstrated in maples. The objective of this study was to use *Quercus phellos* at two different field sites naturally exhibiting varying degrees of reddening, to test the hypothesis that individuals with redder leaves have lower leaf N, and also fewer leaf parasites (measured as number of female lecanium scale insects  $\text{cm}^{-1}$  on over-wintering twigs). For each field site, at least 14 established individuals in sun-exposed parking lots around the Piedmont of North Carolina were sampled. Three south facing branches were collected from each tree and all their leaves were excised during the summer and fall. Soil was also collected beneath each crown in four location within a 1 m radius of each trunk for nutrient analysis. Leaf pigments were quantified using spectrophotometric assays. Additionally, we measured leaf mass per unit area and diameter at breast height of each individual. Data collection is currently underway. P312 - The Vascular Floras of Marion and Lamar County, Mississippi

Allison Wilson, Amos Adams

*University of Southern Mississippi, Hattiesburg, MS*

Vascular plant surveys of Lamar and Marion Counties, Mississippi, are ongoing projects that started in the summer of 2021. The significance of these studies is the documentation and identification of the floristic richness of the counties, which are located in the longleaf pine belt of the Gulf Coastal Plain and are dissected by the Pearl River. Historically, the state of Mississippi has been botanically understudied. Only 11 of the 82 counties in Mississippi have had vascular floras completed. These floras will provide a database for future researchers, conservationists, and land managers while describing the county's various habitats, species diversity, and unique plant communities. Thus far, approximately 177 families, 562 genera, and 1,609 species have been found through field collections and previously collected herbarium specimens. Of the 1,609 species found, 429 are county records. Most notably, in Lamar County *Pinguicula primulaefora* (S3/G3G4), *Macranthera flammea* (S3/G3), and *Echinacea purpurea* (S3/G4) were found; in Marion County *Carex gravida* (S1S2/G5), *Solidago flaccidifolia* (S1/G5), and *Schoenoplectus etuberculatus* (S3/G3) were found. The three largest families within both Lamar and Marion Counties are Asteraceae (156), Cyperaceae (113), and Poaceae (137). Due to both

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histology observed in this study. This information could be valuable for helping to understand the function of anthocyanins in autumn leaves. P310 - Agnes Scott College Herbarium Digitization Project

Isabelle Grovenstein, Jennifer Kovacs

<sup>1</sup> *UNC Chapel Hill, Chapel Hill, NC*, <sup>2</sup>*University of North Georgia, Oakwood, GA*

*Packera obovata* is one of the 75 species in the genus *Packera*. *Packera* is known to have imprecise boundaries between species and cryptic species complexes due to overlaps in vegetative and floral characters between species. *Packera obovata* is widespread across eastern North America, but prefers mafic soils where it is more commonly found. It can be differentiated from congeners with overlapping distributions by the following characters: obovate, serrated basal leaves that are up to twice as long as they are wide, single or loosely clustered stems that are sometimes tomentose, and yellow ray flowers measuring 8-10 millimeters long. With such a wide distribution, there is a high likelihood of regional variance across these populations of *P. obovata*. This paper uses morphometric analyses to evaluate vouchers from the NCU Herbarium that represent the range of *P. obovata* across the southeastern states of Georgia, Alabama, North Carolina, South Carolina, Florida, and Arkansas. Vegetative and floral characters were used in the morphometric analyses to observe any potential trends between populations. Morphometric analyses were performed on the measurements collected from the NCU Herbarium vouchers including: PCoA, nMDS, and cluster analyses. These were used to identify notable variance in local *P. obovata* populations. The objective of this study is to more accurately define *P. obovata*'s physical traits and identify differentiating factors for potential taxonomic revisions.

counties' unique topography, geographic location, hydrology, soils, and various habitats, there are likely more than 1,800 species. P313 - Morphometric Analysis of the *Packera* Obovata Species Complex

Joshua Williford<sup>1</sup>, J. Brandon Fuller<sup>2</sup>

P314 - A Vascular Plant Survey of the Uchee Creek Forever Wild Tract, Russell County, Alabama

T. Wayne Barger<sup>1</sup>, Michael Wayne Morris<sup>2</sup>

<sup>1</sup>State Lands Division, Natural Heritage Section, Alabama Department of Conservation and Natural Resources, Montgomery, AL, <sup>2</sup>Troy University, Troy, AL

The Uchee Creek Forever Wild Tract (UFW) encompasses approximately 1,916 ha in the Chattahoochee River watershed of Russell County, Alabama. Due to the recreational opportunities across diverse habitats, the tract was purchased by the Forever Wild Program on July 7, 2017. Situated in east-central Alabama, three Level IV ecoregions of the Southeastern Plains converge near the tract: Fall Line Hills, Southern Hilly Gulf Coastal Plain, and Flatwoods/Blackland Prairie Margins. Natural plant communities range from upland pine-oak-hickory forests, to mesic deciduous forests, and longleaf pine-turkey oak sandhills. Pockets of calcareous glades, expansive bottomland hardwood forests, bay swamps around springheads, beaver impoundments and associated marshes, seepage areas, and tupelo gum swamps are also present. Many areas of uplands are currently sand pine and loblolly pine plantations and will be gradually converted to longleaf pine sandhills. Ruderal habitats including greenfields, cutover woods, and roadsides are frequently encountered across the tract. Trips to document and voucher plant specimens were initiated March 2019 and are currently ongoing. To date, a total of 761 taxa from 406 genera and 128 families have been collected; approximately half (377 taxa) are considered county records. Species of note include the following: *Brickellia cordifolia* (G3/S2), *Carex socialis* (G4/S1), *Corallorrhiza odontorhiza* (G5/S1), and *Hexastylis harperi* (G4/S2). Currently, the families with the greatest number of taxa present are: Asteraceae (97), Poaceae (89), Cyperaceae (63), and Fabaceae (53). Approximately 12% of the taxa are considered non-native to North America.

P316 - Patterns of commonness and rarity in the Andean tree flora

Ellen Quinlan, Miles Silman

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The high diversity of tropical Andean forests is characterized by many rare species which turnover rapidly along steep gradients. These species are thought to have narrow altitudinal ranges (< 500 m) but may span hundreds to thousands of kilometers latitudinally. Additionally, the high tree diversity is thought to be largely driven by high congeneric turnover and niche partitioning, with many genera containing >10 species within just a few thousand meters. Yet, while some have assessed these patterns at local scales, no study to date has quantified them Andes-wide. We assembled the data from over 200 long-term forest plots spanning the Andes from Venezuela to Argentina and calculated the altitudinal and latitudinal range for each species and genus. Within a few highly diverse focal clades (i.e. *Weinmannia*), we assessed how niche partitioning may vary at broad vs. local scales. Finally, we assessed the relationship between altitudinal range, latitudinal range, and abundance, and used Rabinowitz's (1981) seven forms of rarity to classify the ways in which Andean trees are rare. Preliminary analyses show 38% of species are common (with wide altitudinal and latitudinal ranges as well as high local abundance), while 5% of species are rare in all three categories. The goal of this project was to better characterize the distributions of Andean trees, information crucial to understanding the origin, maintenance, and co-existence of Andean diversity, as well as any conservation effort.

P318 - The effect of ventilated shelters on tree seedling growth within a restored riparian site in Virginia.

Christina Henriksen, Samuel Ventura, Camden Brewer, Elizabeth Stanley, Jared Gordon, Emmaline Clark, Bruce Wiggins James

*Madison University, Harrisonburg, VA*

Riparian forests perform valuable ecological services such as improving water quality, encouraging ecological diversity, increasing carbon sequestration, and decreasing stream temperature. Large-scale agricultural practices intensify nutrient loading into the watershed causing a decline in water quality. Restoring riparian forests offsets negative effects of agricultural land use. Restoration techniques, such as protecting planted trees from herbivory with plastic tree shelters, have been utilized by programs such as the Conservation Reserve Enhancement program (CREP) for 20 years. Some studies suggest trees within un-ventilated shelters benefit from elevated temperature and humidity while other studies indicate that adding ventilation (holes) to tree shelters may result in the accumulation of 55% more biomass than trees grown in un-ventilated controls. The purpose of this study is to quantify the effect of ventilated shelters on seedlings. In the spring of 2022, 162 tree seedlings (*Liriodendron tulipifera*, *Platanus occidentalis*, *Amelanchier canadensis*, and *Asimina triloba*) were planted on an active cattle farm in Linville, VA. Seedlings were planted in ventilated and non-ventilated tree shelters. Field data were collected in the spring for initial height and diameter measurements and at the end of the growing season in the fall of 2022. Chlorophyll content and stomata density were also collected at the end of the growing season. Internal tree shelter temperature and relative humidity was collected three times at random throughout the growing season. Preliminary results demonstrate seedlings grown in unventilated shelters develop significantly less chlorophyll (p-value <0.001) and experience markedly higher overall levels of

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relative humidity ( $p < 0.05$ ), than seedlings in ventilated shelters. No significant differences in seedling height, stem diameter, stomata density, or shelter internal temperatures were found. Correlation between internal shelter climate, shelter type, and plant health could potentially lead to the development of more specific guidance when creating riparian restoration planting plans which utilize protective tree shelters.

#### P320 - Influences of Leaf Inclination and Orientation on Winter Gas Exchange in *Yucca filamentosa*

Alan Huff<sup>1</sup>, Howard Neufeld<sup>2</sup>

*Yucca filamentosa* is an evergreen C3 plant native to the southeast that has long narrow leaves displayed in a radial arrangement. Newly produced leaves are steeply inclined, but become more horizontal with age, often bending and drooping downwards after a period of cold weather. Inclination and radial orientation may have important consequences for light interception and gas exchange, but this has not been studied in this species. Gas exchange characteristics during colder portions of the year are similarly understudied. The goal of this project was to investigate the gas exchange of *Yucca filamentosa* plants located on the Appalachian State University campus during the winter season. Measurements were made on northfacing leaves of different ages using a portable gas exchange system (either Li-6400 or 6800). Maximum rates of photosynthesis were  $10.3 \mu\text{mol m}^{-2} \text{ s}^{-1}$  for leaves of the same orientation on different plants in October. Results from instantaneous measurements and light response curves show that rates of photosynthesis decline as leaves age. Among north facing leaves on one plant, we found maximum rates of  $13.2 \mu\text{mol m}^{-2} \text{ s}^{-1}$  for a non-bent leaf facing up by  $45^\circ$ ,  $5.5 \mu\text{mol m}^{-2} \text{ s}^{-1}$  for a leaf bent down at  $51^\circ$  below the horizontal, and  $7.8 \mu\text{mol m}^{-2} \text{ s}^{-1}$  for a leaf bent down to  $45^\circ$  below the horizontal. We also made measurements of leaf temperatures for leaves in sun and shade. Those in sun can be several degrees warmer than ambient air, which may facilitate gas exchange during cold weather. In the future we will measure dark-adapted fluorescence to determine the degree of photoinhibition due to the combination of cold temperatures and high light. These results will help determine the ecophysiological strategies that this species, normally associated with warmer and drier climates, uses to survive at high elevations in the mountains.

#### P322 - Poultry litter as a potential fertilizer for industrial hemp

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The usage of industrial hemp (*Cannabis sativa*) has become increasingly popular over the years due to its versatility. Though there are many uses for industrial hemp, the most common are fiber, seeds, and biomedical applications. Industrial hemp and marijuana (both classified as *Cannabis sativa*) were considered illegal substances for many years under the Controlled Substance Act of 1970. However, the Agriculture Improvement Act of 2018 authorized the production of hemp and removed hemp and hemp seeds from the Drug Enforcement Administration's (DEA) schedule of Controlled Substances. This took effect on January 1st, 2019. Waste management is a major issue within agriculture and the U.S. is the second largest poultry producer in the world. Compost and animal manure has been used as a fertilizer source for generations due to the potential benefits it poses on soil health. Animal manure has been identified as a great potential source in hemp production, but only a few studies have tested this notion. This study evaluated the usage of poultry litter as a fertilizer for hemp and compared the height in growth between it and an organic commercial fertilizer (Dr. Earth). Our results indicated that the commercially available organic fertilizer had a trend of higher overall growth but was not statistically significant when compared to poultry litter. This demonstrates that poultry litter serves as an attractive alternative to organic commercial fertilizer for hemp production. Due to the recent spike in fertilizer costs and its limited availability because of supply chain delays, identifying locally sourced, alternative fertilizer options for producers is especially needed. As one of America's newest commodities, this work is designed to provide producers insights into the usage of organic fertilizers (i.e., poultry litter) towards commercial production of hemp.

#### P324 - Seed Structure Differences and Germination Trials between *Seymeria cassioides*, *S. pectinata*, and Putative Hybrid, *S. cassioides* x *S. pectinata* (Orobanchaceae)

William Merritt, Benjamin Gahagen

Abraham Baldwin Agricultural College, Tifton, GA

Members of *Seymeria* (Orobanchaceae) are root hemiparasites that mainly favor pines as hosts. *Seymeria cassioides* and *S. pectinata* have a distribution that spans the Southeastern United States, with *S. cassioides* being the more widespread species. While no evidence of natural hybridization occurs, there are distribution and flowering time range overlaps, with *S. pectinata* in peak flowering approximately a month earlier

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Learning how to use new programs and softwares can be challenging, especially for students or researchers who are new to a particular field. Often, researchers are left reading many published papers to determine not only which analyses to run, but also how to run those analyses. In fields such as population or conservation genetics, many of these frequently used programs are dated, sometimes difficult to run, and/or require input file formats that are not currently supported, making these analyses challenging for the novice. In response to this issue, graduate students from an Ecological Genetics course at the University of Memphis generated a GitHub tutorial page detailing how to run commonly used population genetic analyses including genetic variation, population genetic structure, tests for deviations from Hardy-Weinberg Equilibrium, isolation by distance, etc. The purpose in designing this tutorial and repository was to establish a “one-stop-shop” for anyone looking to familiarize themselves with

than *S. cassiodoides*. Additionally, both species appear in longleaf pine ecosystems so it could be possible for a hybridization event to occur. The purpose of this project is to artificially hybridize *S. pectinata* and *S. cassiodoides*, collect the seeds, observe external seed structure of two species and putative hybrid, and conduct germination trials. Pollen was collected from opened *S. pectinata* flowers in September 2022 from a local population in SE Georgia. In October 2022, developing, unopened flowers of a local *S. cassiodoides* population were prepared by removing connate corolla and adnate stamens (unopened). Subsequently, pollen from *S. pectinata* was brushed onto the stigma of *S. cassiodoides* flowers and the flower node was tagged. Seeds of *S. pectinata* (November 2022), *S. cassiodoides* (December 2022), and putative *S. cassiodoides* x *S. pectinata* hybrid (December 2022) were collected, lab dried, and stored at 4°C for future observation and testing. Preliminary results indicate that the putative hybrid favors external seed structure of the *S. cassiodoides* parent plant and germination trials will commence shortly.

#### P326 - PawpawPulation Genetics: An online population genetics tutorial for beginners

Jennifer Mandel<sup>1,2</sup>, Michael Charles<sup>2</sup>, Heather Clendenin<sup>2</sup>, Erika Moore-Pollard<sup>1,2</sup>, Paige Murin<sup>2</sup>, Matthew Pollard<sup>2</sup>, Avery Tucker<sup>2</sup>

population genetic methods and for those considering what analyses to use in their population or conservation genetics research. The designers also had in mind that upper-level undergraduate courses could make use of the repository. Each of the ten how-to pages use a publicly available microsatellite dataset of the North American native pawpaw, *Asimina triloba*, as input for each tutorial demonstration. The ten tutorials are open access and users can follow the how-to pages with the pawpaw data provided or input their own data. Input files, including example file formats, and the resulting output files are included with each method tutorial along with a summary of the results. This GitHub tutorial page is a great tool to learn population genetic methodologies in the classroom or for general research purposes.

#### P328 - Perceptions of Conflict Between Religion and Evolution in Undergraduate Biology Students

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Evolution is one of the most important, yet controversial topics in biology. Even though evolutionary theory is key to our understanding of the diversity and complexity of life, many people are reluctant to accept it, including biology students. Given that undergraduate biology students are our future scientists and science instructors, their perceptions of conflict between religion and evolution have the potential to impact how they communicate about evolution to their religious peers and teach evolution to their students in the future. While perceived conflict between religion and evolution has been measured previously, much of the work is centered on the conflict seen in religious students, leaving the perceptions of conflict in non-religious students relatively unexplored.

We developed a novel measure capable of assessing the levels of perceived conflict between religion and evolution in both religious and nonreligious students and administered the survey to over 11,000 undergraduate biology students in fourteen states.

Factor analysis revealed two distinct measures within the instrument, one for perceived conflict between religion and evolution and one for perceived compatibility. Atheist students perceived more conflict and less compatibility between religion and evolution than Christian students suggesting that they may perceive more conflict and less compatibility than is actually reflected by their religious peers. Non-religious students may benefit from evolution instruction that models religious cultural competence to help them become more effective communicators about science and religion as future scientists, teachers, and healthcare professionals.

#### P330 - Use of COPUS to Characterize Teaching Techniques in Biological Sciences Department

Tamera Hutchinson, Stephanie Mathews

*Campbell University, Buies Creek, NC*

Undergraduate education at this institution is a system designed to graduate students with exemplary academic and professional skills who are prepared for purposeful lives and meaningful service. The instructional techniques used to educate these students significantly impacts classroom engagement as well as the quality of learning. This research aims to accurately characterize the practiced instructional techniques regarding their range and frequency as they are observed in the undergraduate biology courses at this university. Faculty and adjunct professors from the Biological Sciences Department were observed using the Classroom Observation Protocol for Undergraduate STEM (COPUS) tool, and their instructional techniques were recorded using the Generalized Observation and Reflection Platform (GORP) collection system. The observation data was presented to instructors as a tool to reflect on instructional techniques. Their response to the observation data was collected to determine if it encouraged faculty to consider instructional practices and their effectiveness.

#### P332 - Teaching Human Anatomy and Physiology to Undergraduate and Graduate Students Using Virtual Reality

Nathan Hutcheson, Carmen Carrion

*Agnes Scott College, Atlanta, GA*

Virtual reality (VR) is increasingly being utilized to teach human anatomy and physiology (HAP) in both undergraduate and graduate student programs. Our aim is to test the effectiveness of VR to teach HAP to undergraduate and graduate students.

In this pilot study, we will use two different, stand-alone, non-tethered VR units to teach undergraduate and graduate students taking HAP labs. Students will be given pre- and post-test questions to assess the effectiveness of VR usage on learning. Assessment scores will be calculated using 24 questions; 6 questions per each of the 4 VR modules. The questions span across categories of anatomy, physiology, and three-dimensional representations. An assessment score will be calculated by (post-score minus pre-score). We will conduct an ANOVA to investigate the effect of organ module (ie, cardiovascular/respiratory vs digestive/nutrition), VR usage (VR vs no VR), and class rank (undergraduate vs graduate) on assessment scores. An alpha-level of 0.05 will be used. Additionally, we have collected qualitative data by asking a series of 6 open-ended survey questions to gauge students' views of the VR experience and how the VR related to their overall learning experience. Thematic analysis will be used to qualitatively assess students' responses.

For the pilot analysis, we project a total of 70 students to complete the study to account for an ~10% attrition rate. VR was associated with significantly higher assessment scores. We hypothesize to see an overall significant ANOVA with follow-up testing revealing that VR usage is associated with significantly higher assessment scores.

## BBB Talks

### BBB Talk 1 - Biofilm Inhibition in *Escherichia coli* and *Pseudomonas aeruginosa* using Small Molecules as Competitive Inhibitors

Evan Floyd<sup>1</sup>, Anjali Patel<sup>2</sup>, Shelby Blanchard<sup>1</sup>, Tori Glazier<sup>1</sup>, Matthew Simmerman<sup>1</sup>

<sup>1</sup>Mercer University, Macon, GA, <sup>2</sup>Mercer University Department of Biology, Macon, GA

In the world of modern medicine, the issue of antibiotic resistance has become more of a problem than ever before. Because of biofilm's role in assisting the incidence of antibiotic resistance, and its association with various health conditions, there arises a need for studying the inhibition of biofilm formation as an alternative to traditional antibiotic treatment. Recent studies have found that bacteria utilize small auto-inducers to communicate between planktonic cells in a process known as quorum sensing. These signals trigger metabolic cascades that culminate in the production of biofilm. By mimicking the structure of these auto-inducers, our team was able to synthesize and develop a library of 220+ compounds that were used to screen for biofilm inhibiting activity in *Escherichia coli* and *Pseudomonas aeruginosa*. Through triplicate confirmation in Crystal Violet testing and various secondary assays, multiple drugs were found to successfully inhibit biofilm formation in these species without affecting the growth rates of the bacteria. Notably, compounds possessing a benzylic positioned nitro group have been particularly favorable in inhibiting biofilm in both species. These compounds could be applied to clinical settings to help stave off the worsening issue of antibiotic resistance by making bacteria more susceptible to host immune responses and to existing antibiotic treatment.

### BBB Talk 2 - Examining Long-Term Antimicrobial Effect of Modified Titanium on *Candida albicans* for Use in Titanium Implants

Katherine Freel<sup>1</sup>, Deborah Roh<sup>1</sup>, Parvin Fathihshejani<sup>2</sup>, Masoud Mahjouri-Samani<sup>2</sup>, Sahar Hasim<sup>1</sup>

<sup>1</sup>Mercer University, Macon, GA, <sup>2</sup>Auburn University, Auburn, AL

Pure titanium and titanium alloys are widely used for biomedical implants, such as dental implants. Its mechanical properties, low toxicity, and biocompatibility make it an ideal material for implants. Titanium and its alloys also have the ability to inhibit the growth of cells and biofilm formation. Despite this antimicrobial property, there is a high risk of implant failure due to the development of biofilms and infections from several species of bacteria and yeast. Therefore, finding a titanium alloy that further inhibits cell adhesion and biofilm growth is important. This study examines the long-term biofilm inhibition of *Candida albicans* on modified titanium with a coating of titanium disulfide (TiS2). Cell adhesion and viability are measured by counting the number of cells that have attached to the material surface after 6, 24, and 48 hours of incubation. Extracellular Reactive Oxygen Species (ROS) created by cells is measured using 2',7'-dichlorofluorescein diacetate (DCFH-DA) assay. Finally, after removal from the titanium plates, the cells' morphology is examined through mannan and chitin staining, by ConA and Calcofluor white, respectively. The results show that modified titanium samples are effective in inhibiting *C. albicans* growth, suggesting that TiS2 coated materials may be a promising solution for preventing infection in titanium implants and other medical devices.

### BBB Talk 3 - Analyzing the role of Hydrogen Peroxide on Honey's Antibacterial Mechanism

Jonah Dennis

Birmingham Southern College, Birmingham, AL

Rising rates of antibiotic resistance among common pathogenic bacteria has prompted increased interest in natural antimicrobial compounds and substances. One historically used natural antimicrobial is honey. It is hypothesized that honey's antimicrobial mechanism stems from a variety of biochemical compounds such as: 5-hydroxymethylfurfural, flavonoids, carbonyl compounds, and hydrogen peroxide in junction with nonpathogenic bacteria capable of outcompeting pathogens. This research analyzes the impact of hydrogen peroxide honey's antimicrobial properties and assess the ability of honey-based bacteria to outcompete pathogenic bacteria. Three honeys were chosen: manuka, avocado, and alfalfa honeys. Manuka honey is a non-peroxide honey and was chosen due to this fact. Avocado honey is exceptionally dark in color while alfalfa is a very light honey. A darker color is an indicator of high levels of biochemical compounds. Concentrations of H<sub>2</sub>O<sub>2</sub> were determined using an AmplexRed coupled

kinetic assay using a standard curve. Honey borne bacteria were used in a competition assay against known escape pathogens on nutrient agar. It was found that honey borne bacteria were minimally effective against the escape pathogen and that the concentration of H<sub>2</sub>O<sub>2</sub> in avocado and alfalfa honey was sufficient for bactericidal effects to occur. Manuka honey is bactericidal as well yet its mechanism remains unknown.

#### BBB Talk 4 - Soil Solutions: Identifying Novel Sources of Antibiotics from Environmental Bacteria

Jacob Hiatt, Will Roque, Hunter Sjobom, Jalen Garner *Catawba*

*College, Salisbury, NC*

Antibiotic resistant bacterial infections are becoming alarmingly common and are predicted to be the cause of the next major global health crisis. However, over the past thirty years, very few novel antibiotic therapies have been approved for human use. As such, alternative sources of these antimicrobial drugs are desperately needed. Many soil-dwelling microbes naturally produce antimicrobials as a strategy for survival and resource competition in their environments. Hence, soils represent a prominent and important source for antibiotic discovery. Recently, we have isolated and characterized several antimicrobial-producing bacteria from terrestrial soil samples, including an environmental strain of *Pseudomonas putida*.

We found that this organism inhibits the growth of several ESKAPE relatives, laboratory bacterial strains that are related to many notable antibioticresistant pathogens. Interestingly, many *Pseudomonas* species are known for their ability to produce heterocyclic antibiotics. We are performing a forward genetic screen using *P. putida* in an effort to determine which biosynthetic gene may be responsible for its antimicrobial activity. Loss-of-function mutants generated by this screen will also be evaluated using metabolomic analysis to better characterize the compound of interest.

#### BBB Talk 5 - Investigating the bacterial species isolated from commercial probiotic sources

Makenzie Bonner

*Catawba College, Salisbury, NC*

The human gut microbiome is a complex and incredibly important part of human anatomy and physiology. The human microbiome plays a crucial role in maintaining the body's immune system, preventing gut permeability, and supporting the metabolism of nutrients. As important as these are, it is equally important to maintain these bacterial populations. One way to increase bacterial diversity is to ingest probiotic supplements from various sources including pills, yogurt, and other fermented foods. Probiotic sources contain live bacteria and yeast that have potential health benefits. Our goal is to identify the antibiotic substances produced by commercial probiotic supplements and beverages, including Kombucha. In our initial results, we isolated several probiotic species from a commercially available brand of Kombucha. We grew these cultures under physiological conditions that mimic the acidic environment of the stomach during digestion. Initial characterization of our isolates determined that they appeared to be gram-positive bacilli and several produced zones of inhibition against other species in the mixed culture. We will further examine the antibacterial efficacy of our isolates against ESKAPE relatives. The ESKAPE relatives are genetically similar to major antibioticresistant human pathogens like Enterobacteria, Escherichia, and Enterococcus species. To determine the identity of our isolates, we will perform 16S ribosomal sequencing. We are hopeful that the probiotic species isolated from the sources that we test may provide a new source of antibiotic that could work against antibiotic-resistant human pathogens.

#### BBB Talk 6 - The effect of dual photosensitizer-mediated photodynamic therapy on glioblastoma cells

Jamie Solis

*Mary Baldwin University, Staunton, VA*

Glioblastoma (GBM) is the most common form of primary brain cancer, making up over half of all cases. Despite this, there is a lack of new and individualized treatments, due in part to the challenge of chemotherapy drugs being able to pass through the blood-brain barrier. Photodynamic therapy (PDT) is a cancer treatment that involves inducing photosensitivity in cells by allowing them to absorb a special kind of dye called a photosensitizer, then using light exposure to cause cell death. The majority of studies have explored how using a single photosensitizer to mediate PDT affects cells. Multiple studies have shown that photosensitizers activated by longer wavelengths of light allow for better penetration into cells. Another method for combatting this problem is to use two photosensitizers to mediate PDT. In studies using this method, a dual photosensitizer system was more effective than a single photosensitizer. The goal of this study was to explore the effect of a different combination of photosensitizers than those studied previously. The two photosensitizers used were indocyanine green (ICG) and rose bengal (RB) at concentrations of 23.25 $\mu$ g/mL and 16.7 $\mu$ g/mL, respectively. There were groups with each photosensitizer alone and a negative control group. Cells were treated for 18 and 24 hours, then a trypan blue exclusion assay was performed to measure viability. Based on the knowledge that photosensitizers activated by longer wavelengths of light are more effective, I hypothesized that the photosensitizer activated by long wavelengths will perform better than the photosensitizer activated by shorter wavelengths. The differences between the control, RB, and ICG groups were not significant. The average viability of the control group and ICG group were higher at 18 hours than at 24 hours. However, the average viability of the RB group was higher at 24 hours than at 18 hours.

#### BBB Talk 7 - The effects of Sodium Lauryl Sulfate on KRT1 Gene Expression in *Pimephales promelas*

Reina McCullough

*Francis Marion University, Florence, SC*

Sodium lauryl sulfate (SLS) is an anionic surfactant found in many household and personal hygiene products such as toothpaste, shampoo, and laundry detergent. It's used in food as an emulsifier, and it's used in agriculture for crop production. SLS works by lowering the surface tension between two materials using its amphiphilic properties; it binds to proteins and disrupts their quaternary or tertiary structure. The National Institute for Occupational Safety and Health (NIOSH) warns against releasing SLS into the environment as it is toxic to aquatic organisms. Studies conducted on humans found that SLS can cause irritation in the skin, and when combined with warm water it is more irritating. Cytokeratin is a protein found in epithelial cells all over the body. It functions to help form tissues such as hair, nails, organs, and gills in fish. Studies demonstrate that SLS exposure in hamsters causes changes in KRT1 gene expression in cheek epithelia. The disruption of cytokeratin gene expression in fish could be detrimental to their ability to respire as gill epithelia are essential for absorbing oxygen and releasing carbon dioxide. This project aims to investigate the potential dangers SLS has on developing fish by evaluating cytokeratin (KRT1) gene expression in *Pimephales promelas* after exposure to SLS. Larval (24 hours post-hatch) *Pimephales promelas* were exposed to low, medium, and high concentrations of SLS over the first four weeks of their development. Survival and growth were measured to determine any overtly toxic effects of SLS treatments. Morphological defects such as spinal curvature, cardiac edema, and craniofacial abnormalities were also recorded. In addition, we cloned a section of the KRT1 *Pimephales promelas* mRNA and ran qPCR to determine KRT1 gene expression. Our data lend insight into the toxic effects of SLS on developing fish.

**BBB Talk 8 - Mycosporine-like Amino Acids: A Vital Defense Mechanism Against Ultraviolet Radiation in the Painted Sea Urchin *Lytechinus pictus***

Abigail Adams

*Mary Baldwin University, Staunton, VA*

Eggs and embryos of many marine organisms develop in the water column where they can be exposed to ultraviolet radiation (UVR) causing potentially detrimental effects. UV-induced damage impacts embryogenesis because the cell division cycle and cleavage periods are brief, preventing the use of mitotic checkpoints to correct unrepaired DNA. The present experiment was performed to determine the effects of UVR on the early stages of development in the sea urchin *Lytechinus pictus* and whether dietary accumulation of photoprotective compounds, mycosporinelike amino acids (MAAs), reduce UV-induced damage. Eggs from adults on an MAA-rich diet exhibited darker coloring (yellow orange), rather than the noticeably lighter eggs from adults on an MAA-poor diet. Embryos from adults fed an alga lacking MAAs and exposed to UVR exhibited more structural abnormalities, than those from adult urchins fed an alga rich in MAAs. Morphological abnormalities included thickening of blastoderm walls, decreased blastocoel size, blastocoels filled with abnormal cells, exogastrulation, and abnormal spicules. Daily exposure to UVR caused developmental delays and abnormalities, which were not found in nonirradiated embryos. The percentage of *L. pictus* embryos that developed normally was lower in those exposed to UVR, compared to embryos from adult urchins maintained on the MAA-rich diet. The presence of MAAs were found in fertilized eggs, blastulae, gastrulae, prism, and pluteus larval stages in embryos from adult urchins on MAA-rich diet. The identification of MAAs in *L. pictus* embryos suggests that these compounds protect them against UV-induced abnormalities, to at least the fourarmed pluteus larval stage.

**BBB Talk 9 - Analyzing the function of Fus1 during cell fusion in *Saccharomyces cerevisiae***

Chloe DeYoung

*Stetson University, DeLand, FL*

Cell fusion is a ubiquitous process in human life, occurring in events such as fertilization and skeletal muscle development, however, the mechanisms behind cell fusion are understudied. The budding yeast, *Saccharomyces cerevisiae*, undergoes cell fusion during mating and is an excellent model to study cell fusion due to its ability to exist as a haploid or as a diploid, its high doubling rate, and its genetic tractability. Yeast mating involves pheromone release from the opposite mating partner, which leads to cell polarization enabling cell-cell contact. The cell walls are then remodeled at the fusion site allowing plasma membrane fusion to occur. Fus1 is a protein in *Saccharomyces cerevisiae* that localizes to the fusion site and is known to promote cell wall degradation. Fus1 has a C-terminal SH3 domain that has been hypothesized to regulate protein interactions required for fusion, however, this function has not been directly shown. To explore the function of the SH3 domain of Fus1, we created multiple mutations in the coding sequence including a full SH3 deletion, as well as the point mutations H469A and W473A within the SH3 domain. Through quantitative mating assays using a fluorescent dye that allows for visualization of the membrane between mating pairs, we found that Fus1-H469A and Fus1-W473A caused severe fusion defects. These proteins were also mislocalized in polarized cells, potentially explaining their fusion phenotype. Interestingly, the more dramatic complete SH3 deletion caused a less severe fusion defect despite the protein also being. We hypothesize that important protein interactions occur within the SH3 domain that have been altered in our point mutations. Continued assays will involve a high-copy suppressor screen on these mutations as an unbiased method to examine the mechanism of the fusion defect.

**BBB Talk 10 - Triple Negative Breast Cancer: Investigating the role of the Hippo Pathway in Cellular Proliferation**

Victoria Peterson

*Mary Baldwin University, Staunton, VA*

Triple Negative Breast Cancer (TNBC) is a type of breast cancer that does not express estrogen, progesterone, or HER2 receptors. TNBC is the most aggressive form of breast cancer with a low survival rate and with treatments that cause patients with this type of cancer to suffer. Estrogen, progesterone, and HER2 receptors help control cell growth and are also the most common targets of breast cancer therapy. The lack of these receptors makes it hard to control cell growth in TNBC and adds to the difficulties of manufacturing TNBC therapies. The Hippo pathway is a pathway that controls organ size through the regulation of cellular proliferation and apoptosis. YAP is a transcriptional coactivator that works to induce the expression of genes that promote cell proliferation, survival, and migration. Population-based genomic studies have shown that activating Hippo signaling correlates with TNBC breast cancer risk, whereas high YAP expression is associated with poor survival in breast cancer patients, suggesting Hippo/YAP plays a critical role in breast cancer progression. Staurosporine is a kinase inhibitor that affects the activation of the Hippo Pathway in breast cancer cell lines. This research aims to evaluate an opening for control of cellular proliferation in TNBC cells through the Hippo pathway. Can Staurosporine identify a target for therapy that reduces cellular proliferation? Here we examine the activation of the Hippo pathway in MDA-MB231, a TNBC cell line, with Staurosporine administration. RT-PCR (real-time) will be used to evaluate if the expression of CCN1 and CCN2, genes regulated by the Hippo Pathway, are affected by the Staurosporine treatment and correlate with changes in cell proliferation. Because CCN1 and CCN2 are cell cycle regulatory genes, these experiments would provide information on the control of the Hippo pathway and potentially provide another chemotherapeutic option that directly influences cell proliferation in TNBC cells.

BBB Talk 11 - Comparison of mammalian activity between zones of high vs low human traffic bordering a greenway in Raleigh, NC.

Kiera Williams, Jasper Doyle, Claire Boaz, Nancy Gamewell

*William Peace University, Raleigh, NC*

Students in Biology Department at William Peace University have been involved in a two-year study using camera traps to monitor mammalian activity along four zones bordering an urban greenway in Raleigh, NC. The zones include: 1) the paved greenway, 2) the stream bed that parallels the greenway, 3) the forested area adjacent to the stream bed, and 4) a forested corridor between two highly developed areas. Research protocols included monitoring diurnal and nocturnal activity in each zone for all mammalian species recorded on the cameras. Results showed the highest activity levels occurred in the stream bed and in the forest directly adjacent to the stream bed. There was little to no activity on the paved greenway due to extremely high human and canid traffic. Data suggests that mammals do not use convenient walkways or bridges to traverse areas, but rather undertake more risk to avoid pathways frequented by humans.

BBB Talk 12 - Relationship between beak anatomy and bite force in Northern Cardinals (*Cardinalis cardinalis*)

Alicia Deras-Cruz, Joe Poston

*Catawba College, Salisbury, NC*

Functional ecology includes the study of how an organism's traits are adaptive in its environment. Because birds use their beaks to acquire and process food (among other functions), beak shape and size has long been an example of an anatomical trait that reflects a bird's ecological niche. Birds that use force to extract a seed from its hull or shell must generate considerable force, and their deep, conical beaks provide a biomechanical advantage for generating such force. In the eastern US, one of the largest residential seed eating songbirds is the Northern Cardinal (*Cardinalis cardinalis*). Despite their large beaks, and their ability to strike fear in the hearts of bird banders due to their painful bites, there are few studies of Northern Cardinal foraging ecology and how it relates to beak anatomy, and we are aware of no study designed to investigate intraspecific variation in anatomy and feeding. We captured several adult cardinals and measured beak dimensions as well as body size. We also measured bite force with a custom-made force transducer. Similar to studies with other seed-eating birds, we found a positive relationship between bite force and size. Our ongoing study will also measure foraging behavior to see if variation in behavior is related to variation in anatomy or bite force.

BBB Talk 13 - Large Carnivore Numbers, Space Use, and Activity Times in the Sera Rhino Sanctuary of Kenya

Chris Pate

*Bridgewater College, Bridgewater, VA*

The Sera Rhino Sanctuary is a 108 km<sup>2</sup> fenced sanctuary in Kenya where black rhinos are monitored daily for protection from poachers. It is also used for the soft release of rehabilitated African elephant orphans. Large carnivores within the sanctuary could increase stress and mortality for these two endangered megaherbivore species. The goal of my research was to provide wildlife managers with information about five carnivore species within the Sera Rhino Sanctuary, some of which also are of vulnerable conservation status. These species included spotted hyenas, striped hyenas, lions, leopards, and cheetahs. My objectives were to estimate the population composition, assess the space use, and determine the activity times of each species. I examined time-stamped camera trap images taken over 3 years from 29 camera traps positioned throughout the sanctuary to accomplish these objectives. Identifying and counting individuals with techniques previously developed for each species, I identified 12 spotted hyenas, 10 striped hyenas, 2 lions, 5 leopards, and 4 cheetahs. Maps of space use revealed that there is much species overlap in sites used but suggest striped hyenas may avoid areas used by spotted hyenas. Activity times also showed overlap but suggest leopards may avoid being active at the same time as lions. Using all of this information, managers can make informed decisions to better ensure the safety of endangered species within the Sera Rhino Sanctuary. Additionally, further studies could be conducted in the sanctuary to expand on this research such as the impact of water availability

on space use and movement, avoidance behavior in solitary species such as striped hyenas and leopards, and expanding camera trap usage to the southeastern quadrant of the sanctuary to allow for further space use analysis.

BBB Talk 14 - The Effect of Floral Morphology on Pollination in Two *Lilium* Species

Abigail Schmitt

*Mary Baldwin University, Staunton, VA*

Though pollination by Lepidopterans is well-documented, pollination via the wings of Lepidopterans is not as well understood. I hypothesize that the morphological characteristics of certain flowers attract large butterflies as pollinators and the structures lend themselves to pollination mediated by the wings of the butterflies. To test this, I studied two flowers in the *Lilium* genus, *L. superbum* and *L. canadense*, to determine their most efficient pollinators and to look for a case of wing-mediated pollination. These two flowers vary little, except in their structure and color—*L. canadense* is typically bright red and has a funnel-shaped inflorescence, while *L. superbum* is bright orange and has recurved tepals which allow a landing platform for the butterflies. Field observations were conducted to record visitors to the flowers and the type of contact they made with the reproductive parts of the flower. Additionally, time lapse footage was recorded to obtain frequency data for each type of visitor, and pollen counts of collected insect specimens were conducted to determine the pollen load on the bodies of visitors. The data suggest that the most efficient pollinator for *L. superbum* is the pipevine swallowtail (*Battus philenor*), and the most efficient pollinator for *L. canadense* is the ruby-throated hummingbird (*Archilochus colubris*), supporting the hypothesis that the floral morphology of *L. superbum* lends itself to pollination by large butterflies, while the morphology of *L. canadense* does not.

BBB Talk 15 - A test of the threat sensitivity hypothesis in domestic versus wild mice (*Mus musculus*)

Masey Wright

*Mars Hill University, Mars Hill, NC*

The house mouse (*Mus musculus*) is one of the most commonly used animals in laboratory studies examining aging, health, intelligence, memory, and behaviors. The first known use of a “mouse model” bred in captivity for genetic research purposes occurred in 1902. More recent works have suggested that domesticated mice may not be an optimal choice for studies examining behaviors, because generational domestication can lead to modified behaviors that are not comparable to the “natural” behaviors of their wild counterparts. Studies examining predator-prey interactions have focused almost exclusively on the predators’ perspective or how one controls the other in terms of population. The objective of this study is to test the threat sensitivity hypothesis (prey alter predator avoidance tactics based on the level of threat perceived by the individual) in domestic and wild mice to determine if there are behavior differences. To test the hypothesis, we will compare the behavioral responses of domestic and wild caught *Mus musculus* to two escalating levels of predation risks (auditory and visual stimuli). Results of this study will contribute to best practices of when and when not to use domestic mouse models for investigating anti-predatory behaviors in laboratory settings.

## BBB Posters

### Profiling Coloring Agents in Rhizomes of *Curcuma longa* Grown in Georgia Versus Overseas

Bal Khatiwada

*Abraham Baldwin Agricultural College, Tifton, GA*

Abigail Farquhar, Josue Lopez, and Bal Khatiwada

Turmeric powder is widely used as a spice in Middle Eastern and Asian countries. One of the active ingredients of Turmeric Powder, curcumin has been widely studied for its role as an anti-inflammatory and an antioxidant properties. In this study, we are interested in profiling coloring agents present in rhizomes of Curcuma Longa (Turmeric) that are grown locally in Georgia versus overseas. The turmeric powder we purchased overseas has a brighter color as compared to the regular organic turmeric powder from local supermarket. The bright color in turmeric powder may have link to lead chromate-based dye found in other samples. We are interested in finding out the active chemical agents that produce the bright color in the turmeric powder and compare it to the rhizomes grown locally. We plan to profile metal ions present in the turmeric powder by smelting and dissolving the ashes in aqua-regia. The concentration of metal ions dissolved in aqua-regia will be determined using the Coupled Plasma-Optical Emission Spectroscopy (ICP-OES) instrument. We are also interested in finding out synthetic dyes that may have link to the bright color in the powder. We plan to extract the dye using solvent-solvent extraction and identify the dyes using liquid chromatography technique. BBB1 - Effects of Microtubule Stability on Mechanosensory Neuron Morphology in *C. elegans*

Caylin Lepak, Lillian Beavers

*Florida Institute of Technology, Melbourne, FL*

Mechanosensory neurons are responsible for sensing touch, relaying mechanical force into electrical signals. Critical to the function of these neurons are microtubules, which are involved in touch detection and act as the support system of the long axons of these cells. Destabilization of microtubules is linked to several neurodegenerative diseases, but the cellular, genetic, and molecular mechanisms involved are not well understood. Uncovering the genes involved in the complex process of neurodegeneration will be important for future diagnostics and therapeutic treatments. This study proposes to examine how different genes and pharmacological agents affect microtubule destabilization by assessing mechanosensory neurons in a neurodegeneration genetic model. The mechanosensory neurons have distinct morphological phenotypes resulting from increased or decreased microtubule stability, providing an *in vivo* functional readout of microtubule stability. Microtubule stability will be manipulated genetically by the mutation of genes encoding proteins that bind microtubules, as well as genes involved in neuronal development. Pharmacological manipulation of microtubule stability will be done by the introduction of the drugs Taxol and Colchicine. Transgenic expression of GFP in the mechanosensory neurons allows observation of neuronal morphology using fluorescence microscopy. Resultant changes in the Anterior Lateral Mechanosensory (ALM) neuron in *C. elegans* will be tracked.

### BBB2 - Identification and Sequencing of Mutant Bacterial Strains Deficient in Biofilm Production Pathways Inhibited by Novel Lead Compounds

James Haywood, Abdulraheem Kaimari, Priya Patel, Ren Thigpen

*Mercer University, Macon, GA*

Biofilm is an extracellular matrix composed of polysaccharides, polypeptides, and nucleic acids that certain bacteria produce in order to protect themselves from antibiotics and the host's immune system. Biofilms have been implicated in the complication of various diseases; examples include: *Pseudomonas aeruginosa*'s biofilm forming ability correlates with Cystic Fibrosis, *Escherichia coli* biofilms correlate with morbidity of colon cancer, and *Streptococcus mutans* biofilms link with dental caries and cerebral hemorrhaging. Biofilm formation allows for rapid proliferation of antibiotic resistant plasmids through a bacterial population, further complicating the growing problem of antibiotic-resistant bacteria. Mutant strains of clinically significant bacteria with a reduced ability to produce biofilm were isolated. Five clinically significant bacterial species—*Streptococcus mutans*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *Staphylococcus aureus*—were mutated using acridine orange, ethidium bromide, or ethyl methanesulfonate. Initial screening by colony phenotype on Congo Red resulted in hundreds of candidates. Further testing with Crystal Violet thus far revealed more than a dozen lines that have reduced biofilm formation in pathways that our novel biofilm compounds inhibit. The genomes from these lines will be sequenced. Verification of biofilm reduction in the biofilm pathways that our novel drugs inhibit will be carried out on the additional 100+ lines that have been isolated. Plans for a database for assessment will be confirmed. Identifying significant genes related to biofilm production in species of clinically significant bacteria will enhance the general understanding of how these species produce biofilm and give further insight into how to effectively inhibit biofilm production with additional novel lead compounds.

### BBB3 - Analyzing the Effect of a Point Mutation on the Plasma Membrane Fusion Protein Prm1p in *Saccharomyces cerevisiae*

Matea Susac

*Stetson University, DeLand, FL*

Cell fusion is an integral part of life and is involved in key processes such as myogenesis and zygote formation. Despite its importance, it is severely understudied. *Saccharomyces cerevisiae* has served as a model organism to understand this essential life process. Various proteins have been identified as playing some role in fusion, including Prm1p, a pheromone-induced protein suspected of being involved in plasma membrane fusion. While many proteins have been shown to regulate early steps in the fusion process in yeast, Prm1p is the only protein known to regulate membrane fusion. In previous studies, two-way *prm1Δ* mutants showed significant defects in fusion, including an inability to fuse or lysis upon contact, which may indicate Prm1 plays a role in cell membrane stabilization. While Prm1p is not a fusogen, understanding more about its mechanism may help uncover the plasma membrane fusogen, a protein that has remained elusive to this day. To analyze the role of Prm1p further, we created a point mutation in two neighboring, highly conserved residues in the extracellular loop, Prm1p-PF240AA, using site-directed mutagenesis on plasmids containing PRM1 tagged with GFP. When this plasmid was expressed as the only copy of Prm1p in yeast cells, we observed a significant decrease in cell fusion compared to the wildtype protein. When the localization of Prm1p-PF240AA was analyzed through fluorescence microscopy, we found a similar significant decrease in localization to the fusion site. This study is one of the few analyzing the roles of various domains in Prm1p, and the results may suggest that this extracellular domain may be necessary for fusion-specific interactions. Further research on this and other domains may shed light on the mechanism of Prm1p in membrane fusion.

#### BBB4 - Observing environmental challenges on the phenotypes of FAM210B mutants in *Arabidopsis thaliana*

Olivia Rizzo

*Athens State University, Athens, AL*

The FAM210B proteins are highly conserved across eukaryotes. In humans, the mitochondrial targeted FAM210B protein has been identified as having a role in iron import for heme synthesis. In addition to a mitochondrial targeted FAM210B protein, the green lineage has a FAM210B domain containing protein targeted to the chloroplast. Recently, other studies have shown that the knockout and knockdown of the chloroplastic FAM210B in *Arabidopsis thaliana* plays a role in the photosynthetic complex assembly. We hypothesize that the FAM210B proteins in the green lineage are involved in iron transport, and this affects cytochrome *c* assembly for both the *bcl* complex in the mitochondria and the *bcl* complex in the chloroplast. However, neither the knockout mitochondrial FAM210B (SALK\_082302) nor the knockdown chloroplastic FAM210B (SALK\_06267C) have displayed a discernable phenotype under standard growth conditions. To explore the importance of these genes' role in heme import, we are attempting to induce a phenotype through environmental manipulation, including the use of a low iron media.

#### P204 - Evaluation of cytotoxic and apoptotic effects of several regional plant-based extracts on human squamous cancer cell lines

Nicholas Spader

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The south has always been a place where home remedies and scientific-based medicine go hand in hand. Throughout time, natives and other groups have used homemade salves and ointments for what could have potentially been carcinoma or other forms of skin cancers. Through literature study, certain plants of interest have been identified. Plants used in various assays were either grown in the ABAC greenhouse or collected from local areas. Two extracts of interest have been used in the application of cell cytotoxicity assays, cell proliferation assays, and a cellular invasion assay using skin cancer cell lines. The cytotoxicity assay demonstrated cytoprotective properties when applied to cells. The cell proliferation assay confirmed this observation by showing increased cellular proliferation in cells treated with the extracts. The wound healing assay demonstrated increased cellular invasion into an area of artificial damage. Overall, these extracts have been shown to demonstrate increased cell survival, increased cell proliferation, and increased cellular invasion when used to treat subcutaneous cancer cells in vitro. In summary, the extracts demonstrated a prosurvival phenotype indicative of enhanced wound healing.

#### BBB5 - A Seasonal Analysis of Bacterial Antibiotic Resistance Levels in the Holston Watershed, Northeast Tennessee

Rachel Turner<sup>1</sup>, Frauke Argyros<sup>1</sup>, George Argyros<sup>2</sup>

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<sup>1</sup> *Emory & Henry College, Emory, VA*, <sup>2</sup> *Emory and Henry College, Emory, VA*

Overuse of antibiotics in medical, veterinary, and agricultural industries has led to increased discharge of antibiotics into the surrounding environment. These discharged antibiotics alter the natural microbial environment, and also pose a risk to human health. During the summer of 2022, we analyzed seasonal levels of bacterial antibiotic resistance from three recreational and two wastewater treatment plant locations within the Holston watershed of northeast Tennessee. Preliminary testing revealed all isolates are gram-negative bacteria. Following preliminary identification, microbial susceptibility testing was performed using 12 common-use antibiotics. We hypothesize seasonal fluctuation in antibiotic resistance across sampling sites, and higher resistance in downstream effluent of wastewater treatment plants. Antibiotic susceptibility testing revealed bacterial isolates (N=38) were 97.4% resistant to Bacitracin, 84% to Penicillin, 77% to Novobiocin, and 71% to Cephalothin from the sampled summer population. Additionally, 50% of the isolates were resistant to Ampicillin, 44% to Vancomycin, and 14% to Erythromycin. Intermediate levels of susceptibility were exhibited at 39% to Erythromycin, 24% to Vancomycin, 21% to amoxicillin, 13% to Chloramphenicol, 11% to Cephalothin, 8% to ampicillin, and 3% to Amikacin. Isolates exhibited only 5% intermediate susceptibility to Tetracycline, Neomycin, and Novobiocin. Of isolates tested, 90% were susceptible to Neomycin, 87% to Tetracycline, 86% to Amikacin, 74% to Chloramphenicol, and 42% to Amoxicillin.

BBB6 - Examining Cell Fusion Defects of a Point Mutation in *FUS1* in *Saccharomyces cerevisiae*

Rebekah Brawley

*Stetson University, DeLand, FL*

Cell fusion is a necessary process for events that occur throughout human development, yet the mechanisms underlying cell fusion are understudied. *Saccharomyces cerevisiae*, also known as budding yeast, is an ideal model organism to study fusion. Mating in yeast involves pheromone release, shmoo formation, cell wall remodeling and plasma membrane fusion. Fus1p is a known regulator of cell fusion that localizes to the fusion site in mating cells. Fus1p consists of 3 conserved domains: a transmembrane domain, internal domain, and SH3 domain. The transmembrane domain is the only domain with a known function, and the internal domain is yet to be studied. My research sought to better understand the function of the internal domain through analyzing the effects of a point mutation, Fus1p-L263A, on mating efficiency and Fus1p localization. The Fus1p-L263A mutation was made using site directed mutagenesis and expressed as the only copy of Fus1p in yeast cells. We found that Fus1p-L263A cells show a slight reduction in fusion compared to WT cells in a qualitative mating assay against a fusion compromised mating partner. To determine if Fus1p-L263A was localized to the fusion site, we performed a pheromone induction and examined the localization of GFP labeled Fus1p using fluorescence microscopy. Preliminary evidence shows that Fus1p appears to be similarly localized at the shmoo tip in both Fus1p-L263A and WT cells. This suggests that a single point mutation is not sufficient in affecting Fus1p's function, or that the internal domain is redundant with the SH3 domain for some protein interactions. While Fus1p-L263A did not cause severe fusion defects, future experiments, including a quantitative mating assay, are needed to examine its effects on fusion more wholly. Further work analyzing the phenotypes of other mutations in the Fus1p internal domain can help provide insight into the internal domain's function.

BBB7 - The role of genetic distance in the kin recognition responses of the model angiosperm *Arabidopsis thaliana*

Ruth Nichols, Emily Soucy, Davonya Cheek Florida

*Tech, Melbourne, FL*

Many plants have evolved the ability to distinguish between members of the same or different accession and alter their behaviors accordingly. Common responses include changes in shoot and/or root growth, nutrient acquisition, as well as exudate production, all of which can influence plant success. We hypothesize that accession recognition is modulated by the extent of genetic relatedness between the two accessions in question whenever plants of the same species are forced to interact. Prior studies have established that nutrient restriction can amplify accession recognition impacts, providing an additional tool for distinguishing between the generic and genetic relatedness models. Here we evaluate accession recognition phenotypes between several well-established accessions of *Arabidopsis thaliana*: Col-0, La-1, Aa-0, Xxx-0, and Ws-0. Young plants (<21 days) will be evaluated for changes in root length and area with the ImageJ and SmartRoot software packages to provide information about their Root Structure Architecture (RSA).

BBB8 - Testing the Utility of Herbarium Specimens as Repositories of Arthropod eDNA

Carlos Guerrero

*Reinhardt University, Waleska, GA*

Metabarcoding of environmental DNA (eDNA) has increased in popularity in recent years. Typically, eDNA is extracted from water, soil or air, and metabarcoding is used to better understand the species present in or near that particular sample area. However, the use of plant tissues as repositories of eDNA has not yet been widely explored. Our study seeks to determine the utility of herbarium specimens as sources of arthropod eDNA. To test this, DNA will be extracted from locally collected herbarium specimens and arthropod eDNA will be amplified using previously published arthropod-exclusive primers. Amplified eDNA will be sequenced to identify arthropod visitors to the plants. If successful, use of herbarium specimens for this technique will provide ample opportunities to examine plant-arthropod interactions, such as pollination and herbivory, across space and time.

BBB9 - Developing an Electroporation Method for Transforming *Streptomyces nymphaeiformis*

Heather Knott

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These data suggest antibiotic resistance in the Holston watershed is prevalent. All antibiotics tested were ineffective or only intermediately effective to some isolates tested. This is concerning due to high recreational use of these waterways during summer months. Both wastewater treatment plant sites displayed similar patterns of resistance and susceptibility between upstream and downstream samples, indicating the treatment process does not appear to alter propagation of antibiotic resistant microorganisms. Further research will compare resistance trends across the subsequent winter and spring seasons to assess impacts of seasonality.

*Streptomyces* species are notoriously difficult to transform. *Streptomyces nymphaeiformis* is no different, so a method of electroporation was used to attempt to transform the cells. Multiple growth stages were used in order to alter the degree of development of the cell wall. The procedure did not kill the cells, but the cells were not transformed. Due to the lack of transformation with *S. nymphaeiformis*, transformation was attempted on two other *Streptomyces* strains, *S. lividans* and *S. coelicolor*. Neither was successfully transformed to thiostrepton (*tsr*) resistance, nor did they grow on a plate lacking thiostrepton. One possibility for the lack of transformation and growth was flocculation of the cells in the broth after electroporation. The flocculation may have prevented the electric pulse from shocking all the cells or transferring the sample to the plate for selection was made more difficult. Finding a method to prevent flocculation may allow for successful transformation using an electroporation method.

#### BBB10 - Quorum-Sensing Inhibitors Targeting Growth and Biofilm Production in Multidrug-Resistant *Candida auris*

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*Candida auris* (*C. auris*) is a multidrug-resistant fungal pathogen that leads to significant mortality, spreads easily in clinical settings, and is challenging to identify. Biofilms are associated with increased resistance to antifungal drugs and difficulty detecting and treating infections, making it important to develop strategies to inhibit *C. auris* biofilm formation. In this study, several quorum-sensing inhibitors are designed to interact with small quorum-sensing molecules, such as farnesol, to inhibit biofilm formation in *C. auris*. Twenty-one clinical isolates of *C. auris* are co-incubated with different concentrations of cholecalciferol and novel compounds Lys-28 and Tyr-6. The minimum inhibitory concentration, growth curves, and crystal violet assays determine these compounds' biofilm inhibition and antifungal effect. Cholecalciferol shows significant growth reduction and biofilm inhibition in most *C. auris* strains. Lys-28 and Tyr-6 also display growth and biofilm reduction in a few strains. In conclusion, among these compounds, cholecalciferol acts as a promising antibiofilm agent against drug-resistant strains of *C. auris*. BBB11 - Analyzing the role of Hydrogen Peroxide on Honey's Antibacterial efficacy

Jonah Dennis

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Rising rates of antibiotic resistance among common pathogenic bacteria has prompted increased interest in natural antimicrobial compounds and substances. One historically used natural antimicrobial is honey. It is hypothesized that honey's antimicrobial mechanism stems from a variety of biochemical compounds such as: 5-hydroxymethylfurfural, flavonoids, carbonyl compounds, and hydrogen peroxide in junction with nonpathogenic bacteria capable of out competing pathogens. This research analyzes the impact of hydrogen peroxide honey's antimicrobial properties and assess the ability of honey-based bacteria to out compete pathogenic bacteria. Three honeys were chosen: manuka, avocado, and alfalfa honeys. Manuka honey is a non-peroxide honey and was chosen due to this fact. Avocado honey is exceptionally dark in color while alfalfa is a very light honey. A darker color is an indicator of high levels of biochemical compounds. Concentrations of H<sub>2</sub>O<sub>2</sub> were determined using an AmplexRed coupled kinetic assay using a standard curve. Honey borne bacteria were used in a competition assay against known escape pathogens on nutrient agar. It was found that honey borne bacteria were minimally effective against the escape pathogen and that the concentration of H<sub>2</sub>O<sub>2</sub> in avocado and alfalfa honey was sufficient for bactericidal effects to occur. Manuka honey is bactericidal as well yet its mechanism remains unknown. BBB12 - Impact of Ultraviolet Light and 3D Coated Materials on the Growth of *Candida* Species

Sahar Anis Ali, Jheel Patel, Sahar Hasim, Dorina Mihut

Mercer University, Macon, GA

Recently, advances in 3D printing have been widely used in biomedical devices due to enhanced productivity and cost-efficiency; however, these devices are associated with a likelihood of nosocomial infections as their surface is prone to microbial adhesion and biofilm formation. One of the strategies to prevent or eliminate microbial adhesion is surface modification and coating. Our current research investigates the growth and biofilm inhibition of *Candida* species (*Candida albicans* and *Candida auris*) when coated with pristine 3D-printed polymeric flexible BioMed clear resin. Various thicknesses of copper and silver are created using the magnetron sputtering system. The 3D materials coated with copper and silver are then submerged in a media containing *C. albican* or *C. auris* cells with and without ultraviolet (UV) light for eight hours. Next, the cell growth, and Reactive Oxygen Species (ROS), are measured through the colony count assay, and ROS detection kit. A significant decrease in cell viability and adhesion was shown in the presence of 100 nm copper and 300 nm silver-coated 3D printed materials. Additionally, exposure to UV light increased ROS production. This finding could be a promising way to prevent *Candida* biofilm formation on 3D implants and medical devices.

#### BBB13 - Assessing the relationship between the biofilm formation pathway and expression of collagen-binding proteins in *Streptococcus mutans* OMZ175 strains using novel competitive inhibitors.

Daniel Alejandro Mendoza, Ren Thigpen, Ji Won Park, Mary-Maria Mbanaso

Mercer University, Macon, GA

Infection with *Streptococcus mutans* is epidemiologically associated with dental caries, infective endocarditis, and cerebral hemorrhages. This range of conditions is often explained through the different virulence factors linked with varying serotypes. While serotype c is heavily associated with oral complications, other serotypes have been associated with endovascular infections. Interestingly, the groups that infect at a vascular level carry the *cnm* gene, which encodes for a collagen-binding protein. Through targeted disruption of the collagen-binding pathway, these infections may be

selectively combatted. Therefore, the current study aims to identify compounds that inhibit collagen-binding in OMZ175, a *cnmc*-carrying strain of serotype f. Over 400 compounds were produced via dehydration synthesis of 20 amino acids and 75 carboxylic acid derivatives using a modified D3 protocol. These compounds are being screened with the Crystal Violet assay for their effect on biofilm production in OMZ175 *cnm*<sup>+</sup> and *OMZ175 Δ nm*. Compound-induced inhibition or acceleration of biofilm production in OMZ175 *cnm*<sup>+</sup> is being used to select potential collagenbinding inhibitors. This criterion is based on findings correlating the *cnm* gene and the development of the biofilm matrix. Possible compounds elucidating a correlation between these phenomena are glutamate-based or phenylalanine-based drugs with acid derivatives containing a benzene ring. Following screening, potential compounds will be reassessed for their effect on biofilm production in all strains with a newly patented biofilm quantification assay derived from Congo red testing. Secondary assays (doubling time, use-dilution test) will also be performed to assess whether any observed differences are not merely due to bacteriostatic or bactericidal effects. Future studies will involve performing a collagen-binding assay to determine whether the selected compound disrupts the collagen-binding pathway of OMZ175 and fine-tuning potential inhibitors by designing and testing novel compounds in this system.

#### BBB14 - Bactericidal Effects of High-Energy Visible Light on Acute Otitis Media Pathogens

Anh Phan

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Acute otitis media (AOM) is an infection of the middle ear caused by inflammation of the Eustachian tube, preventing fluid from draining. The accumulation of fluid and mucus in the middle ear is suitable for bacteria such as *Moraxella catarrhalis*, *Streptococcus pneumoniae*, and *Haemophilus influenzae* to colonize and cause infection. There are approximately 709 million new cases of AOM diagnosed annually worldwide. AOM is mainly treated with watchful waiting, but if that is unsuccessful, antibiotics are prescribed, specifically amoxicillin. With the significant incidence rate of AOM and the amount of amoxicillin prescribed, pathogenic bacteria are developing antibiotic resistance at a high rate. Our previous studies have shown that planktonic bacteria exposed to high-energy visible light (405 nm) resulted in a 99.99% reduction in survival. In this study, I examined the effect of high-energy visible light on biofilms of these organisms. *M. catarrhalis*, *S. pneumoniae*, and *H. influenzae* were allowed to form biofilms for three days and then continuously exposed to no light (control) or high-energy visible light for up to 4 hours. Survival was quantified at each hour. Data for *M. catarrhalis* suggest that high-energy visible light is effective against biofilms of this organism. The effects of high-energy visible light on *S. pneumoniae* and *H. influenzae* biofilms are currently being studied. Understanding the impact of high-energy visible light on biofilms of AOM pathogens is important because biofilms are thought to play a significant role in AOM and are a contributing factor for antibiotic resistance in these organisms.

#### BBB15 - Investigation of the Effects of Novel Anti-biofilm Compounds on Biofilm Production in *Staphylococcus aureus* Nor-A Wild-type and Mutant Strains

Abdulraheem Kaimari, Dontavious Jones, Ryan Brownlee

*Mercer University, Macon, GA*

Quorum-sensing is a density-dependent communication mechanism utilized by bacteria to restrict specific gene expression to high cell-density conditions where collaboration is most beneficial. Once a bacterial population reaches a threshold, one intercellular response is the production of a surface-coating virulence factor called biofilm. Biofilms prevent a host's immune system from detecting and eradicating them. Recent studies indicate that efflux pumps may be involved in the process of biofilm production. These pumps are transmembrane transport proteins that extrude various substances, including toxins, quorum-sensing molecules, and biofilm components. The NorA efflux pump in *Staphylococcus aureus* is the most biofilm-related pump studied, as subsequent overexpressor (SA-1199B) and knockout (SA-k1758) strains have been developed to analyze the relationship between NorA activity levels and biofilm production. Known NorA efflux pump inhibitors (EPI), to date, cannot be used clinically due to their bactericidal and/or bacteriostatic effects, which significantly harm our normal flora. Thus, discovering novel compounds that act on the NorA efflux pump without bactericidal and bacteriostatic effects is essential for maintaining a positive symbiotic relationship with beneficial bacteria. NorA EPI structures were obtained through a literature search and were compared to the structures of 853 previously designed quorumsensing analogs. Three were chosen for further testing: Proline trans-3,5-Difluorocinnamic acid (Pro 61), Proline trans-3-nitrocinnamic acid (Pro 68), and Tyrosine trans-cinnamic acid (Tyr 16). Biofilm was stained with crystal violet and absorbances were recorded (550 nm). Disc diffusion and Use dilution assays qualitatively studied the bacteriostatic and bactericidal effects of our drugs. The bacterial growth curve assay (600 nm) studied changes in bacterial growth patterns. Minimum inhibitory concentrations were determined (μg/mL) for ideal biofilm inhibition. Pro 61, Pro 68, and Tyr 16 significantly reduced biofilm in SA-1199 and SA-1199B, but not in SA-k1758, which are similar effects of known NorA EPIs reported in the literature.

#### BBB17 - Activity of Crude Plant Extracts on an MCF-7 Breast Cancer Cell Line

Everette Rhymer, Carmony Hartwig

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The natural herb *Artemisia vulgaris* has documented antimicrobial and antitumor effects. *Artemisia vulgaris* is a part of the same genus as *A. annua*, from which the highly effective antimalarial artemisinin is derived. Despite the major advances in biomedical technologies, cancer is amongst the leading causes of death worldwide; and while effective treatments and chemotherapeutics do exist, many come with an exorbitant monetary cost and severe side effects. Another threat to global human health is antibiotic and antiparasitic resistance. The previously mentioned antimalarial

artemisinin from *A. annua* used to be the most promising drug to fight malaria infections, but in recent years resistance has emerged. Similarly, antibiotic resistance in some of our most deadly bacterial infections is on the rise globally. Therefore, it is of vital importance that we again look toward naturally-derived, novel compounds that may have activity toward these deadly pathogens. The use of crude extracts of *A. vulgaris* is an innovative way of finding the solutions to these problems. The Fred Stanback Jr. Ecological Preserve (FSJEP) at Catawba College is a highly diverse and species-rich habitat that is home to not only this natural herb, but several other plant species that have potential anticancer and antimicrobial effects. Here we discuss the antitumor effects of crude ethanol extracts prepared from *A. vulgaris*, *Poncirus trifoliata*, and *Lindera benzoin* on MCF-7 breast cancer cells, as measured through resazurin viability assays.

#### BBB18 - Acute exposure to trace metals reduces cell viability

Jalen Garner

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Trace metals are elements such as copper, iron, and magnesium that occur at very low levels in the environment. Though all living organisms need small amounts of certain trace metals for normal function, exposure to high levels can be toxic, potentially leading to dysfunction of the nervous and immune systems, the appearance of skin lesions, and carcinogenesis. While there are regulations for acceptable metal levels in food and beverage items, there are few that govern the production of ceramic tableware and serveware. However, metallic compounds are frequently used in the production of these items to enhance product durability. Here, we used human and non-human cell lines to assess the cellular effects of acute exposure to metals similar to those found in ceramics. We report that cells treated with moderate concentrations of dissolved metallic compounds, including metal sulfates and metal oxides, exhibited a decrease in viability compared to the control. Based on this finding, we are evaluating the mechanism by which metal exposure reduces viability by assessing cellular metabolic profiles, evaluating the capacity for cellular proliferation, and searching for markers of DNA damage.

#### BBB19 - The antimicrobial activity of an environmental strain of *Bacillus koreensis*

William Roque, Hunter Sjoberm, Jacob Hiatt

*Catawba College, Salisbury, NC*

In nature, microorganisms have long been competing with each other for survival. Many species produce antimicrobial compounds that target the physiological processes of competing organisms. Several of these antimicrobials have served as the basis for antibiotic therapies. However, overuse and misuse of these drugs has resulted in the increased frequency of antibiotic-resistant infections. Although it is imperative to develop effective alternatives, very few antibiotics have been approved for use since 1987. Previously, our group has worked to isolate and identify antimicrobial-producing bacteria in terrestrial soil samples. Here, we altered our approach and began screening for antibiotic-producers in aquatic and semiaquatic soil samples, as the diversity of species present is very different than that of terrestrial microenvironments. Using bioactivity-guided screening of bacteria collected from the shoreline of a freshwater pond, we observed that one bacterium, identified as *Bacillus koreensis*, inhibits the growth of several ESKAPE-relatives, laboratory bacterial strains that are genetically similar to prominent antibiotic-resistant pathogens. Although *B. koreensis* has been reported to have anti-fungal activity, it has not yet been demonstrated to have antibacterial activity. Further work was done to characterize this environmental strain of *B. koreensis*, including Gram-staining, motility-testing, and metabolic analysis. Currently, we are working to identify the gene or gene cluster responsible for antimicrobial activity by performing a forward genetic screen. Loss-of-function mutants obtained from this screen will also be used to characterize the antimicrobial compound through metabolomic analysis.

#### BBB20 - Enhancing detection of the multi-drug resistant *Candida auris* by macrophages through caspofungin unmasking of the cell wall

Bradley Akin, William dos Santos

*Mercer University, Macon, GA*

*Candida auris* (*C. auris*) is a multidrug-resistant fungal pathogen that has become a significant threat to human health. One of the challenges in detecting *C. auris* is its ability to evade the host immune system by masking its cell wall. *C. auris* cell walls are made of chitin and  $\beta$  (1-3)-glucan, masked with a layer of mannosylated glycoproteins. Caspofungin is the newest antifungal drug in clinical use to inhibit the synthesis of  $\beta$  (1-3)glucan, an essential fungal cell wall component. In this study, we focus on unmasking the cell wall of *C. auris* using caspofungin to improve detection by macrophages, immune cells that play a crucial role in recognizing and removing pathogens in the host. After three-hour incubation of each *C. auris* clinical strain with caspofungin (50 ng/ml), the level of unmasking is detected by Concanavalin A, a specific staining reagent for the mannan present in the cell wall. Next, each unmasked strain is incubated with macrophages, and lactate dehydrogenase concentrations in supernatants are measured using CytoTox 96® Non-Radioactive Cytotoxicity Assay kit. Overall, our data demonstrated primary evidence suggesting that some strains are unmasked in the presence of sublethal caspofungin concentration, which promotes the detection by innate immune cells and increases the level of phagocytosis.

#### BBB21 - Understanding how Human T-cell Leukemia Virus (HTLV) infection alters host cell metabolic signatures.

Mackenzie Martin

*Department of Biology, Catawba College, Salisbury, NC*

Human T-cell Leukemia Virus (HTLV-1) is a human retrovirus known to cause a lifelong infection which may result in the development of a fatal form of leukemia known as Adult T-cell Leukemia (ATL). While HTLV-1 is classified as an oncovirus, it is unclear how the virus causes infected immune cells to become cancerous. To date, two viral proteins have been implicated in this process: Transactivating factor (Tax) and the HTLV-1 basic leucine zipper factor (HBZ). Both of these proteins are important for different parts of the retroviral replication cycle, but HBZ is the only viral protein that is known to be produced by the virus at all stages of infection. HBZ is a viral transcription factor capable of regulating the expression of both viral genes and cellular genes. It is thought that many of the pro-cancer changes that occur in HTLV-infected cells arise from changes in cellular gene expression, due in part to the activities of HBZ. Alterations in gene expression are well documented to affect the metabolic signature of a cell. In fact, cancerous cells have metabolic profiles that are distinct from those of healthy cells. It was recently reported that HTLV activity is influenced by glucose and oxygen availability, but it is unknown how the virus impacts the metabolism of its host cell. Here, we evaluate whether HBZ expression alters cellular metabolism in a way that is consistent with the pro-survival metabolic signatures observed in other cancer types.

#### BBB22 - Effects of Ethanol on Liver Cancer Cells

Summer Dobson

*Catawba College, Salisbury, NC*

The majority of liver disease and liver cell death is due to long-term consumption and sometimes even the short-term abuse of alcohol. Alcohol is linked to cell damage in the liver as it is the primary organ for metabolism of ethanol. The goal of our research is to examine how ethanol exposure affects HepG2 human liver cancer cells. Initially, we will perform a viability assay to determine non-lethal dosages of ethanol that will be used during our study. Then, we will measure reactive oxygen species (ROS) production and DNA damage in HepG2 cells treated with varying concentrations of ethanol. Since ROS are produced during alcohol metabolism, we expect to see an increase of ROS production and DNA damage in the ethanol-treated cells. Furthermore, we will explore how ethanol exposure affects the expression of genes and proteins involved in oxidative stress/inflammation-mediated pathways, including the NF- $\kappa$ B and MAPK pathways. Our results will be important to understanding the particular molecular pathways involved in alcohol-induced cellular damage in HepG2 cells. Overall, our findings can contribute to increasing awareness of the likelihood of cancer development and other consequences of alcohol consumption.

#### BBB23 - "Evaluation of cytotoxic and apoptotic effects of several regional plant-based extracts on human squamous cancer cell lines"

Nicholas Spader

*Abraham Baldwin Agricultural College, Tifton, GA*

The south has always been a place where home remedies and scientific-based medicine go hand in hand. Throughout time, natives and other groups have used homemade salves and ointments for what could have potentially been carcinoma or other forms of skin cancers. Through literature study, certain plants of interest have been identified. Plants used in various assays were either grown in the ABAC greenhouse or collected from local areas. Two extracts of interest have been used in the application of cell cytotoxicity assays, cell proliferation assays, and a cellular invasion assay using skin cancer cell lines. The cytotoxicity assay demonstrated cytoprotective properties when applied to cells. The cell proliferation assay confirmed this observation by showing increased cellular proliferation in cells treated with the extracts. The wound healing assay demonstrated increased cellular invasion into an area of artificial damage. Overall, these extracts have been shown to demonstrate increased cell survival, increased cell proliferation, and increased cellular invasion when used to treat subcutaneous cancer cells in vitro. In summary, the extracts demonstrated a prosurvival phenotype indicative of enhanced wound healing.

#### BBB24 - Adaptation of Novel HEFY Assay for Verification of Biofilm Inhibition by Novel Competitive Inhibitors in *Candida albicans* and *Candida parapsilosis*

Matthew Simmerman, Tori Glazier, Evan Floyd, Shelby Blanchard, Jackson Sundgren

*Mercer University, Macon, GA*

Antimicrobial-resistant fungal strains have recently emerged as a major problem facing the modern world. A possible solution for this problem is the use of biofilm inhibitors to prevent or stop the formation of biofilm, which is believed to play a role in drug resistance. Biofilm production has been found to largely be regulated by quorum sensing, in which individual cells communicate with each other through small signaling molecules. By creating compounds that resemble these autoinducers, progress has been made in reducing the formation of biofilm in a variety of fungal species through competitive inhibition. However, research in biofilm inhibitors has been complicated by difficulties in accurately quantifying biofilm growth, especially in undergraduate labs. To solve this problem, the HEFY assay was developed as an inexpensive, efficient, and accurate biofilm quantification method. Using 3D-printed petri dish lids, this assay allows for easy quantification of biofilm growth on Congo Red media. The HEFY assay was previously shown to be effective in multiple bacterial species, however, its adaptation to fungal biofilm quantification required multiple adjustments because of the growth requirements of many fungal species and the sensitivity of the Congo Red indicator. By using SDA with an

adjusted pH, we were able to effectively grow and quantify biofilm produced by *C. albicans* and *C. para*. Additionally, we found multiple compounds in both species that appeared to inhibit biofilm formation. The adaptation of the HEFY assay for fungal biofilm quantification is useful because it provides for a reliable secondary or primary assay that can be used in conjunction with the Crystal Violet assay to study biofilm formation by fungal species. In particular, this is useful for undergraduate labs that seek to do research in fungal biofilm.

BBB25 - Molecular effects of BPA-induced DNA damage in MCF-7 breast cancer cells

Emily Brasseur<sup>1</sup>, Kasey McLamb<sup>2</sup>

Due to its molecular similarity to estrogen, BPA also has the potential to act as an estrogenic chemical, including in estrogen receptor-positive MCF-7 breast cancer cells. Our goal is to test the effects that BPA has on MCF-7 breast cancer cells through DNA damage and the regulation of genes and proteins involved in the cell cycle and DNA damage response pathways. Initial results confirm that BPA treatment (10mM), increased cellular proliferation significantly, as demonstrated by our experiments using a fluorescent Resazurin assay. We tested BPA-induced DNA damage via Comet assays and found that 10mM BPA induced DNA strand breaks. We further tested how BPA affected gene expression via Reverse Transcription-PCR. Genes of interest involved in cell cycle control and DNA damage repair were CCND1, CCNE1, CHK2, TP53 and ATM. We found that BPA induced expression of TP53 and ATM in MCF-7 cells. Finally, we plan to examine protein expression in control vs. BPA-treated cells using Western blot analysis. Together with our initial results showing that BPA increases MCF-7 cell proliferation and induces DNA damage, we expect an upregulation of genes and proteins associated with cell cycle control and DNA damage response pathways in the BPA-treated cells. Our potential results could show the risk to human health associated with contact and consumption of BPA on a regular basis.

BBB26 - Human Health Risks Associated with Chemical and Biological Hazards Surrounding Radium Springs

Jade Slaybaugh, Jankembreya Hill

*Thomas University, Thomasville, GA*

Radium Springs has historically been a potential health risk to the public due to the increased levels of radium-226 present in the water. This radionuclide-rich environment is potentially unsafe to swim or drink from. Areas nearby Radium Springs, GA provide a unique habitat as radium has penetrated the soil and spring water. The springs have a direct relationship with two major water systems, the Apalachicola-Chattahoochee Flint River Basin (ACF) and the Floridan Aquifer. Radium Springs connects to these two major water systems and it is hypothesized that radium contaminated water from the spring travels through the ACF and the Floridan Aquifer. These areas also serve as points for runoff such as antibiotics, fecal matter, and other environmental contaminants associated with human activities from the urban and agricultural lands nearby. This research analyzed the radionuclide, chemical, and biological properties of Radium Springs and the surrounding area. Selected sites include 1) Flint River Aquarium in Albany, GA, 2) Radium Springs Gardens, Albany GA, 3) Camilla boat launch in Camilla, GA, 4) Flint River in Bainbridge, GA, 5) reference sinkhole at Warwick, Thomasville, GA which served as a control. This study addressed water quality, radium presence and prevalence, and the microorganism community within this unique environment.

BBB27 - Quantification of Microplastic Pollutants in water with Genetically Modified M13 Bacteriophage

Aubrey Stewart, Lauren Locke

*Troy University, Troy, AL*

Microplastic pollution in lake, river, and oceans has become a big world-wide environmental concern and threat for human health. In this research project, we aim to develop a novel method of measuring polyethylene (PE) microplastic pollutants in water by using genetically modified M13 bacteriophages strongly binding to PE. Biopanning of phage display will be performed to select M13 phages with a high affinity to PE. Among the selected phages in biopanning, the best candidate phages will be further narrowed to be chosen by comparing their  $K_d$  values with PE.  $K_d$  values will be also used for calculating how many M13 phages bind to a certain amount of PE pollutant microparticles. The chosen phages will be used to form a complex with PE in water and then the phage-PE complex will be separated by centrifugation and filtration process. The difference of optical density values (at 269 nm for M13 phage quantification) before and after the PE-phage complex separation process will be used to quantify the initial amount of PE pollutant microparticles in water.

BBB28 - Severe drought decreases weevil infestation and affects masting patterns in six oak species

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Bisphenol A (BPA) is a colorless chemical commonly used in the production of plastics. Studies have found that exposure to BPA can have harmful effects in various model organisms. The main mode of BPA exposure occurs when BPA leaches into food or liquids that the plastic containers hold.

Truman Douglass

*Samford University, Birmingham, AL*

The predictions of climate change models for central Alabama include increased frequency and severity of drought. In 2016, Alabama experienced the most severe drought on record, with consecutive months of no rainfall. As a consequence, local forests experienced extended stress, dropping leaves and fruits early and abruptly, with high rates of mortality in some habitats. Notably, oak trees experienced a near total failure of the acorn crop. Oaks (*Quercus* spp.) display a system of pulsed reproduction called masting, where individuals in an area coordinate a large pulse of acorn production every 2-5 years, depending on the species, with low levels of acorn production in intervening years. Masting cycles may be an adaptation to suppress populations of seed predators, which include the specialist acorn weevils (*Curculio* spp.) that infest and feed on the inside of developing acorns, damaging the cotyledon and/or embryo and reducing seedling survival and performance. Weevil infestation rates are often high, exceeding 50 percent of acorns. We monitored acorn production and weevil infestation in 6 Oak species for 6 years after the 2016 drought. We found that acorn production was high and weevil infestation was very low in 2017, the year after the severe drought, but synchronization of acorn production among oak individuals appeared to be disrupted. Weevil infestation rates have slowly recovered in subsequent years, with some fluctuations. Oak species in the red and white oak sections have synchronized masting cycles. Severe drought may temporarily decrease weevil populations and infestation rates of acorns, potentially facilitating oak reproduction in the year after a drought.

**BBB29 - Will selective nutrients continue to drive native pollinators preference toward native plants in Cherokee National Park?**

Hannah Walker

*TriBeta-Mu Kappa Chi Chapter, Louisville, KY*

Climate change has taken a toll on the environment including native plants and their pollinators. One effect has been a shift in phenology for many native plants and insects. In addition, non-native plants are now invading that flower earlier with larger floral displays than native plants, thus affecting pollination of native plants. Shifts in flowering time of native plants may be due to climate change effects or competition for pollinators with invasive species. In this study, phenology shifts were analyzed by comparing visitations of native bees to spring beauty (*Claytonia virginica*) to an aggressive invasive plant (*Ficaria verna*). Local shifts in the flowering time of spring beauty were also analyzed using seasonal observations in an urban park in Louisville, KY and historical records. There were no conclusive results to this. No differences were found between invaded and uninvaded sites. One possible explanation is that spring beauty is a perennial plant and less dependent on pollinators than annual plants. Further study in this system is needed.

**BBB30 - An analysis of biodiversity in microbial communities over time due to heavy industrial activity in Birmingham, Alabama**

Kevin Drace

*Birmingham-Southern College, Birmingham, AL*

Prolonged industrial activity has shown to produce many significant effects on an environment. Birmingham, Alabama has a known history of heavy steel and iron production, taking a severe toll on microbial life hidden deep within the soil. Harsh conditions over time have led to increased levels of stress that these microbial communities have now had to acclimate to, likely altering both species and genetic diversity. This experiment aims to better analyze the species of bacteria collected from sites that have been exposed to toxic heavy metals, as it compares over an extended period of time. Utilizing Oxford Nanopore sequencing technology, how much microbial communities are changing in response to these new conditions will be determined. Techniques including both alpha and beta diversity will be used to analyze how both polluted and unpolluted samples have been affected by this constant industrial exposure over time.

**BBB31 - Exploring the Microbial Biodiversity of Upstate South Carolina**

Justin Leonhardt

*Lander University Psi Theta Chapter of BBB, Mauldin, SC*

Even though soil microbes dominate the biosphere, we lack knowledge about how these microscopic communities are changing due to encroaching human and animal activities. For this study, soil samples will be taken throughout upstate South Carolina at established camera trap sites. DNA was extracted from 0.25g of each soil sample using Quiagen DNeasy PowerSoil Pro Kit following the standard manufacturer's protocol. Concentrations of extracted DNA were assessed using a Nanodrop spectrophotometer to ensure successful DNA extraction. DNA samples were sent to University of Michigan Medical School's Microbiome Core for library preparation and MiSeq high-throughput sequencing. The V4 region of the 16S rRNA gene was amplified for downstream sequencing with the commonly used primers 16Sf-V4 (515f) and 16Sr-V4 (806r). Acquired DNA sequences were filtered for quality and analyzed using MOTHUR v 1.48.0 following the MiSeq SOP with modifications. Alpha and beta diversity standard metrics were used to measure the similarities between microbiome structures and compositions, while considering the human and animal activity level for the soil sample results for all of camera sites. Results have provided insights into the impact of animal and human disturbances on soil microbiomes at selected sites throughout upstate South Carolina. Understanding the composition and structure of the natural, soil microbiome may help expand the scientific knowledge of mammalian and microbial community ecology and provide site owners with useful information about the wildlife and soil health on their land.

BBB32 - Microbial Soil Biodiversity of Lander University's Campus

Jeilymar Jimenez

*Lander University, Greenwood, SC*

University campuses represent a unique environment with a wide range of microorganisms due to high human population densities and increased animal activity. After a brief literature review, it was noted that very little research has been published on the diversity and bioactivity of soil microbial communities on college campuses. Soil samples were collected throughout the college campus using convenience sampling and each soil sample was analyzed independently for microbial community analyses. DNA was extracted from each soil sample using Quiagen DNeasy PowerSoil Pro Kit following the standard manufacturer's protocol. Concentrations of extracted DNA were assessed using a Nanodrop spectrophotometer to ensure successful DNA extraction and quantification for sequence library preparation. DNA samples were sent to University of Michigan's Microbiome Core for library preparation and next generation sequencing. The V4 region of the 16S rRNA gene was amplified for downstream sequencing with the commonly used primers 16Sf-V4 (515f) and 16Sr-V4 (806r) and a previously developed protocol. Sequencing was accomplished via a MiSeq high-throughput sequencer. Acquired DNA sequences were filtered for quality and analyzed using MOTHUR 1.48.0 following the MiSeq SOP with modifications. Results were analyzed to determine the composition and structure of the Lander University soil microbiome. Additionally, species diversity was determined to better understand the ecological complexity of a single sample (alpha diversity) or between samples (beta diversity). Also, results were analyzed for the presence of antibiotic producing and antibiotic resistant organisms. This study has provided an initial characterization of the soil microbial communities collected on the Lander University college campus in Greenwood, South Carolina.

BBB33 - Monitoring water quality in Abbeville County, South Carolina - A citizen science project

Hailey Holcomb

*Erskine College, Due West, SC*

The South Carolina Adopt-a-Stream Program (SC AAS) was created to get the public involved in the monitoring and health of local waterways through citizen science. To our knowledge, Abbeville County waterways had never been monitored through SC AAS. Seven streams in Abbeville County were sampled and tested for pH, dissolved oxygen content, and E. coli levels to determine the overall water quality and health of each stream. In addition, macroinvertebrate community assessments were also taken for three of these streams. The data gathered was compared to South Carolina Department of Health and Environmental Control (SCDHEC) data and SC AAS where possible. Nearly all of the streams monitored were in or near the acceptable range of each parameter tested. Each stream will need further testing to create baseline data and paint an accurate picture of what is considered normal for that stream.

BBB34 - Mycoremediation of Mercury: Bioremediation Potential of Fungi

Alexander Gehron

*Catawba College, Salisbury, NC*

Heavy metals are a common form of environmental pollution emitted by activities ranging from burning fossil fuels to producing medical supplies. Mercury is an important heavy metal contaminant contributed by coal-fired power plants as well as other sources. In the field of pollution management, there is a promising approach to removing heavy metals from the environment through bioremediation. This study focuses on mycoremediation: using fungi to clean the environment. We will report the result of our research to measure the abilities of a few species of fungi to remove mercury from environmental samples.

BBB35 - Vertebrate activity in and around artificial rock piles in the southern Blue Ridge of Georgia

Abby Ware

*Reinhardt University, Waleska, GA*

Humans have used artificial cover such as brush and rock piles for years with the assumption that they provide benefit to animals. To create microhabitats for vertebrate animals, in 2018 we established 10 rock piles on private conservation land in northern Cherokee County, Georgia. Piles were constructed mainly out of slabs of reinforced concrete with a large central cavity made from a culvert and are found in forest stands that receive prescribed burns. In 2019, 2020, and 2022 we monitored 7 piles and 7 paired comparative sites using 15-meter drift fences with camerafitted bucket traps to count smaller vertebrates. In 2023 we monitored 5 piles and comparative sites each with wildlife cameras and pitfall traps to count mid-sized to large animals and small mammals, respectively. Data from bucket traps showed higher overall vertebrate activity at piles than at similar sites away from piles. Various taxa, including reptiles and small rodents, were more active near piles but many pictures could not be identified to the species level. The set of vertebrates counted at piles was also more species rich than the vertebrates counted away from piles, suggesting that these piles are an important focal point for activity of a wide range of vertebrates. We will also present preliminary data from pitfalls and game cameras and discuss future plans for this project.

BBB36 - The Effects of Roundup® on the Reproduction Rate and Life Span of the Freshwater Rotifer *Philodina acuticornis*

Olivia Jans

*Erskine College, Due West, SC*

Glyphosate, the active ingredient in Roundup® Weed and Grass Killer Concentrate Plus, is the most used herbicides worldwide. In the United States alone, approximately 300 million acres of crop land are treated annually with glyphosate containing herbicides. Studies indicate that up to 40% of applied glyphosate has the potential to reach bodies of water due to soil runoff, depending on the agricultural season and the rainfall intensity. Even low quantities of glyphosate in freshwater ecosystems have been shown to be detrimental to organisms at different trophic levels. To examine the effect of Roundup® on *Philodina acuticornis* life expectancy and reproduction, rotifers collected within 24 hours of hatching were incubated at various concentrations (0.1 mg/l, 0.05 mg/l, and 0.025 mg/l) reflecting Roundup® concentrations reported in natural ecosystems. Rotifers incubated in media without Roundup® served as the control group. Each day the adult rotifers were counted and transferred to fresh media with the appropriate Roundup® concentration. After 24 hours, the previous wells were checked again to determine the number of offspring produced. Results of this study indicate that environmental concentrations of Roundup® have a significant impact on the life expectancy of the rotifers, and rotifers exposed to the highest concentration (0.1 mg/l) survived only a few days. Survival was extended at lower concentrations but was still decreased compared to the control. Rotifers cultured in the presence of Roundup® also showed reduced reproduction levels, producing an average daily and total live offspring count lower than the controls.

BBB37 - Catfish movements in response to frequent scouring effects in Crabtree Creek, NC.

Claire Boaz, Nancy Gamewell, Jasper Doyle, Kiera Williams

*William Peace University, Raleigh, NC*

Students in Biology Department at William Peace University have been involved in a study monitoring movement of channel catfish (*Ictalurus punctatus*) and have tagged over 500 fish during the past two years. We wanted to determine the public health risks of PCB contaminated fish moving long distances from the head waters where the initial PCB release occurred. Our previous study showed fish that were tagged at the headwaters were recaptured 22 miles downstream at the confluence of the Neuse River. Our current research focused on whether catfish stay in a specific area of a stream unless otherwise forced downstream by scouring events. If fish consistently are pushed downstream, these contaminated fish pose health risks to confluence waterways, especially those subject to human activities. Our tag and release data shows that channel catfish tend to remain in localized reaches of the stream until major flood events occur. Very few tagged fish were able to maintain their position during these scour events. In addition, these tagged fish were never recaptured so were unable to recolonize.

BBB38 - Survey of Soil Microbiota with Poly 3-hydroxybutyrate (PHB) Degrading Capabilities

Ana Price, Yahaira Galan Cruzes, Ben Bielby, Carmony Hartwig

*Catawba College, Salisbury, NC*

The continual pollution of our environment with polymer plastic derivatives has made it increasingly important to find contemporary approaches for plastic and microplastic decontamination. Recent novel investigations have identified microbial partners in our environment capable of degrading a variety of plastic polymers, such as polyethylene, polyester and poly 3-hydroxybutyrate (PHB). We therefore sought to survey microbial communities from our local ecological preserve and within the Catawba College campus to determine if resident microbiota may possess enzymes capable of degrading PHB. Samples were collected, and diluted, before being inoculated onto PHB-infused Nutrient agar plates according to procedures set forth by Egusa et. al. (2018). PHB-degrading strains will be isolated and genotyped, in addition to quantifying the enzymatic rate. The enzymatic breakdown of PHB presents an opportunity to explore this concept of biologically degraded plastics as a solution to the disastrous global plastic waste problem.

BBB39 - Impact of Upstream Pasture on Habitat Use of Longnose Dace (*Rhinichthys cataractae*) in the Shenandoah Valley: An Investigation of Substrate Preference

Alison Keister

*Bridgewater College, Bridgewater, VA*

Agricultural and livestock runoff are major contributors to the presence of environmental contaminants in waterways. The combination of runoff from animal waste, herbicides, pesticides, and fertilizers from upstream areas influences water quality and the health of aquatic species. Environmental pollutants also influence behaviors of aquatic species. The purpose of this study was to understand the variation in habitat use of longnose dace (*Rhinichthys cataractae*) living in streams with different amounts of upland pasture. The longnose dace is a small minnow species native to North America and commonly inhabits fast-moving riffles and large cobble or boulder substrates. We collected longnose dace from a stream with a high proportion of upland pasture (Eidson Creek) and dace from a stream with a lower proportion of upland pasture (Narrow Passage Creek) and evaluated their substrate preference in the laboratory. We tested fish individually in a tank split evenly lengthwise with cobbles (avg. diameter = 121.2 mm) and pebbles (avg. diameter = 25.6 mm). We found that Eidson Creek fish spent the same amount of time over cobbles and

pebbles (matched pairs t-test,  $p = 0.929$ ), while Narrow Passage fish spent significantly more time over cobbles than pebbles (matched pairs t-test,  $p = 0.0187$ ). The fish living in Eidson Creek, which drains more upland pasture, may have a decreased ability to make the best habitat decisions. Other aquatic species may experience similar changes in behavior due to drainage from upland pasture and these behavioral changes have the potential to impact how species interact.

#### BBB40 - Impact of Water Quality on Axolotl Growth

Allison O'Brien

*Bridgewater College, Bridgewater, VA*

Axolotls (*Ambystoma mexicanum*) grow at different rates when housed in different water temperatures, despite being fed the same amounts of food. When axolotls are housed in cold water they exhibit lower uptake of oxygen, and therefore lower metabolism. This current research is aimed to examine if the slower metabolism of axolotls housed in cold water results in fewer molecules used as energy sources, hence more nutrients used in biomass accumulation, resulting in a higher growth rate. A group of young axolotls was randomly divided, with half housed at 20°C and half at 10°C. Following feeding, animals were transferred into clean tanks and kept there for 48 hours. After this time measurements were performed for various water quality factors, such as levels of ammonia, nitrites, nitrates, phosphates, pH, and total dissolved solids (TDS). Data compared between the two temperature conditions shows higher catabolism of proteins at higher water temperature, resulting in higher levels of ammonia, nitrites, and TDS in the waste water. The higher concentration of metabolites and overall poor quality of warm water may be detrimental to axolotl growth. The results help to generate novel data on the production of metabolites during daily life of axolotls, and to gain a better understanding of axolotl metabolism in different environmental conditions.

#### BBB41 - The effects of exposure to Great Pee Dee River contaminants in fathead minnows (*Pimephales promelas*)

Emily Llewellyn, Rikhya Ford, Angelina Jayapuram, Summer Bailey

*Francis Marion University, Florence, SC*

The Great Pee Dee River was once home to a large number of recreational fish species, however populations of these species have declined over time. The Great Pee Dee (GPD), is known to be a heavily contaminated river system, however effects of GPD contaminants on fish are unclear. Previous work has documented high contaminant loads in tissues of GPD fish; these loads have been associated with a suite of impaired health indicators including poor gonad differentiation and widespread tissue pathology. Therefore, we hypothesize that GPD contaminants represent a barrier to fish repopulation and may have an adverse effect on development of fish embryos. To test this hypothesis we exposed *Pimephales promelas* larvae (24 hours post hatch) to one of two treatments: clean water (control) or GPD water. Water was changed every 24 hours. We then monitored larval survival and growth for four weeks. We also documented morphological abnormalities such as cardiac edema, spinal curvature, and craniofacial defects to determine any overt effects of GPD water exposure. Finally, we examined the transcriptional effects of GPD exposure by performing qPCR on a suite of genes known to be altered by contaminant exposure: CYP1A (hydrocarbons), AR (androgen receptors), ER (a gene that controls estrogen receptors), VTG (vitellogenin), and MT (metallothionein). These data will lend insight into the barriers to population recruitment in the GPD, and will provide valuable information to wildlife managers responsible for fish stocking programs. BBB42 - The Butterflies of South Carolina

Caleb Bruner, Christian Watley

*Erskine College, Due West, SC*

Abstract-

Acknowledgements / co-author Dr.

Harry LeGrand, Jr.

Tom Howard

Dr. Dennis Forsythe

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LeGrand and Howard (2022) have continued to document the distribution and occurrences of butterfly species in North Carolina and South Carolina over the last 28 years. 166 distinct species of butterflies have been identified in South Carolina. It is important to note that this research is heavily dependent on the sampling effort or lack thereof, in each county and/or province. Utilization of tools such as the DNR website, the *Butterflies of North Carolina: their Distribution and Abundance. 29th Approximation*, iNaturalist, and some field sampling ourselves, we have worked to identify the locations of various butterfly species with a focus on species that have not been documented in certain areas. This information is then used to determine if a certain species requires conservation efforts to save the population, or if the butterfly has been identified correctly in the case of a rare sighting. This information will be useful to individuals concerned with both butterfly conservation and casual butterfly watching. The goal of this poster is to tabulate and analyze the South Carolina butterfly distribution data.

#### BBB43 - Underwater and In Air Hearing Capabilities of *Trachemys scripta* sp.

Abigail Peterson

*Virginia Wesleyan University, Virginia Beach, VA*

The function of the semiaquatic turtle's ear is poorly understood and not much is known about their hearing range or sensitivity. As a semiaquatic species, they encounter two unique auditory soundscapes. While sound travels more quickly in water vs. air, it also carries less intensity due to the density difference. Thus, for an animal that is equally at home in both environments, a functional hearing system would have to account for these two situations. For this study, we examined the differences between the startle behavioral responses of four red-eared slider/yellow bellied slider hybrids (*Trachemys scripta elegans* x *Trachemys scripta scripta*) to sound in air and in water. Turtles were collected using traps in Lake Taylor, Virginia Beach, VA, a lake located on the campus of Virginia Wesleyan University. Four turtles were measured, identified based on shell length, and kept in 1500L tanks in an aquatic facility. Each turtle was observed individually over a month period to differentiate behaviors and develop a working ethogram. For trial sessions, turtles were exposed to tones ranging from 250-1250 Hz using either an air or underwater speaker (Lubell Labs, Inc). On average, the turtles responded to lower frequency (750-1000 Hz) in water when compared to the in-air environment (1000-1250 Hz). Furthermore, when startled, each turtle responded quickly (within 0.03 seconds of a startle frequency being played) and there was no direct correlation between sound pressure (dB) and response duration. Finally, this study determined that the type of startle response, such as a Head Raise (HR) in air or Swimming (SW) in water greatly impacted the startle response duration. The results from this study suggest that *Trachemys scripta* sp. react with different behaviors and to slightly different frequencies in these two environments.

#### BBB44 - A Comparison of Antibiotic Resistance Levels in Feral, Semi-Feral, and Domesticated Horses

Sarah Walters<sup>1</sup>, Frauke Argyros<sup>1</sup>, George Argyros<sup>2</sup>

antibiotic resistance present in fecal isolates from feral, semi-feral, and domesticated horse herds located in Shackleford Banks, North Carolina; Linton, North Dakota; and Bristol, Virginia, respectively. Following bacterial identification, which primarily revealed *Escherichia coli*, *Klebsiella pneumoniae*, and *Proteus vulgaris* to be the cultured species, Kirby-Bauer Disk Diffusion antibiotic susceptibility testing was performed on these isolates with 18 common-use antibiotics. It is hypothesized that a correlation of increased antibiotic resistance will be observed in horse populations with higher degrees of human contact. Sampled isolates from all three populations of horses were resistant to the same four antibiotics. Shackleford isolates were susceptible to 10 of 18 antibiotics tested; North Dakota isolates were susceptible to 8 of 18 antibiotics tested; and Bristol isolates were susceptible to 7 of 18 antibiotics tested. Degree of intermediate susceptibility followed the same trend. These data support our hypothesis of increasing degree of antibiotic resistance correlated with degree of human contact and interaction. Antibiotic resistance levels are highest in the domesticated horses of Bristol, Virginia followed by the semi-feral horses of North Dakota, and the feral horses of Shackleford Banks. Further research is needed to determine the sources of antibiotics and/or resistant microorganisms facilitating these shifts towards antibiotic resistance in these populations, as well as to help mitigate further trends towards increasing antibiotic resistance.

#### BBB45 - Exploring the Relationship between Human Behaviors, Oral Microbiota, and Periodontal Disease

Janiya Downs, Francisco Camacho, Carmony Hartwig Catawba

*College, Salisbury, NC*

Periodontal disease is the result of a common, but often preventable, infection of the gum tissue within the oral cavity that surrounds the tooth. Disease occurs when plaque-forming bacteria invade pockets of the gum layer surrounding the tooth. Periodontitis, if left uncorrected, can lead to more serious problems, such as bone and tooth loss. Good oral hygiene is one of the most important habits to have for an overall healthier lifestyle. It follows that human behaviors such as vaping, smoking, consuming high amounts of sugar, etc. may influence not only the likelihood of periodontal disease over time, but also the composition of the oral microbiota. Prior research has demonstrated a higher abundance and species diversity of oral microbiota in individuals with progressive periodontal disease (Mombelli, 2000). Our research question is therefore focused on determining if there is a correlation between human behaviors, diet, age, etc. with 1) the composition (diversity) and abundance of the oral microbiome, and 2) levels of periodontal disease. We hypothesize that, similar to prior research; certain behaviors will correlate an increased abundance and diversity of the oral microbiome, and also with an increased progression of periodontal disease (i.e. improper daily dental care). In addition to sampling and plating of oral microbiota for colony characteristics and enumeration, future plans include a 16S metagenomics analysis of collected samples that will provide a more comprehensive snapshot of oral microbiota from our human participants.

#### BBB46 - Prevalence of Skeletal Pathologies in Non-Human Primates: An Osteological Study Utilizing Natural History Collections

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The horse's gastrointestinal tract exhibits a wide variety of bacteria. However, due to the widespread use of antibiotics for both prophylaxis and treatment of infection, many bacteria have evolved resistance, sometimes to multiple chemotherapeutics. We assessed prevalence and degree of

Savannah Mynatt

*Emory and Henry College, Emory, VA*

Occurrence of healed-fractures in wild populations is established in the literature, but effects on survivability for individuals is unclear. Although studies have been conducted investigating skeletal pathologies in primates and other taxonomic groups, few have addressed prevalence of such conditions relating to behavior that could lead to such pathologies, or why certain species are more likely to sustain these injuries and survive over others. The purpose of this study was to determine prevalence of healed long-bone fractures in old world primates (Hominoidea) assessed by examination of skeletal material from Natural History Collections. We hypothesize higher prevalence of fractures in arboreal over terrestrial/semiterrestrial species due to greater likelihood of injuries sustained from collisions and falls associated with locomotor behaviors (e.g., brachiation), and that survivability, to the point of well-healed fractures, is correlated with degree of gregariousness as species with collaborative feeding and highly organized social systems are more likely to survive such injuries. Skeletons of 211 individuals collected during the Harvard University Asiatic Primate Expedition (1937), and African Expedition (1926) representing old world apes housed at the Museum of Comparative Zoology, Harvard University were examined for evidence of healed fractures. Taxa represented four species: Gibbon (*Hylobates lar*, n= 155), Gorilla (*Gorilla gorilla*, n= 23), Chimpanzee (*Pan troglodytes*, n= 22), and Orangutan (*Pongo pygmaeus*, n= 11). Prevalence of healed-fractures was 31% for all species examined, combined. Gorilla's exhibited the highest prevalence of healed-fractures (65%), Orangutans (54%), Gibbons (23%), Chimpanzees (9%). Although all species exhibited fractures more than previously published studies, sample size may confound interpretation in all but *Hylobates lar*. Our hypothesis was partially supported in that survival of fractures appears to occur at higher percentages in more gregarious species; semi-terrestrial species had a higher prevalence than arboreal species, possibly due to death resulting from falls prior to healing.

#### BBB47 - The Decline of the Eastern Hemlock and the Importance of Its Micropropagation

Jonathan Cooper

*Erskine College, Due West, SC*

The Eastern Hemlock Tree (*Tsuga canadensis*) is a vital foundation species within the eastern North American ecosystem. Thus, its drastic decline since the introduction of the Hemlock Woolly Adelgid (*Adelges tsugae*) is cause for alarm and action. Solutions to the problem of the Hemlock Woolly Adelgid may take different forms, one of which may be the micropropagation of the Eastern Hemlock to boost population numbers and preserve potentially advantageous traits. Disinfestation of shoot material has been attempted using several published methods for field samples of woody plants. Trials thus far have not yielded needed quantities of clean material, due to sensitivity of shoot material to the disinfestation process as well as persistent fungal contaminants. Variations of methods yielding the best results so far continue to be tested.

#### BBB48 - Predictors of variation in the secondary metabolites of *Usnea strigosa*

Ayla Edmiston, Carley Cannon, Lisa McDonald

*Lander University, Greenwood, SC*

The basic chemical composition of common lichens is often well known because it is required for the successful identification of lichen taxa; however, spatial and temporal variation in the suite of secondary metabolites produced by lichens is not well understood. Using a common lichen, *Usnea strigosa*, we investigated chemical variation both temporally and at multiple spatial scales ranging from intra-individual variation to population-level variation. Lichens were collected from locations throughout the study area, and thin-layer chromatography was used to identify and compare the suites of potential secondary chemicals (PSCs) found in our samples. On the broadest spatial scale, we found substantial variation in PSCs both between and within populations throughout Upstate South Carolina. On a smaller spatial scale (within the city of Greenwood), we still detected variation both between and within sites. Within single sites, we were able to identify variation at the substrate level. However, we found virtually no detectable intra-individual variation in PSCs, and we failed to detect significant temporal variation in PSCs. These results suggest that there may be significant undescribed variation in the secondary chemistry of *U. strigosa* across many spatial scales. This contrasts with historical evidence that the chemistry of *U. strigosa* is simple, with a small number of known chemotypes found in predictable locations throughout its large geographic range.

#### BBB49 - Going nuts: Eastern gray squirrel behavior is unaffected by predator activity

Chandler Russian, Lisa McDonald, Emily Prince

*Lander University, Greenwood, SC*

The Eastern gray squirrel, *Sciurus carolinensis*, is common across the Eastern United States. Their success may be due, in part, to their ability to avoid predation. For this study, we asked how squirrels' behavior might differ in areas with different predation risk. Our hypotheses were that in areas with high predator activity, squirrels would have lower overall activity, be observed in groups more frequently, and be more nocturnal. To test these hypotheses, we observed the behavior of gray squirrels and their predators using camera traps at 26 camera stations across 6 sites in Upstate South Carolina. Each image was labeled by time of day, species present, and number of individuals. We calculated squirrel activity as the number of squirrel observations per camera day. We used the observed frequency of potential squirrel predators (i.e., foxes, feral cats, bobcats, and coyotes) to categorize stations as having high, medium, or low predation-risk levels. We discovered that the squirrels were not significantly more nocturnal or more likely to form groups in the presence of higher predation risk. We also determined that mean squirrel activity was greater at

locations with both high and medium predator-risk levels than in locations with low predator risk, which was the inverse of what we expected. This may be because predators are attracted to areas with high squirrel activity or because some areas provide better habitat for both squirrels and predators. Overall, our results suggest squirrels do not alter behavior as expected in response to predation risk.

#### BBB50 - Behavioral responses of white-tailed deer (*Odocoileus virginianus*) to predation risk

Elisabeth Howansky, Zachrey Swartzentruber, Lisa McDonald, Emily Prince

*Lander University, Greenwood, SC*

White-tailed deer (*Odocoileus virginianus*) engage in specific behaviors to minimize predation risk. Because humans also pose a threat, deer may respond to human disturbance in the same ways they respond to other predators. For this study, we sought to determine whether the behaviors of deer differ between areas with differing levels of threat from humans and other predators. Because deer are more visible in open habitats, we hypothesized that with increasing habitat openness deer would increase vigilance and group formation to better detect and avoid predators. We hypothesized that deer would also increase vigilance and group formation with increasing human disturbance. Lastly, we hypothesized that deer would be more nocturnal at sites with higher human disturbance to avoid encountering diurnal humans, but that deer would be more diurnal in open habitats to avoid nocturnal predators. To test these hypotheses, camera traps were set up in locations across Upstate South Carolina with varying habitat openness. We calculated a human disturbance score for each location using the frequency of images containing human activity. The observations were also used to calculate the group formation, vigilance, and nocturnality of deer. We found that deer were more vigilant and more likely to form groups in open habitats and sites with high human disturbance. We also found that nocturnality was higher in sites with high human disturbance and, contrary to our prediction, in more open habitats. This may be because the benefit of decreased visibility outweighs the risk of encounters with nocturnal predators. Overall, our results indicate that white-tailed deer do modify their behavior in response to perceived predation risk.

## Panels

### PANEL: ESRI Panel Discussion: Career Pathways for Applied Environmental Scientists

Historically, there were traditional career paths for students graduating with biology degrees. However, as global awareness around the interconnectedness of our environmental, social and economic systems increases, so too does demand for applied environmental knowledge across a wide array of industries. This panel will expose students to representatives from the public, non-profit and technology sectors whose career paths deviated from the “classic.” Students will hear about their experiences in college, in their careers and advice about navigating professional development. Audience members will also have an opportunity to ask the panelists questions in a light, fun and engaging session. The panel will feature Liv Haney, a Product Manager with Ecobot; Wesley Knapp, Chief Botanist for NatureServe, and Lesley Starke, Plant Conservation Program Manager for the North Carolina Department of Agriculture and Consumer Services.

### PANEL: Hitting the Ground Running with Your Graduate School Applications --Hosted by ASB's Human Diversity Committee

Jennifer Mandel, University of Memphis, Department of Biological Sciences, Memphis, TN, Chinyere Knight, Tuskegee University, Tuskegee, AL and Veronica Segarra, Goucher College, Baltimore, MD

This 2nd annual workshop hosted by ASB's Human Diversity Committee has been designed to better prepare our undergraduate student members to apply to graduate school. Through dynamic discussion with more experienced ASB member panelists, this workshop will raise participants' awareness of the key elements of successful graduate school applications—especially those that are often not explicitly described to or required from undergraduate applicants. After a short introductory presentation, an interactive moderated discussion will ensue with the help of experienced ASB members who will serve as panelists and answer prepared and real-time participants' follow up questions related to the topic. There will also be short exercises throughout the workshop that will help attendees experience successful strategies for career development. The workshop will end with a short review of the take-home messages to remember.

### PANEL: NSF Research and Training Opportunities for Biologists

Join NSF Program Officers to learn about new and ongoing NSF funding opportunities of relevance to biologists at all career stages. This session will cover how NSF funds research, the grant proposal process, and funding opportunities through Biology core programs and current special solicitations. Overviews of Research Experiences for Undergraduates (REU) opportunities, the Graduate Research Fellowship Program (GRFP), Postdoctoral Research Fellowships in Biology (PRFB), the Faculty Early Career Development Program (CAREER), the Building Research Capacity of New Faculty in Biology Program (BRC-BIO) for early-career faculty at non-R1 institutions, and the Mid-Career Advancement (MCA) program will be provided. A question-and-answer session will follow the overview of the funding and training opportunities.

### PANEL: Promoting Student Success Through Implementation of Specifications Grading in STEM Courses

Jennifer Hurst-Kennedy<sup>1</sup>, Mary Beth Anzovino<sup>1</sup>, Alessandra Barrera<sup>1</sup>, Amy H Erickson<sup>1</sup>, Shoshana Katzman<sup>2</sup>, Michael Morton<sup>1</sup>, Karen Perrel-

Gerson1, Jennell Talley1, Omar Villanueva1 and Cynthia Woodbridge1, (1)Georgia Gwinnett College, Lawrenceville, GA (2)Agnes Scott College, Decatur, GA

Have you ever wondered if there was a grading method where students would be willing to try harder to show they have mastered content and spend less time begging for extra credit or extra points? Specifications grading might be your new best friend. Come learn how to use specifications grading in any course to enhance student learning and engagement. Panelists will include Chemistry, Math, and Biology faculty discussing how they implement specifications grading in their courses. We will also provide attendees with a "Specs Starter's Guide" with resources on how to use specifications grading in their classes.



## Workshops

### WORKSHOP: Build your own open-source hardware GPS data logger for wildlife movement studies

Patrick Cain, Georgia Gwinnett College, Lawrenceville, GA; Toledo Zoo and Aquarium, Toledo, OH and Matthew Cross, Toledo Zoo and Aquarium, Toledo, OH

Global Positioning System (GPS) technology has been a boon to animal spatial ecology studies, allowing researchers to obtain vast quantities of data, even for cryptic organisms. Oftentimes, the main limitation to wide-spread application of this technology is the cost, which can dictate the number of individuals outfitted with GPS technology, thereby limiting sample sizes. Here, we discuss the use of Arduino in developing a low-cost, customizable, open-source hardware GPS logger for use in animal movement studies. We also present results from field tests with eastern box turtles (*Terrapene carolina*) in northwestern Ohio. These GPS loggers have the potential to augment existing projects and facilitate studies that would be otherwise cost-prohibitive. This workshop will introduce attendees to the basic concepts of open-source, do-it-yourself technology and some of its applications. Attendees will learn how to assemble a GPS data logger through a guided exercise, and will be able to take home their creation. Sessions will include a brief lecture, basics of soldering tutorial (via a customized LED exercise), guided GPS unit construction, field testing, downloading and visualization of data, casing and attachment options, customization options, and future projects.

### WORKSHOP: Honey, I Blew-Up the Molecules: 3D Printing as a Multi-faceted and Multi-disciplinary Pedagogical Practice

Carmony Hartwig, Amanda Bosch and Zachary Trivett, Catawba College, Salisbury, NC

Interested in engaging your students with hands-on models that bring abstract theories to physical 'life'? Join us on a journey into the world of 3D printing molecular models to help students understand more complex concepts such as drug-molecule interactions, mutational effects on protein structure, and macromolecular composition. During this session, we focus on ways to expand 3D print models into larger course projects that incorporate multiple digital pedagogies such as: Poster presentation with Augmented Reality (AR) apps, exploration of enzyme active sites with Virtual Reality (VR) programs...and beyond! We will brainstorm ways to use this practice in diverse curricula, and in a variety of applications, from drone technologies to field work.



### WORKSHOP: Spatial Analytics Workshop with ArcGIS Pro and ArcGIS Online

There are a wide variety of analytics that biological and environmental professionals may need to apply in their careers and many ways to achieve these. In this 1 hour workshop, Esri will teach participants how to conduct presence-only predictive modeling, hot spot and cluster analysis, interpolation with kriging and more with ArcGIS Pro. We'll discuss how to enhance analyses with the R-bridge, python and Jupyter notebooks

before we briefly show how these analyses can be published to ArcGIS Online. Once in ArcGIS Online, users can create a wide array of information products to help interpret results for a public audience.

#### WORKSHOP: SISRIS: Supporting Inclusive and Sustainable (collections-based) Research Infrastructure for Systematics

Scientific discoveries derived from biological collections depend on the people who create the infrastructure of preserved specimens. However, currently accepted metrics of professional success for taxonomists ignore the essential functions of collection, identification, and annotation of research specimens that support published research. This results in an incomplete picture of the contributions of those working in biological collections. Recently, web-based informatics tools have been developed to improve attribution of specimens, so that collectors and determiners can track their contributions to subsequent research discoveries made using their work and receive greater professional recognition. In this workshop, participants will learn how to create an inclusive and sustainable research ecosystem for collections'-based research by increasing attribution of the people. This half-day workshop will comprise four hours of instruction about the structure and use of the web-based informatics tool Bionomia and the other software services that interact with this platform, including ORCID, GBIF, Wikidata, and Zenodo. The workshop will emphasize (1) how participants can use these tools as part of their collections-based research activities and (2) how increased attribution can be used as a means to effect community-level change in how taxonomic expertise is acknowledged. Participants will practice attributing specimens to historical collectors from underrepresented groups, which will directly increase the visibility of hidden figures in natural history collections.

Shawn Krosnick, Tennessee Tech University, Cookeville, TN and Andrea Weeks, George Mason University, Department of Biology and Ted R. Bradley Herbarium, Fairfax, VA

## Field Trips

### FIELD TRIP: Monadnock Forests and Fire Ecology at Pilot Mountain State Park

Steph Jeffries, NC State University, Raleigh, NC and Dane Kuppinger, High Point University, High Point, NC

Join us at Pilot Mountain to learn about Piedmont monadnocks and fire history. Pilot Mountain, part of the Sauratown Mountains, supports xeric and sub-xeric forest communities which are distinct from the surrounding Piedmont forests. Unlike much of the Piedmont, these communities have been significantly shaped by both anthropogenic and naturally occurring fires. We plan to hike the Pilot Knob Trail and the Grindstone Trail to see effects from the 2021 fire and learn about the role of fire in these forests and some of the long-term community ecology research that is being done at Pilot Mountain. Participants can choose a shorter hike (0.8 mi) that ends before lunch, or can add a longer trail (3.5 mi, one-way downhill) to extend the field trip and wrap up by mid-afternoon.

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Stiltner

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**PRES. NO.**  
Thomas

**LAST NAME**  
Michelle

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LAST NAME	FIRST NAME	PRES. NO.	LAST NAME	FIRST NAME	PRES. NO.
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Willis	Caroline				
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Windham	Michael				
Winterton	Elyssa				
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Wolff	Robert				
Wolfram	Shelby				
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Woods	Natasha				
Woods	Ashleigh				
Wright	Dahja				
Wright	Alexandra				
Wright	Douglas S.				
Wright	Masey				

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Wynne	Aurora	128			

# THE 84TH ANNUAL MEETING OF THE ASSOCIATION OF SOUTHEASTERN BIOLOGISTS

WINSTON-SALEM, NC

23 MARCH - 26 MARCH 2023



T-shirt designed by Elijah Wells Campbell  
University, NC.

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(ISSN 1533-8436)

ASB Affiliate Organizations



SE CHAPTER



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TRIBETA DISTRICTS I & II

# Association of Southeastern Biologists 2023

## Meeting-At-A-Glance

Thursday, March 23 - 26, 2023

Benton Convention Center

### Thursday, March 23

Time	Event	Location
9:00 am – 4:00 pm	Exhibitor Set-up	Salem Ballroom 2/3
11:00 am – 9:00 pm	Registration Open	Salem Ballroom Foyer
11:00 am – 2:00 pm	Exhibitor's Coffee Break (exhibitors only)	Salem Ballroom 2/3
1:00 pm – 2:00 pm	ASB Executive Committee Meeting	Salem Ballroom 2/3
2:00 pm – 4:00 pm	SABS Executive Council Meeting	Hearn A - Marriott
6:30 pm – 8:00 pm	<b>WELCOME AND PLENARY</b> Welcome: ASB 2023 R.H. Martin Plenary Speaker   Pulitzer-Prize Winning Journalist and Author Ed Yong	Winston Ballroom 2
8:00 pm - 9:30 pm	Book signing - Foyer Welcome Reception - Ticketed Event	Salem Ballroom 2/3

### Friday, March 24

Time	Event	Location
7:00 am – 5:00 pm	Registration Open	Salem Ballroom Foyer
7:00 am – 5:00 pm	PowerPoint Preview Check – Lightning Talks	Salem Ballroom 2/3
7:00 am – 8:15 am	ASB Past President's Breakfast Meeting	Hearn C - Marriott
8:00 am – 10:00 am	<b>ASB POSTER PRESENTATION MORNING SESSION</b> Presenters are asked to stand with their poster during one session	Salem Ballroom 2/3
8:00 am – 5:00 pm	Exhibits Open	Salem Ballroom 2/3
9:30 am – 12:00 pm	SYMPOSIUM: Conservation, Natural History, and Systematics of Crayfishes - a Southeastern perspective.	Winston Ballroom 2
10:00 am – 11:45 am	<b>TALK SESSION 1</b> MICROBIOLOGY A PLANT COLLECTIONS AND HERBARIA ECOLOGY: ECOSYSTEMS & LANDSCAPES A ANIMAL: BEHAVIOR ANIMALS: ENTOMOLOGY ECOLOGY: POPULATIONS	Winston Ballroom 1A Winston Ballroom 1C Winston Ballroom 3C Salem Ballroom 1A Salem Ballroom 1B Salem Ballroom 1C
10:00 am – 10:55 am	ESRI Panel Discussion: Career Pathways for Applied Environmental Scientists	Winston Ballroom 1B
10:00 am – 11:00 am	Honey I Blew Up the Molecules: 3D Printing as a Multi-faceted and Multidisciplinary Pedagogical Practice WORKSHOP	Winston Ballroom 3B
11:00 am – 12:00 pm	Spatial Analytics Workshop with ArcGIS Pro and ArcGIS Online	Winston Ballroom 1B
11:45 am - 1:00 pm	TRIBETA Joint Business Meeting and Mixer (all delegates MUST attend)	Winston Ballroom 1C
12:00 pm - 1:15 pm	Lunch ASB Diversity Committee ASB Education Committee SABS Lunch	Hearn C - Marriott Hearn D - Marriott Winston Ballroom 3B

1:00 pm – 3:15 pm	SYMPOSIUM: Conservation, Natural History, and Systematics of Crayfishes - a Southeastern perspective.	Winston Ballroom 2
1:15 pm – 3:00 pm	<b>TALK SESSION 2</b> GENETICS AND CELL & MOLECULAR BIOLOGY A ECOLOGY: FRESHWATER ECOLOGY: CONSERVATION A PLANT FLORISTICS ANIMALS: HERPETOLOGY ECOLOGY: COMMUNITIES A	Winston Ballroom 1A Winston Ballroom 1C Winston Ballroom 3C Salem Ballroom 1A Salem Ballroom 1B Salem Ballroom 1C
1:30 pm – 3:00 pm	NSF research and training opportunities for biologists	Winston Ballroom 1B
1:30 pm – 3:00 pm	Hitting the Ground Running with Your Graduate School Applications --Hosted by ASB's Human Diversity Committee	Winston Ballroom 3A
1:30 pm – 3:00 pm	SISRIS: Supporting Inclusive and Sustainable (collections-based) Research WORKSHOP	Winston Ballroom 3B
3:00 pm – 5:00 pm	<b>ASB POSTER PRESENTATION AFTERNOON SESSION</b> Presenters are asked to stand with their poster during one session TriBeta Posters	Salem Ballroom 2/3
4:45 pm - 5:45 pm	SABS/BSA Student Reception	Winston Ballroom Foyer
6:00 pm - 10:00 pm	<b>FRIDAY NIGHT SOCIAL</b> - Ticketed Event Live Music, Food, Drinks, Games	Roar

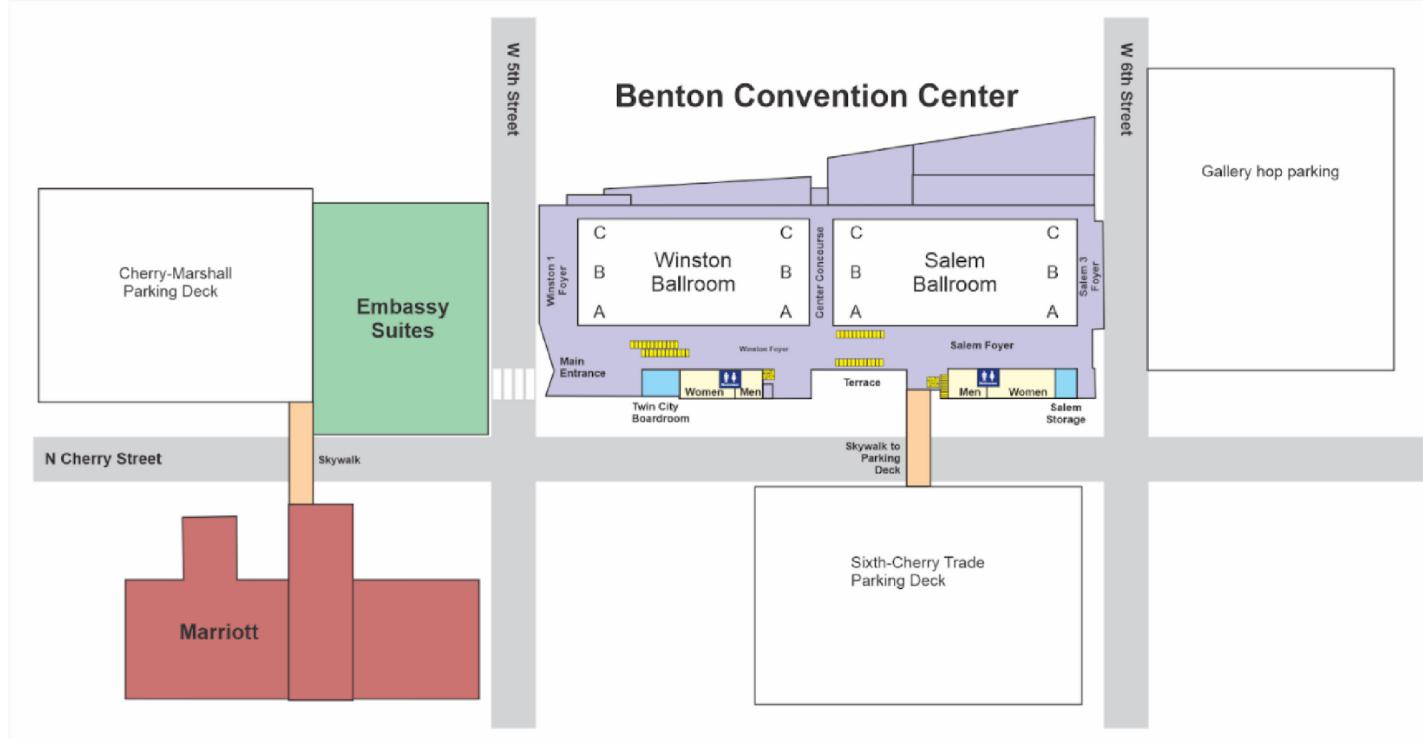
**Saturday, March 25**

Time	Event	Location
7:00 am - 12:00 pm	Registration Open	Salem Ballroom Foyer
7:00 am – 5:00 pm	PowerPoint Preview Check – Lightning Talks	Salem Ballroom 2/3
8:00 am – 10:00 am	<b>ASB POSTER PRESENTATION MORNING SESSION</b> Presenters are asked to stand with their poster during one session TriBeta Posters	Salem Ballroom 2/3
7:30 am – 6:00 pm	TRIBETA Officers & Judges Room	Twin City Boardroom
10:00 am – 12:00 pm	TRIBETA SESSION 1	Winston Ballroom 1C
10:00 am – 11:45 am	<b>TALK SESSION 3</b> MICROBIOLOGY B ECOLOGY: ECOSYSTEMS & LANDSCAPES B PLANT SYSTEMATICS ANIMALS: ENTOMOLOGY ECOLOGY: PHYSIOLOGICAL	Winston Ballroom 1A Winston Ballroom 3C Salem Ballroom 1A Salem Ballroom 1B Salem Ballroom 1C

10:00 am – 12:15 pm	Conservation Genetics - Symposium	Winston Ballroom 1B
10:00 am – 12:00 pm	SYMPOSIUM: Conservation, Natural History, and Systematics of Crayfishes - a Southeastern perspective.	Winston Ballroom 2
10:00 am – 12:00 pm	SISRIS: Supporting Inclusive and Sustainable (collections-based) Research Infrastructure for Systematics - SYMPOSIUM	Winston Ballroom 3A
10:00 am – 12:00 pm	Promoting Student Success Through Implementation of Specifications Grading in STEM Courses	Winston Ballroom 3B
10:00 am - 11:45 pm	PULSE	Hearn C - Marriott
12:00 pm - 1:15 pm	SHC SE Chapter Lunch and Business Meeting ESA Lunch and Business Meeting	Hearn C - Marriott Hearn D - Marriott
1:15 pm – 3:00 pm	TRIBETA SESSION 2	Winston Ballroom 1C
1:15 pm – 2:45 pm	<b>TALK SESSION 4</b> GENETICS AND CELL & MOLECULAR BIOLOGY B ECOLOGY: CONSERVATION B SCHOLARSHIP OF TEACHING AND LEARNING ANIMALS: HERPETOLOGY ECOLOGY: COMMUNITIES B	Winston Ballroom 1A Winston Ballroom 3C Salem Ballroom 1A Salem Ballroom 1B Salem Ballroom 1C
1:15 pm – 3:00 pm	Southeastern Chapter of ESA-sponsored Symposium: Revisiting some of the plants that William Bartram wrote about in his 'Travels' to North Carolina, 1775 and 1776 - SYMPOSIUM	Winston Ballroom 1B
1:15 pm – 3:00 pm	SYMPOSIUM: Conservation, Natural History, and Systematics of Crayfishes - a Southeastern perspective.	Winston Ballroom 2
1:15 pm – 3:00 pm	SISRIS: Supporting Inclusive and Sustainable (collections-based) Research Infrastructure for Systematics - SYMPOSIUM	Winston Ballroom 3A
1:15 pm – 3:00 pm	Joro Spider Summit - SYMPOSIUM	Winston Ballroom 3B
3:00 pm – 5:00 pm	<b>ASB POSTER PRESENTATION AFTERNOON SESSION</b> Presenters are asked to stand with their poster during one session	Salem Ballroom 2/3
5:00 pm – 6:00 pm	TRIBETA Joint Session and Awards	Winston Ballroom 1C
5:00 pm – 6:00 pm	ASB Members Meeting (All ASB members invited)	Winston Ballroom 3A
7:00 pm – 9:00 pm	<b>ASB AWARDS BANQUET</b> - Ticketed Event	Winston Ballroom 2

**Sunday, March 26**

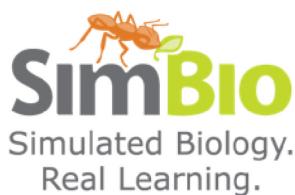
Time	Event	Location
7:00 am - 12:00 pm	<b>FIELD TRIP TO PILOT MOUNTAIN</b>	Meet up location TBA
7:00 am – 12:00 pm	EC Breakfast and meeting (for Current and Incoming EC members only)	Hern A - Marriott



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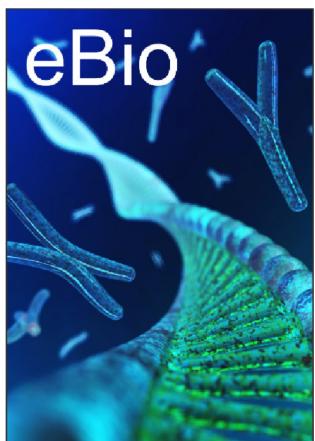
## 2023 Exhibitors



College of Pharmacy & Health Sciences



## ***eBIO & The Southeastern Naturalist: Official Journals of the ASB***



*eBio* (ISSN 2165-6657), is a peer-reviewed official journal of ASB that publishes original articles and notes focused on the biology of organisms as it relates to the structure, function, and development of their internal systems, and as it pertains to their use in medical and other applications not directly related to the ecology and conservation of species or their habitats. Subject areas include, but are not limited to, biochemistry, biotechnology, cell biology, developmental biology, genetics and genomics, immunology, microbiology, molecular biology, neurobiology, physiology, parasitology, and toxicology.

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*The Southeastern Naturalist* (Print ISSN #1528-7092 and Online ISSN #1938-5412) is a peer-reviewed official journal of ASB that publishes original articles focused on natural history research related to all aspects of the biology and ecology of terrestrial, freshwater, and marine organisms and the environments of the southeastern portion of North America.