26th Annual

BIOSYMPOSIUM



2017

James Madison University

Biology Department

*Thursday, April 20 - Friday, April 21, 2017*

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**C** Research that is part of CGEMS (Center for Genome and Metagenome Studies)

Digital posters (in room 2009)

\* Research that was supported by summer funding gifts

**g** Graduate student research

**Schedule: Thursday April 20th**

**SESSION 1 11:00am-1:00pm POSTERS 2nd Fl. Foyer**

P1 Katherine M. Bland, Christopher J. Handwerk, Z. Logan Holley, Zachary O. Casey, Dr. George S. Vidal

**Exploring cerebral cortical synaptic pruning in models of Autism Spectrum Disorder**

P2 **g** Sarah McGrath, Francis Gebhard, Walker Spradlin, Cole Roberts, Dr. David McLeod

**A review of larval amphibian field enclosure techniques and introduction of a novel field device**

P3 Jacob Gumpf, Behan Alavi, Dr. Oliver Hyman, Dr. David McLeod

**Detection of *Batrachochytrium dendrobatidis* but not *B. salamandrivorans* in Bornean frog populations**

P4 Taylor Hutchinson, Rana Ihsan, Dr. Tim Bloss

**Characterization of misfolded protein stress inducers in the different cell types of *C. elegans***

P5 Lauren Pope, Jillian Breault, Dr. Jonathan Monroe

**Quaternary Structure of β-Amylase 2 in *Arabidopsis thaliana***

P6 Graham Price, Anne Masters, and Dr. Marquis T. Walker

**Identifying the genetic locus and characterizing the pathophysiology of *Drosophila* retinal degeneration mutant, rdgI**

P7 Madeline Henwood, Shaw Camphire, Dr. Terrie Rife

**Characterizing tau in the nucleus**

P8 Elizabeth Rogers, Stephanie Sharpes, Dr. Kyle Seifert

**The Effects of Spacer Variation and Hydrocarbon Tail Length on the Antibacterial Activity of Novel Tris- and Bis-Cationic Amphiphiles**

P9 Justin Rissmiller, Shannon Palmer, Dr. Bisi Velayudhan

**Effect of accelerated feeding and estrogen treatment on Brunner’s glands and submucosal thickness in the duodenum of diary calves**

P10 *poster cancelled*

P11 Marissa Reilly, Dr. Bisi Velayudhan

**Quantification of the enteroendocrine cells in the small intestine of dairy cattle in response to accelerated feeding**

P12 Patricia Brown, Dr. Kevin Hatala, Heather Dingwall, Dr. Brian Richmond, Dr. Roshna Wunderlich

**Foot Structure and Function in Habitually Unshod Children**

P13 Danielle Orlandi, Mark McGowan, Dr. Anthony Tongen, Dr. Roshna Wunderlich

**The Effects of Cage Size in Captivity on Physical Activity Levels in *Propithecus coquereli***

P14 Matthew Morrissey, Guy Stewart, Dr. Heather Griscom

**The effects of varying environmental conditions on American chestnut hybrids in an Appalachian Cove Forest in West Virginia**

P15 Molly Campillo, Dr. Joseph Harsh

**Expertise in "seeing" in visual data: An eye tracking and think aloud using graphical representations**

P16 Bryan Scalf, Jenny Russel, Dr. Chris Lantz

**Assessment of Faithful Interleukin-3 Production by Transgenic Fluorescent Reporter Mice**

P17 Andrew Ayad, Rohan Bamania, Elizabeth Dunlap, Mason Earp, Robert Hacker, and Dr. Michael Renfroe

**The effect of stressors on beta-carotene production in *Dunaliella bardawil***

P18 Wendy Chen, Marlene Flores, Matthew Lowry, Hannah Smith, Morgan Smith, and Dr. Michael Renfroe

**Inducing the production of astaxanthin in *Haematococcus pluvalis***

P19 Andrew Ayad, Rohan Bamania, Wendy Chen, Elizabeth Dunlap, Mason Earp, Marlene Flores, Robert Hacker, Matthew Lowry, Hannah Smith, Morgan Smith, and Dr. Michael Renfroe

**Effect of synthetic seed storage conditions on emergence of *Saintpaulia rupicola***

P20 Anjali Batra, Phoebe Cook, Dr. Idelle Cooper

**Color Variation within Hawaiian damselfly, *Megalagrion calliphya***

P21 Taylor Derby, Monique Waldman, Dr. Marta Bechtel

**Characterizing Decellularization Methods in the Rabbit Cornea Scaffold**

***Moved from Friday's poster session:***

P51 \* Cecilia Rogers, Dr. Heather Griscom

**Investigating the ecology and habitat modeling for *Solanum conocarpum* on St. John, USVI** *(moved to Thursday's poster session)*

P52 Anna Nordseth, Dr. Heather Griscom

**Coffee carbon stocks, pests and diseases under varied shade management: a review** *(moved to Thursday's poster session)*

**Schedule: Thursday April 20th**

**SESSION 2 1:00pm-4:45pm ORAL PRESENTATIONS Biosci 2007**

12:55 Introduction by Dr. Grace Wyngard

1:00 T1. Sara Schoen, Dr. Dave McLeod

**Defining Characteristics and Class Sizes Within the *Limnonectes kuhlii* Complex using Statistical Analysis of Morphometric Traits**

1:15 **g** T2. Peter Riley and Dr. Mike Renfroe

**Development of Media for in vitro Symbiosis between *Burkholderia phytofirmans* and *Dionaea muscipula***

1:30 \* T3. Milos Lesevic, Grace Bauer, Dr. Terrie Rife

**Nuclear alpha-synuclein and its impact on transcription of Nitric Oxide Synthase I**

1:45 T4. Heather Maher, Dr. David S. McLeod

**Delimiting frog species using head-shape analyses.**

2:00 T5. Zachary Casey, Dr. George Vidal

**Capturing ex vivo electrophysiological data from Cerebral Cortex**

2:15 **C** T6. Grant Rybnicky, Dr. Steven Cresawn

**Repeaterator: a tool for visualizing DNA repeat motifs in Actinobacteriophage genomes**

**2:30** : : : : : : : : **BREAK** : : : : : : : :

2:45 **g** T7. Kelly Livernoche, Dr. Heather Griscom

**The effect of cattle management on soil carbon; implications for climate change**

3:00**g** T8. Romie D. Powell, Dr. Justin Brown

**The Role of Ambient Temperature in the Recovery from Major Surgical Stress in Rats**

3:15 **g** T9. Jessie Mandirola, Dr. Patrice Ludwig

**Oyster Restoration Utilizing Artificial Substrates: Implications for Mitigating Oyster Decline in the Chesapeake Bay**

3:30 **C**\* T10. Courtney Stout, Nick Dunham, Dr. Chris Berndsen, Dr. Raymond Enke

**Epigenetic regulation of photoreceptor-specific transcription factor binding in the genomes of retinal neurons**

3:45 T11. Grant Rybnicky

**Qualification of qPCR positive control at Merck Elkton**

4:00 **C**\* T12. Claire Gormley, Kristen Hoffman, Dr. Tim Bloss

**Characterization of Cell-type Specific Responses to Misfolded Protein Stress in *C. elegans***

4:15 T13. Roxana Behrooz, Sean M. Gay, Dr. Mark Gabriele

**Complementary multimodal compartments in the developing midbrain**

4:30 **C g** T14. Curtis Kapsak, Kevin Libuit, Erika Gehr, Dr. James Herrick

**Determining the prevalence of antibiotic resistance plasmids found in native stream bacteria in the Shenandoah Valley**

**Schedule: Friday April 21st**

**SESSION 3 9:00am-noon ORAL PRESENTATIONS BIOSCI 2007**

9:00 Introduction by Dr. Cynthia Bauerle, Dean of College of Science and Mathematics

9:05 T15. Gregory Mansour, Melissa Encinias, Matthew Riordan, Cody Cubbage, Dr. Dean Cocking

**Persistence and Sustainability: Trace Mercury in “Uncontaminated” Rockingham County VA Terrestrial Ecosystems Represent a Widespread Pattern of Occurrence**

9:20 \* T16. Melissa Encinias, Gregory Mansour, Matthew Riordan, Cody Cubbage and Dr. Dean Cocking

**Detectable Traces of Mercury Exist Within Invertebrate Food Webs of Low Contamination Shenandoah Valley Terrestrial Ecosystems**

9:35 **g** T17. Elizabeth Rogers, Dr. Kyle Seifert

**The Antibacterial and Biofilm Disruption Activity of Novel Amphiphiles**

9:50 T18. Naomi Gilbert, Dr. Steven Wilhelm, Dr. Morgan Steffen

**Survey of microbial urea degrader diversity in Lake Shenandoah and the Shenandoah River**

10:05 **g** T19. Pat Harmon, Dr. Christine May

**Revealing the Current Relationship between Stream Acidification and Fish Species Richness: What is the Status after Two Decades of Recovery?**

**10:20** : : : : : : : : **BREAK** : : : : : : : :

10:30 \* T20. Giavanna Verdi, Taylor Berrena, Miriam Kabore, Rachel Pallister, Mara Gaykema, Dr. Corey Cleland

**Rat hind limb nociceptive withdrawal response to heat and mechanical stimuli depends on initial position of the paw but not stimulus location**

10:45 \* T21. Matt Kohler, Dr. Amanda Storm, Dr. Jon Monroe

**Effects of the microenvironment surrounding Cys433 in *Arabidopsis* β-amylase-1 and -3 on the sensitivity to glutathionylation by nitrosoglutathione**

11:00 \* T22. Hayley Fried, Naomi Gilbert, Taylor Wright, Michael Gay, Dr. Morgan Steffen

**A Survey Of The Bacterial Diversity And Function In Lake Shenadoah (Virginia, U.S.A.)**

11:15 T23. Mary Pegalow, Seerat Mavi, Diana Grigoryan, Corey Cleland

**Integration of limb or tail nociceptive withdrawal responses and postural body movements in unrestrained rats**

11:30 T24. Jillian Breault, Lauren Pope, Dr. Amanda Storm, Dr. Jonathan Monroe

**Characterizing the salt dependence of β-Amylase2 in Arabidopsis**

11:45 **g** T25. Phoebe Cook, Rebecca Rasmussen, Edward Hsieh, Dr. Jonathan Brown, Dr. Idelle Cooper

**Sexual selection on color variation in *Megalagrion calliphya***

**Keynote Presentation:**

Dr. David Rizzo

University of California - Davis

*From the beginning: a brief history of an*

*emerging forest disease*

***Bioscience 1007, 12:20-1:10***

1:10-2:00 LUNCH RECEPTION FOR PRESENTERS - Second floor foyer

**Schedule: Friday April 21st**

**SESSION 4 FRI 2:00pm-4:00pm POSTERS 2nd Floor Foyer**

P22 \* Toma Matveeva, Jared Martin, Dr. Kimberly Slekar

**Investigating the role of ß-NAC on stress response in *Saccharomyces cerevisiae***

P23 Amanda Crandall, Kristianna Bowles, Rhiannon English, Cole Roberts, Nathan Robinson, Dariia Yerahova, Dr. Bruce Wiggins

**A Comparison of Riparian Characteristics and Resulting Water Quality in Restored Agricultural Systems**

P24 Kristianna Bowles, Amanda Crandall, Rhiannon English, Megan Moore, Cole Roberts, Nathan Robinson, Dariia Yeharova, Dr. Bruce Wiggins

**Using LiDAR to generate digital elevation models for watershed delineation and canopy height measurements**

P25 Kristianna Bowles, Amanda Crandall, Cole Roberts, Rhiannon English, Dariia Yeharova, Nathan Robinson, Dr. Bruce Wiggins

**Effects of Riparian Restoration and Land Use on Water Quality in the Shenandoah Valley, Virginia**

P26 Dominic Sales, Michael Pamonag, Dr. Kristopher Kubow

**Influence of fiber orientation and fiber movement on cell polarization and migration**

P27 Reef Buckhalter, Jessica Cornell, Peyton Coady, Dr. Marta Bechtel, Dr. Kristopher Kubow

**Development of a Cell Culture System to Study the Effects of Physical Confinement and Spatial Arrangement on the Maintenance of Chondrocyte Phenotype**

P28 Kristin Sammons, Lexi Deak, Dr. Corey Cleland

**Selective Stimulation of A-delta Nociceptors in Rat Hind Limb and the Resulting Nociceptive Withdrawal Response**

P29 Seerat Mavi, Mary Pegelow, Diana Grigoryan, Dr. Corey Cleland

**The effect of noxious tail simulation on the nociceptive withdrawal response of unrestrained rats**

P30 Don Afful, Aubrey Siebels, Ariel Childs, Alex Zeher, Corey Cleland

**Sensory Mechanisms Underlying the Escape Response to Looming Stimuli in Crickets**

P31 Ashley Ballengee, Paulina Bauer (co-first authors), Dr. Corey Cleland

**Skin temperatures in the nociceptive withdrawal response of the rat tail**

P32 Courtney Neumeyer, Rebecca Livermore, Gregory Steffensen, Dr. David McLeod

**A Review of Overwintering Strategies in Three Species of North American Amphibians**

P33 Jenna Salter, Dr. Bryan Cage, Dr. David McLeod

**Revolutionary Methods of Telemetry for the Ecological Study of Eastern Box Turtles**

P34 Shelby Snowden, Dr. Kerry Cresawn

**An Exploratory Study to Understand Elementary School Students’ Conceptions of Food Chains**

P35 Chelsea Romanchuk, Ashley Warrington, Brooke Thompson, Dr. Conley K. McMullen

**Updating floristic surveys of the Edith J. Carrier Arboretum and the Smith Creek restoration area**

P36 Sam Hetrick, Lilly Nelson, Romie Powell, Alexander Schmidt, Dr. Justin Brown

**The Role of Brain Stem 5HT1A and GABA-A Receptors in the Thermoregulatory Response to Hypoxic Stress**

P37 Madison Frongello, Gabriela Constancia, and Dr. Marquis T. Walker

**Characterizing the role of the TRPML cation channel in *Drosophila* innate immunity**

P38 **g** Matthew Harris, Amanda Crandall, Mark Petre, Chloe Wines, Dr. David McLeod

**Ultrasonic Vocalization in Southeast Asian Frogs**

P39 \* Esraa Aldkheil, Dr. Nathan Wright

**Characterization of the Titin ZIg9/10-Obscurin Ig58/59 Complex**

P40 \* Rachel Barborek, Kendyl Combs, Adam Fischel, Althea Neighbors, Will O'Connor, Osna Samady, Ryan Samuel, Rebekah Tenney, Dr. Susan Halsell

**Identifying the molecular components of cold nociception in *Drosophila melanogaster***

P41 **C** Andie Gargiulo, Ashton Holub, Melika Rahmani-Mofrad, Dr. Ray Enke

**RNA sequencing analysis of the human retinal transcriptome**

P42 Emily Miller, Ellen Jones, Faith Hartley, Dr. Joseph Harsh

**A closer look at interactions and activities within undergraduate research experiences through weekly journals**

P43 John Nguyen, Jonathan Popham, Dr. Joseph Harsh

**The Influence of Kinesthetic Games on College Students' Mental and Physical Well-being**

P44 Kathryn McGee, Dr. Patrice Ludwig

**The Effect of Artificial Substrate on Initial Eastern Oyster Growth**

P45 **g** Sarah McGrath, Charles Holmes, Connor Keelan, Dr. Morgan Steffen

**Effect of nutrient amendments on lake microbial communities**

P46 Megan Coceano, Guadalupe Reynoso, Emily Walsh, Dr. Morgan Steffen

**Characterization of freshwater microbes relevant to cyanobacterial harmful algal blooms**

P47 **C** Sophie Jurgensen, Charles Holmes, Dr. James Herrick

**Whole-genome epidemiology of environmental *Salmonella*: optimization and integration into an undergraduate course**

P48 \* Sean Gay, Roxana Behrooz, and Dr. Mark Gabriele

**Double-labeling neuroanatomical staining protocols for differentiating auditory midbrain neuronal subpopulations**

P49 Joy Zhang, Dr. Terrie Rife

**The effect of alpha-synuclein on gene expression in SK-N-MC cells.**

P50 Rebecca J. Livermore, Ranjit Z. Virk, Dr. Katrina Gobetz

**A comparison of diet between fossil and modern equids using plant remains from dental plaque**

P51 \* Cecilia Rogers, Dr. Heather Griscom

**Investigating the ecology and habitat modeling for *Solanum conocarpum* on St. John, USVI** *(moved to Thursday's poster session)*

P52 Anna Nordseth, Dr. Heather Griscom

**Coffee carbon stocks, pests and diseases under varied shade management: a review** *(moved to Thursday's poster session)*

P53 \* Charles Holmes, Dr. Heather Griscom

**Variation in mycorrhizal colonization of american ginseng (*Panax quinquefolius*) from varying soils and forests: a preliminary study**

P54 Shelby Saroka, James Collins, Dr. Chris Lantz

**Establishment of Genotyping Protocol for Interleukin-3 Transgenic Fluorescent Reporter Mice**

P55 Marina Barmanova, Dr. Tim Bloss

**Role of BTF3/ICD-1 in control of cancer-associated gene expression in *C. elegans***

P56 Sydney Ashton, Dr. Rocky Parker

**Searching for sex differences in snake skin**

P57 Monifa Williams, Zachary Schuhmacher, Patrick Kilkenny, Klebert Feitosa, Dr. Roshna Wunderlich

**Characterization of intrinsic foot muscles using novel Polydimethylsiloxane  
PDMS methodology and EMG measurements**

P58 **g** Elizabeth Rogers, Stephanie Sharpes (co-first authors), Dr. Kyle Seifert

**The Antibacterial and Biofilm Disruption Activity of Novel Tris-Cationic Amphiphiles**

P59 Morgan Hennessy, Dr. Oliver Hyman, Dr. Elizabeth Doyle, Dr. Ray Enke

**Integration of Geospatial Analysis into Biology Undergraduate Curriculum**

**Abstracts ORAL PRESENTATIONS**

**T1** Sara Schoen and Dr. David McLeod

**Defining Characteristics and Class Sizes Within the *Limnonectes kuhlii* Complex using Statistical Analysis of Morphometric Traits**

Reliable methods for delineating species’ boundaries are essential to the identification, assessment, and, ultimately, the conservation of global biodiversity. This is especially true for those taxa that comprise species complexes, and for which confounding morphology may preventing us from recognizing the true diversity of a region. Historically, morphometric analyses of linear body measurements have been used to assess differences between species. In some cases, however, these data may be skewed due to sexual dimorphism, hypertrification, or age. The *Limnonectes kuhlii* complex, a group of Southeast Asia fanged frogs, exemplifies this conundrum with a general male-biased size dimorphism and the hypertrophy of the head in males. This study evaluates two different measurements for assessing “body size” in the context of species delineation. The historically used character of snout–vent length is compared to body length as possible way to begin delineating boundaries within the *L. kuhlii* complex and as a means for testing the idea that size classes exist within this group. Results of this study suggest that body length is less biased and may have broader application in morphometrics than snout–vent length. Results of body size analysis also demonstrate that generalized size classes exist within the *L. kuhlii* complex and that these can be used in species delimitation.

**T2** **g** Peter Riley and Dr. Mike Renfroe

**Development of Media for in vitro Symbiosis between *Burkholderia phytofirmans* and *Dionaea muscipula***

*In vitro* biotization with plant growth promoting rhizobacteria (PGPR) can reduce the stresses associated with plant tissue culture propagation and acclimatization of plants to normal humidity outside tissue culture vessels. *Dionaea muscipula* (venus flytrap) is an endangered plant primarily propagated through tissue culture and thus a good candidate for biotization.  The PGPR *Burkholderia phytofirmans* strain: PsJN was observed to have a negative interaction with *Dionaea muscipula* *in vitro* characterized by a mucoid phenotype in the bacteria and ensuing plant tissue necrosis.  An alternative media using maltose instead of sucrose was developed to address this problem and tested alongside the standard sucrose based Venus flytrap growth medium. Biotized flytraps grown in maltose media as opposed to sucrose media produced significantly more propagules and biomass over the observed time (140 days).  Also no visible biofilm was observed on the maltose media in contrast to sucrose media all of which exhibited biofilm production by the bacteria.

**T3** \*Milos Lesevic, Grace Bauer, Dr. Terrie Rife

**Nuclear alpha-synuclein and its impact on transcription of Nitric Oxide Synthase I**

Parkinson’s disease (PD) is a chronic, progressive neurodegenerative disease affecting approximately 50,000 Americans and over 10 million people worldwide. PD is characterized by stiffness of limbs, postural instability, loss of balance, and deposits of a protein called alpha-synuclein (a-syn) into Lewy Bodies. The onset of symptoms is caused by the loss of dopamine-producing neurons in the substantia nigra. One of the proteins that is implicated in the neuronal death process is nitric oxide synthase I (NOS1). NOS1 produces a neurotransmitter called NO that mediates dopamine neurotransmission when produced in normal quantities but when produced in high quantities it causes neuronal death. Studies of post-mortem PD brains have shown that NO neurotransmission in basal ganglia is altered. Interactions between NOS1 and the a-syn proteins are not well understood. However, preliminary results in our lab indicate that a-syn may act as a transcription factor that regulates NOSI. Presence of a-syn impacts expression of NOSI promoter cloned in front of a luciferase reporter gene and expressed in human SK-N-MC cells. Our research has verified and attempted to further elucidate how alpha-synuclein works to control NOS1 expression

**T4** Heather Maher, Dr. David McLeod

**Delimiting frog species using head-shape analyses**

In contemporary systematics, molecular and morphometric data are commonly used for lines of evidence when delineating species boundaries. Morphometric data are typically derived from discrete linear measurements of morphological characters. Whereas these linear data capture much of the relevant morphological differences between taxa, they do not allow us to utilize shape-based characters effectively. This research explores the use of landmark and shape-based morphological data (geometric morphometrics) in quantitative analyses. Geometric morphometrics was applied to members of the *Limnonectes kuhlii* complex to determine whether head shape could be used to determine differences between species. In addition, qualitative data was transformed into quantitative data that could be subjected to standard statistical analyses. Results of this study demonstrated that there was a statistically significant difference in head shape between three clades. This additional morphological data was used along with body measurements and molecular data to identify a new Cambodian species in the *L. kuhlii* complex.

**T5** Zachary Casey, Dr. George Vidal

**Capturing ex vivo electrophysiological data from Cerebral Cortex**

Neurons are dynamic, electrically excitable cells that have exquisite molecular control over their synaptic function. Synapses are often dysregulated in developmental neurological disorders such as autism spectrum disorder and schizophrenia, as well as in cases of dementia such as Alzheimer's disease. To understand the cellular function of the molecules that regulate synaptic function, we seek to acquire functional data from individual neurons and their synapses using whole-cell voltage-clamp electrophysiology. Here, we engineer a "rig" to capture ex vivo electrophysiological measurements from individual neurons of the cerebral cortex. Our rig is built to detect various dynamic processes of the neuron, including miniature postsynaptic currents, synaptic potentiation, and synaptic depression. These data can be used to understand synaptic plasticity and the balance of excitation and inhibition in the cerebral cortex, especially in models of neurological disorders and dementia.

**T6 C** Grant Rybnicky, Dr. Steven Cresawn

**Repeaterator: a tool for visualizing DNA repeat motifs in Actinobacteriophage genomes**

Horizontal gene transfer plays a large role in microbial genetic diversity. Bacteriophages can facilitate this diversity through transduction, the uptake and dispersal of microbial DNA between bacterial hosts. However, bacteriophages themselves experience horizontal gene transfer through mobile genetic elements and recombination. Unlike their hosts, bacteriophages cannot easily be mapped onto a phylogenic tree as they do not all possess a common trait like the 16s RNA gene. There are tools to compare bacteriophages such as Gepard and Phamerator that compare nucleotide identity across bacteriophage genomes, but none that fully capture evidence of horizontal gene transfer and display it in an intuitive manner. Programs to identify repeat motifs also exist, but many lack the ability to display genomic information and regard repeats in an isolated manner. To address this problem, I have developed Repeaterator, a tool to visualize DNA repeat motifs within Actinobacteriophage genomes. Much like Phamerator, Repeaterator displays bacteriophage genomes and their annotations visually using the D3.js library from a Mongo database. Instead of comparing multiple genomes, Repeaterator compares a genome to itself to map the occurrence of DNA repeat motifs in the context of gene annotations. Additionally, other genomic information can easily be overlaid on the visualization, including GC content or the direction in which genes code. Repeaterator provides powerful new insight into the evolutionary origins of Actinobacteriophage genomes and can be easily adapted to analyze other genomes.

**T7** **g** Kelly Livernoche, Dr. Heather Griscom

**The effect of cattle management on soil carbon; implications for climate change**

In naturally occurring ecosystems, forests function as substantial carbon sinks, storing carbon in soil and biomass that would otherwise exist in the atmosphere as carbon dioxide. However, agricultural fields have the potential to act as reservoirs of soil carbon as well. Rotational cattle pastures, where cattle are moved between enclosed sections of pasture, may improve soil carbon content compared to conventional pastures. In rotational cattle pastures, a more even distribution of manure increases plant biomass, and increased cattle movement decreases soil compaction, thereby reducing erosion and loss of soil carbon. This study quantified differences in soil carbon and bulk density (soil compaction) within and between a high-frequency rotational pasture (HFR), a low frequency rotational pasture (LFR), and a conventional non-rotational (NR) pasture. Soil samples were collected from top, middle, and bottom slope positions and were separated by soil depth (0-10, 10-20, and 20-30 cm). Bulk density was determined using dry soil weights, and soil organic carbon was analyzed with the loss-on-ignition technique. Soil carbon was greater overall in the HFR (6.58%) and LFR (6.68%) pastures compared to the NR pasture (3.47%; p < 0.001). Bulk density was also greater in the NR pasture (0.96 g/cm3) compared to the HFR (0.79 g/cm3) and LFR (0.80 g/cm3) pastures (p < 0.001). There was no difference in soil carbon (p = 0.94) or bulk density (p = 0.62) between the HFR and LFR pastures. Soil carbon was also affected by soil depth in all pastures (p < 0.001). Soil carbon was not related to slope position in the NR pasture, as was expected (top: 3.30%, middle: 3.30%, bottom: 3.29%; p = 0.24). This study suggests that rotational cattle pastures could be one pathway for mitigating climate change through greater carbon sequestration and soil carbon storage.

**T8** Romie D. Powell, Dr. Justin Brown

**The Role of Ambient Temperature in the Recovery from Major Surgical Stress in Rats**

Laboratory animals routinely undergo surgical instrumentation for experimental use and housed at ambient temperatures ranging from 20ᴼC - 26ᴼC during recovery per recommended guidelines. However, rats typically prefer ambient temperatures (Tamb) of ~27 ᴼC (Brown and Le 2011). Rats are often housed at normal room temperature (~22 ᴼC). While this is comfortable to those maintaining them, it may lead to a cold thermal stress for the rats. It is hypothesized that housing rats at ambient temperatures away from their preferred Tamb could lead to a thermal stress which adversely affects surgical recovery. To address this, adult rats (220g-320g) were surgically instrumented with a radiotelemetry probe (DSI), which allows non-invasive measurement of core temperature (Tc). A cannula (21ga) was also inserted into the brainstem to allow microinjection of drugs as part of a separate project. For >1 week of recovery, the rats were housed at either 21ᴼC, 24ᴼC, 27ᴼC (control), 30ᴼC, or 33ᴼC Tamb while Tc, food and water intake, and body weight changes were measured. Rats housed at 21ᴼC and 24ᴼC struggled to recover a normal Tc initially. This delayed the onset of normal circadian cycling in the 21ᴼC group. Rats housed at 33ᴼC recovered quickly and maintained a slightly elevated Tc compared to control (27ᴼC). The 30ᴼC and 33ᴼC groups reestablished Tc circadian rhythms faster than the control, but the 33ᴼC struggled to maintain that rhythm near the end of the recovery week. The return of body weight to pre-surgical levels was delayed in rats housed at the warm Tamb (30ᴼC & 33ᴼC) or the very low Tamb (21ᴼC). The effect of Tamb on daily food and water intake after surgery was not remarkable except for on the 5th day of recovery in the 30ᴼC and 33ᴼC groups when water intake increased. The 21ᴼC and 24ᴼC increased food intake after the 4th day in comparison to the control (27ᴼC). These data suggest that rats maintained at 27ᴼC recovered from surgical stress more readily. These rats returned to pre-surgical body weight more quickly and demonstrated a normal thermoregulatory circadian rhythm earlier than the cold (21 or 24ᴼC) or warm (30ᴼC or 33ᴼC) rats. Rats housed at > 30ᴼC may have been exposed to a heat stress which affected weight gain and surgical recovery initially and impaired normal circadian cycling later in the week. Rats housed at 21ᴼC may have been cold stressed which also affected weight gain and thermoregulatory recovery. It is suggested that rats be maintained at their preferred Tamb of 27ᴼC during the week following surgery to minimize thermal stress and thereby facilitate recovery. This reduction in stress facilitates the return to a normal physiologic state and consequently enables more reliable data collection from these animals.

**T9 g** Jessie Mandirola, Dr. Patrice Ludwig

**Oyster Restoration Utilizing Artificial Substrates: Implications for Mitigating Oyster Decline in the Chesapeake Bay**

The high national priority of developing artificial oyster reef substrates is a consequence of scarcity and consequential expense of natural oyster shells available for restoration. Restoration is imperative because the historical abundance of native oyster populations has declined 99% over the past 100 years. A common artificial reef substrate is concrete made with silica sand. However, conventional concrete results in reduced long-term oyster survival and increased competition with other organisms like barnacles. The aim of this research is to test alternative concrete formulations in oyster restoration efforts. Specifically, we field-tested the statistical hypothesis that wild juvenile oysters would strike to and grow on concrete casts of oyster shells made with limestone sand as often as striking to casts made of concrete with silica sand because limestone sand has elevated levels of calcium, which is known to attract oyster larvae for settlement and growth. Natural oyster shells were used as a control and reference substrate. An initial Kruskal-Wallis post hoc test suggests that after one spawning and recruitment period (14 weeks, 2016), shell casts made with limestone sand attracted similar abundances (149, n=80) of oyster spat as normal concrete casts (174, n=80). However, both shell cast varieties attracted fewer spat than natural oyster shells (365, n=80; p < 0.001). Over time, greater settlement on the natural shell led to increased mortality due to competition for space and resources. Therefore, the settlement on the concrete formulations was more stable and could lead to greater long-term success compared to the natural shells. The long-term effects of the limestone-rich concrete formulation are currently being examined, however, this study suggests that concrete that benefits oyster spat can be advantageous as an alternative substrate for oyster restoration due to similar abundances found on both formulations. We suggest that future restoration efforts should consider using alternative substrates that provide benefits to oyster spat and help to reduce overall cost while maintaining optimal recruitment success.

**T10 C**\* Courtney Stout, Nick Dunham, Dr. Chris Berndsen, Dr. Raymond Enke

**Epigenetic regulation of photoreceptor-specific transcription factor binding in the genomes of retinal neurons**

The retina is a neuronal tissue lining the back of the eye containing rod and cone photoreceptors that make vision possible. Highly regulated transcriptional networks control differentiation and maintenance of photoreceptors in the retina. DNA methylation of cytosine bases in genomic DNA is an epigenetic modification correlated with repression of gene expression. Currently in our lab, the biochemical relationship between DNA methylation and the ability of retina-specific transcription factors to bind in the genome is being studied. These transcription factors, known as cone-rod homeobox (CRX) and neural leucine zipper (NRL), have been shown to act synergistically to control photoreceptor expression in the retina. Preliminary data has supported the hypothesis that DNA methylation is critical for modulating cell-specific binding of CRX to target recognition sites. DNA binding domain coding sequences of the human CRX and NRL proteins were cloned into bacterial expression vectors, and were expressed and purified for downstream biochemical analysis. In vitro gel shift assays are currently being used to determine the ability of CRX and NRL to bind unmethylated and methylated oligonucleotides. Collectively, these studies will contribute to a better understanding of how epigenetic modifications influence the development, homeostasis and pathology of human retinal neurons.

**T11** Grant Rybnicky

**Qualification of qPCR positive control at Merck Elkton**

Merck & Co., Inc. is a global healthcare company headquartered in Kenilworth, NJ that generated $39.8 billion in revenue in 2016. With locations worldwide, the Company has a manufacturing facility located in Elkton, VA. Originally a chemical manufacturing facility, the Elkton site has been converting to bioprocessing. Through the Future Talent Program’s internships and Co-Ops, the Company provides opportunities for undergraduate students to gain industry experience in bioprocessing prior to graduation. Within any bioprocess, it is imperative to maintain system control to ensure safety and quality of the end product. System control is measured through critical process parameters which can have an ultimate effect on critical quality attributes (CQAs). CQAs directly determine the characteristics of a drug substance (DS) or end product. Residual DNA is a CQA of many products produced through bioprocessing, and can cause allergic reaction and other immune responses in patients. To measure this CQA, DNA from DS and in-process samples is extracted and then quantified by qPCR. Extraction reagents must be qualified prior to measuring residual DNA concentration to ensure proper functionality. A positive control substance was created, verified as a positive control, and the range of a positive result determined to address this. This positive control was qualified by two analysts using multiple manufacturing lots of extraction reagents. Upon completion of this protocol, samples can be tested for residual DNA and DS can be released for use in patients.

**T12** **C**\* Claire Gormley, Kristen Hoffman, Dr. Tim Bloss

**Characterization of Cell-type Specific Responses to Misfolded Protein Stress in *C. elegans***

Cells experiencing misfolded protein stress can become debilitated and die, consistent with this stress being linked to numerous diseases, including neurodegeneration. When misfolded proteins accumulate in the endoplasmic reticulum (ER), the unfolded protein response (UPR) initiates mechanisms that resolve this stress or trigger apoptotic cell death, dependent on the severity and/or duration of the stress. The nascent polypeptide-associated complex (NAC) is a heterodimeric chaperone that mediates proper protein folding and localization during translation; depletion of the NAC promotes misfolded protein stress in the ER, resulting in the initiation of the UPR in affected cells. The relationship between the NAC and the UPR is not well understood, nor is it known if this relationship differs depending on cell type. Our goal is to characterize this relationship in the model organism *C. elegans*, where cell lineages display variable sensitivities to misfolded protein stress. Via RNA interference, we are depleting the NAC in worm strains expressing cell type-specific fluorescent proteins and characterizing the nature, number and position of these cells throughout the life of the worm. Depletion of the NAC in worms expressing neuron-specific red fluorescent protein decreased the number of observable neurons in the ventral nerve cord while mislocalizing a subset of the neurons that remained. Concordantly, affected worms displayed movement defects consistent with disruption of ventral nerve cord function. Conversely, depletion of the NAC in worms expressing hypodermal cell-specific yellow fluorescent protein (YFP) resulted in an increase in YFP-positive cells starting early in embryogenesis and continuing into adulthood, potentially explaining why worms depleted of the NAC were very often misshapen in a way consistent with defective hypodermal cell function. Both the neuronal and hypodermal cell results are consistent with previous findings in *C. elegans* showing neurons more susceptible to death during misfolded protein stress relative to other cell types, e.g. hypodermal cells. Through these experiments, we hope to better understand the role of the NAC during misfolded protein stress response, and whether this role changes relative to the cell type experiencing the stress.

**T13** Roxana Behrooz, Sean M. Gay, Dr. Mark Gabriele

**Complementary multimodal compartments in the developing midbrain**

The auditory system is responsible for encoding hearing. Research in our laboratory focuses on the inferior colliculus (IC), a major relay hub situated in the midbrain, that is subdivided into a central nucleus and surrounding dorsal and lateral cortices. Recent studies show that the lateral cortex of the inferior colliculus (LCIC) is actually multimodal, receiving inputs from not just auditory sources, but also somatosensory and visual structures. The precise organization of the patterned inputs to LCIC and their development has yet to be fully established. Mounting evidence suggests a modular framework with surrounding extramodular zones that provide an anatomical substrate for input-output arrays. Previously, we identified acetylcholinesterase (AChE), cytochrome oxidase (CO), glutamic acid decarboxylase (GAD), nicotinamide adenine dinucleotide phosphate-diaphorase (NADPH-d), and parvalbumin (PV) as discrete markers of LCIC layer 2 modular fields. The present study builds upon these findings and establishes calretinin (CR) as a complementary extramodular marker. CR-specific labeling was observed in LCIC zones surrounding presumptive layer 2 modules at all ages, yet became increasingly more distinct at later developmental stages. This finding somewhat contrasts previous results in developing rat in which LCIC CR patterns were more evident prior to hearing onset (Lohmann and Friauf, 1996). NADPH-d and CR double-labeling confirms a complementary modular/extramodular LCIC substrate that is established during the early postnatal period. Ongoing studies aim to determine the alignment of similarly organized Eph/ephrin guidance expression patterns in relation to these markers and developing multimodal projection patterns.

**T14 C** **g** Curtis Kapsak, Kevin Libuit, Erika Gehr, Dr. James Herrick

**Determining the prevalence of antibiotic resistance plasmids found in native stream bacteria in the Shenandoah Valley**

Plasmids in agriculturally-impacted bodies of water may play a significant role in the dissemination of antibiotic resistance (AR). High bacterial loads in stream sediment and selective pressures introduced by agricultural practices may facilitate the exchange and recombination of genetic material, creating reservoirs of AR genes that can potentially be accessed by fecal and other animal and human pathogens. Transmissible, tetracycline-resistance plasmids were captured “exogenously” from stream sediment samples by conjugating stream sediment bacterial cells with a rifampicin-resistant strain of Escherichia coli. One plasmid, pEG1-1, conferred resistance to tetracycline, tobramycin, kanamycin, ticarcillin, piperacillin, piperacillin-tazobactam, and cefepime. A method to sequence multi-drug resistance plasmids using both Oxford Nanopore MinION and Ion Torrent Personal Genome Machine sequencers was developed to sequence plasmid pEG1-1. A hybrid assembly of the sequence data from both platforms generated a single 73,320 bp contig. Analysis of the genome revealed pEG1-1 to be an IncP-1β plasmid with two mobile genetic elements – a tn21-related transposon and an in104 complex integron – both of which carry multiple antibiotic resistance genes. In the past year, a total of 85 tetracycline-resistance plasmids have been exogenously captured from native stream bacteria among three different sampling sites along Cook’s Creek. These plasmids will be tested for their conferred antibiotic resistances and a subset of these plasmids will be sequenced on both the Oxford Nanopore MinION and an Illumina MiniSeq for further genomic analysis.

**T15** Gregory Mansour, Melissa Encinias, Matthew Riordan, Cody Cubbage, Dr. Dean Cocking

**Persistence and Sustainability: Trace Mercury in “Uncontaminated” Rockingham County VA Terrestrial Ecosystems Represent a Widespread Pattern of Occurrence**

Forest ecosystems in the Shenandoah Valley of Virginia are not directly exposed to major sources of Hg contamination. Rockingham Co was assumed to be suitable as a low level control in comparison with sites exposed to major industrial contamination in Waynesboro VA for studies in the late 1900’s. Subsequently the presence of low level Hg from regional background sources has been demonstrated. This study was conducted from 2014-2016 and expanded from two sites in to four sites in 2016. A composite air index, soil, and decaying organic material “duff” were analyzed for total Hg concentrations using a Perkin Elmer Flow Injection Spectrophotometer dedicated to Hg analysis. The samples were digested in hot concentrated nitric and sulfuric acid. The results demonstrated a detectable amount of Hg was present within the various samples with extensive variability. This prompted the question of whether there is an ambient level of mercury in terrestrial ecosystems. An extensive literature search aggregated Hg values from control sites used in studies globally. Comparing the findings from the Rockingham Co. sites indicated that the Hg presence is below the global average for control sites yet remains detectable. None of the concentrations attained at these sites are great enough to be considered a health hazard. Annual sampling is needed to provide longitudinal data to account for seasonal variation and identify long-term trends.

**T16** \*Melissa Encinias, Gregory Mansour, Matthew Riordan, Cody Cubbage, Dr. Dean Cocking

**Detectable Traces of Mercury Exist Within Invertebrate Food Webs of Low Contamination Shenandoah Valley Terrestrial Ecosystems**

Mercury (Hg), in the form of inorganic and methyl mercury, has been increasing in the environment due to airborne deposition. While this increase can be attributed to both natural and anthropogenic sources, Hg exposure is known to negatively impact biological systems, even at low levels. Airborne Hg pollution puts environments, previously thought to be uncontaminated, at risk. To identify the possible long term implications of Hg accumulation in “uncontaminated” terrestrial environments, sites, in and around Rockingham Co. VA, were sampled over the summers of 2014, 2015, and 2016. At each site, habitat and invertebrate samples were analyzed for Hg concentration using a Flow Injection Mercury System (FIMS). Analysis revealed Hg concentrations, in both the habitat and invertebrate samples, above a zero level but below the concentrations of contaminated ecosystems. Results from the first year supported the prediction of increasing Hg concentrations in invertebrates based on bioaccumulation patterns. Additional analyses of subsequent invertebrate data verify presence under background conditions. The results demonstrate a need for further study of Hg accumulation in “uncontaminated” regions as a baseline for potential increased presence over time.

**T17 g** Elizabeth Rogers, Dr. Kyle Seifert

**The Antibacterial and Biofilm Disruption Activity of Novel Amphiphiles**

Approximately 23,000 people die every year in the United States as a direct result of antibiotic resistant infections caused by organisms such as Pseudomonas aeruginosa and Staphylococcus aureus. The risk of transmission, particularly among hospital patients, is increased by the formation of bacterial biofilms, which protect pathogens against chemical removal. Several novel series of amphiphiles have been synthesized, and their antimicrobial activity was tested against seven different bacteria, including P. aeruginosa, Escherichia coli, and S. aureus. The minimum inhibitory concentrations (MIC) of these compounds were compared in order to determine the effects of structural change on antimicrobial activity. The ideal size of the hydrophobic tail region was 12 carbons per tail for all tris-cationic double-tailed series, with MIC values as low as 4 micromolar for Gram-negative species and 2 micromolar for Gram-positive species. The most effective compounds were used in a crystal violet stain biofilm disruption assay. The 12-carbon tail derivatives disrupted up to 70% of established P. aeruginosa biofilms as efficiently as tobramycin at similar concentrations, approximately 63 micromolar. Continued research into these compounds as novel antimicrobials will include study of synergistic effects, mechanism of action and efficacy against new emerging pathogens.

**T18** Naomi Gilbert, Dr. Steven Wilhelm, Dr. Morgan Steffen

**Survey of microbial urea degrader diversity in Lake Shenandoah and the Shenandoah River**

One of the primary drivers of cyanobacterial harmful algal blooms (cHABs) in freshwater systems is nutrient loading, particularly of nitrogen and phosphorus. There has been an increased focus on assessing the role of nitrogen (N) in freshwater lakes and rivers that suffer cHABs. Urea, a widely-used, N-rich fertilizer, is a source of interest due to its abundance in freshwater ecosystems, primarily caused by anthropogenic nutrient loading. While recent work has shown that cHAB population succession may favor the toxic cyanobacterium *Microcystis*in urea-rich waters, the diversity of the associated bacterial community capable of degrading urea has yet to be determined. Therefore, we generated targeted sequence libraries of the gene encoding for the alpha subunit of the urease enzyme, *ureC*, from samples collected during summer (2015) from two model freshwater systems, Lake Shenandoah (LS) and the Shenandoah River (SR) (Virginia), to reveal potential urea-degrading members in threatened freshwater ecosystems. The total microbial community with urea-degrading capabilities was dominated by Proteobactiera in all samples, while Cyanobacteria was present in low abundance. This may be a result of the physical environment of LS and SR, or the low abundance of Cyanobacteria may be due to limitations in the custom database constructed for *ureC*amplicon analysis. LS and SR communities were similar overall, with the exception of a higher relative abundance of Acintobacteria *ureC* sequences in SR.  Further analyses will aim to characterize more members with the *ureC*gene and expand upon the foundation built for community analyses based on*ureC.* These results provide better insight into the diversity of an important gene involved in bacterial urea degradation needed to resolve the microbial freshwater urea cycle.

**T19 g** Harmon, Dr. Christine May

**Revealing the Current Relationship between Stream Acidification and Fish Species Richness: What is the Status after Two Decades of Recovery?**

Prior to limitations placed on atmospheric emission of sulfur dioxide, Shenandoah National Park streams were heavily impacted by acid deposition. Acidification of these streams resulted in a depletion of acid neutralizing capacity (ANC) and a loss of fish species diversity. Differences in bedrock composition of park watersheds produce variation in stream response to acidification such that siliciclastic watersheds provide the lowest ANC, basaltic-carbonate watersheds provide the highest ANC, and granitic watersheds provide an intermediate ANC. Previous results by Bulger et al. (1995) indicated a significant relationship between ANC and fish species richness in thirteen park streams, such that low ANC streams in siliciclastic watersheds support less fish species diversity than relatively higher ANC streams in granitic and basaltic-carbonate watersheds. Given the potential for acidification recovery in the past two decades, has the relationship between fish species richness and ANC changed? The current study investigated changes in this relationship from 1995 to 2016 in the same thirteen park streams distributed among the three major bedrock regions of the park. Increases in fish species richness primarily occurred in well-buffered streams underlain with granitic and basaltic-carbonate bedrock while minimal increases in fish species richness were observed in siliciclastic watersheds. The strong correlation between fish species richness and ANC observed in 1995 still exists in 2016, indicating that park streams may still be experiencing legacy effects of past acidification.

**T20** \*Giavanna Verdi, Taylor Berrena, Miriam Kabore, Rachel Pallister, Mara Gaykema, Dr. Corey Cleland

**Rat hind limb nociceptive withdrawal response to heat and mechanical stimuli depends on initial position of the paw but not stimulus location**

Rats rapidly withdraw their hind limb in response to noxious stimulation, which is known as the nociceptive withdrawal response (NWR). The first aim of my project was to determine whether the location of mechanical stimuli and the initial position of the paw preceding the NWR influenced the direction of the NWR in intact, unanesthetized rats. Second, after determining that the direction of the NWR depended on the initial position of the paw, we tested whether the rat used proprioceptive sensory feedback or corollary discharge to identify the position of the paw prior to stimulation. Based on previous studies, we hypothesized that the response would be both directly away from the location of the stimulus and also depend on initial paw position, with the latter arising from proprioceptive sensory feedback rather than corollary discharge. In the first set of experiments, rats were stimulated with localized mechanical (“Von Frey” monofilament or 30-gauge needle) stimuli to five spots widely distributed over the plantar surface of the hind left paw. The NWR was quantified as a vector between the initial and final position of the stimulated paw. Unexpectedly, stimulus location did not significantly influence the direction of the response, falsifying our hypothesis. However, the initial position of paw was variable, suggesting an influence on the direction of response. Correlation between the initial location and the change in location rostral/caudally and lateral/medially revealed a significant and inverse effect on response direction. Thus, if the paw was initially rostral, it would move caudal after stimulation; if the paw was initially caudal, it would move rostral. In the second set of experiments, rats were stimulated by heat with an infrared laser to a single region of the paw, which was placed on an independently movable glass plate. The plate was repositioned rostral-caudally (forward-backward) just before evoking the NWR to dissociate proprioceptive sensory feedback from corollary discharge. The NWR was unaffected by repositioning the paw prior to the evoking the NWR, consistent with proprioceptive feedback being used to determine the direction and magnitude of the NWR. Taken together, our results suggest the CNS in intact rats primarily uses proprioceptive information about limb posture, but not stimulus location, to determine the direction of the NWR movement. Thus, the NWR appears designed to both maintain posture, as well as protect the paw from injury.

**T21** \* Matt Kohler, Dr. Amanda Storm, Dr. Jon Monroe

**Effects of the microenvironment surrounding Cys433 in Arabidopsis β-amylase-1 and -3 on the sensitivity to glutathionylation by nitrosoglutathione**

Glutathionylation is a reversible post-translational modification involving the transfer of glutathione to the thiols of specific cysteine residues in proteins. While the mechanism behind glutathionylation is known, the specificity of cysteine glutathionylation is not understood. Using β-amylases (BAMs) 1 and 3 from *Arabidopsis thaliana*, which have different sensitivities to nitrosoglutathione (GSNO), as a model, I attempted to provide insight into why some cysteines are glutathionylated by GSNO and others are not. Our lab found that GSNO inhibits BAM3 activity by glutathionylating Cys433 *in vitro*, yet BAM1 is unaffected by GSNO despite containing a cysteine at the same position. At a physiological level, the glutathionylation of BAM3 may be important as BAM3 may be inhibited by a NO-induced modification under cold stress. After comparing sequence alignments and homology models of both enzymes, I hypothesized that H430, N432, and S434 contribute to the sensitivity of Cys433 in BAM3 to GSNO, and that the corresponding amino acids in BAM1, D430, L432, and A434, contributes to its insensitivity. To test this hypothesis, I made three BAM3 mutants with the amino acid substitution H430D, N432L, or S434A and the corresponding BAM1 mutants, D430H, L432N, and A434S. All of the BAM1 mutants were active but only the BAM3-H430D mutant was active. The BAM1-D430H and BAM1-A434S mutants were inhibited by 45% and 20%, respectively, by GSNO whereas the WT BAM1 control was insensitive to GSNO. The active BAM3 mutant was just as sensitive to GSNO as the BAM3 control. Therefore, D430 and A434 may contribute to the insensitivity of Cys433 to glutathionylation by GSNO in BAM1.

**T22** \* Hayley Fried, Naomi Gilbert, Taylor Wright, Michael Gay, Dr. Morgan Steffen

**A Survey Of The Bacterial Diversity And Function In Lake Shenandoah (Virginia, U.S.A.)**

Widespread eutrophication is a recognized threat to the sustainability of ecosystem function in freshwater lakes. The impacts of associated cyanobacterial harmful algal blooms (cHABs) include toxicity, hypoxia, and disruption of the availability of potable water to surrounding communities. Factors such as loading of nitrogen and phosphorus and rising temperatures may have contributed to accumulation of cyanobacteria biomass in the model ecosystem Lake Shenandoah, located in Rockingham County, Virginia. However, limited data exist on the ecological status of the system. Therefore, we performed a study to assess the bacterial diversity of the system during summer months over the course of two years. Clone libraries of bacterial 16S rRNA and ureC, the gene encoding the alpha subunit of the ureases enzyme, were generated from samples collected in May, June, July, and August of 2015 and 2016. Phylogenetic composition of Lake Shenandoah analyzed from targeted bacterial 16S genomic data revealed the presence of bacteria related (>97% identity) to *Limnohabitans* sp., *Polynucleobacter* sp., *Clostridium perfringens*, and *Methylomicrobium alcaliphilum*. Analysis of ureC sequences revealed a mixture of Cyanobacteria and Alpha- and Betaproteobacteria capable of urea degradation in this system. Paired with nutrient and environmental data, this preliminary study provides for the first time an understanding of microbial function in Lake Shenandoah.

**T23** Mary Pegalow, Seerat Mavi, Diana Grigoryan, Corey Cleland

**Integration of limb or tail nociceptive withdrawal responses and postural body movements in unrestrained rats**

The nociceptive withdrawal response (NWR) occurs when an animal rapidly withdrawals a body part in response to a noxious stimulus. In previous studies, rats were restrained in a tube or box during stimulation of the tail or foot, but it was hypothesized that constraint may have blocked additional postural body movements. The specific aim of this study was to examine postural changes in the body of rats when delivered a noxious heat stimulus to the tail or foot in an unrestrained, as opposed to a restrained, setting. Rats were first anesthetized with isoflurane and black marks were placed on the feet, tail, and body. The animals were then placed on an open glass table with a video camera underneath to capture movement of the animal and localized heat stimuli were delivered via infrared laser to either the tail or the plantar surface of the feet. It was found that when the tail or feet were stimulated, the stimulated body part was withdrawn approximately one second following stimulation, nearly 100% of the time. When stimulating the tail, animals also tended exhibit forward body movement, as well as foot movement. When the feet were stimulated, animals tended to pitch the body forward and take additional foot movements, but no tail movement was observed. Stimulus location on the tail had no effect on the rotation of the animals, but individual animals exhibited significantly different magnitudes of angular change. In summary, rats appear to use similar local withdrawal strategies, but different postural strategies, which may be missed when rats are restrained.

**T24** Jillian Breault, Lauren Pope, Dr. Amanda Storm, Dr. Jonathan Monroe

**Characterizing the salt dependence of β-Amylase2 in Arabidopsis**

β−amylases hydrolyze starch in leaves at night to provide energy in the absence of light. Arabidopsis has nine β−amylases (BAMs), five of which are catalytically active. BAM2 is the most unique of the active BAMs, exhibiting allosteric regulation and a strict salt dependence. The optimal level of KCl for activity was about 300 mM, which is quite high but is close to the ionic strength found in the chloroplast stroma. Comparing the amino acid sequences of 15 eudicot orthologs of BAM2, we found that all contain a unique N-terminal domain enriched in aspartic and glutamic acids (26%) whereas the catalytic domains contained only 14% acidic amino acids. Thinking that this acidic domain might play a role in BAM2’s salt dependence, we created a mutant BAM2 protein lacking the N-terminal acidic domain by introducing a restriction site into the cDNA sequence and then deleting the N-terminal region. The mutant protein was then expressed in E. coli and purified using His-tag affinity chromatography. Interestingly, this mutant showed higher activity than wild type BAM2, no requirement for salt, and a lower Km for starch. This suggests that the N-terminus of ΒΑΜ2 plays an important role in regulating the activity of BAM2.

**T25 g** Phoebe Cook, Rebecca Rasmussen, Edward Hsieh, Dr. Jonathan Brown, Dr. Idelle Cooper

**Sexual selection on color variation in *Megalagrion calliphya***

Sexual dimorphism is commonly assumed to be caused by sexual selection. The Hawaiian damselfly *Megalagrion calliphya* is an excellent system in which to study the causes of dimorphism because it has a dimorphism in color within females as well as between males and females. Males are always red, while females are either red (andromorphic) or green (gynomorphic). Populations vary in the frequency of these female morphs, from monomorphic green through dimorphic to monomorphic red. We tested the hypothesis that female coloration is under sexual selection due to male sexual harassment, which we expect to be greater in populations with high male frequencies or ratios of andromorphs. We also tested two possible forms of sexual selection on female morph. If andromorphs are male mimics, we expect that they will be harassed less than gynomorphs, but if frequency-dependent sexual selection is driving the dimorphism, we expect that the more common morph in a population, whichever it is, will receive more harassment. We measured sex and morph frequencies and performed behavioral trials in five populations on Hawaii Island. We did not find a relationship between male frequency and harassment, and we did not see either predicted pattern of sexual selection. There were no significant differences between harassment rates of andromorphs, gynomorphs, and males. These results do not support either the male mimicry or frequency-dependent sexual selection explanations for color dimorphism. However, the morph frequencies do follow an elevational cline, suggesting that this dimorphism may be a result of ecological rather than sexual selection.

**Abstracts POSTERS**

**P1** Katherine M. Bland, Christopher J. Handwerk, Z. Logan Holley, Zachary O. Casey, Dr. George S. Vidal

**Exploring cerebral cortical synaptic pruning in models of Autism Spectrum Disorder**

Autism spectrum disorder (ASD) has no known cure and affects about 1 in 68 individuals in the United States. ASD is usually accompanied by an excess of synapses on cerebral cortical neurons. Around birth, the cortex normally overproduces synapses. During childhood, the brain then undergoes a period of normal development in which synapses are pruned away, leaving only synapses that are important for neural function. Our hypothesis is that deficient pruning causes the excess synapses found in ASD. Here, we explore several candidate ASD molecules that may control the timing and location of synaptic pruning on individual neurons of the cerebral cortex. By modifying the expression of these candidate molecules in individual neurons in vivo, we are able to determine the molecular mechanisms of dysregulated synaptic number in ASD.

**P2** **g** Sarah McGrath, Francis Gebhard, Walker Spradlin, Cole Roberts, Dr. David McLeod

**A review of larval amphibian field enclosure techniques and introduction of a novel field device**

Biological research sometimes requires the study of organisms in their natural environment due to the difficulty of replicating many biotic and abiotic factors in the laboratory. Amphibians are frequently used as model organisms for field studies, particularly those dealing with ecology and ecotoxicology, due to their dual life stage natural history and the manageability of keeping amphibians in semi-captive or captive conditions. The use of specific devices or techniques used to contain amphibians, especially larval amphibians which are aquatic, are found infrequently throughout the literature and vary greatly in design, cost, and use. Here, we summarize the various devices previously used in amphibian field research, address potential concerns with in-field enclosures, and provide comprehensive comparisons to assist further research in this area. We also introduce a novel in-field device that is cost effective and easily transportable as a new alternative to aid future anuran studies.

**P3** Jacob Gumpf, Behan Alavi, Dr. Oliver Hyman, Dr. David McLeod

**Detection of *Batrachochytrium dendrobatidis* but not *B. salamandrivorans* in**

**Bornean frog populations**

The pathogenic fungi *Batrachochytrium dendrobatidis* (Bd) and *B. salamandrivorans* (Bs) have both been shown to cause the disease chytridiomycosis in amphibians and have been implicated in worldwide declines of amphibian populations. Surveys conducted to determine the distribution and host range of these fungi have been focused on regions in the Americas and Europe, with less attention given to South East Asia. This study surveyed amphibians from Brunei and Malaysia on the island of Borneo, a hotspot of amphibian diversity where these fungi have not been detected. 203 anurans representing 49 different species were swabbed and tested for the presence of both Bd and Bs using qPCR. Of the 203 samples [DM1] tested, none tested positive for Bs (0% prevalence, 95% confidence intervals of 0.0-0.4), and three individuals tested positive for Bd (1.5% prevalence, 95% confidence intervals of 0.0 ≤ 0.01 ≤ 0.04). The three Bd positive samples came from 3 different species (*Limnonectes kuhlii*, *Ansonia longidigita, Meristogenys whiteheadi*) collected in the Malaysian state of Sabah from an area of high tourist visitation. Further studies on host distribution, the prevalence of Bd, and the effect of Bd on Bornean amphibians are necessary to prevent a potential loss of Bornean amphibian diversity.

**P4** Taylor Hutchinson, Rana Ihsan, Dr. Tim Bloss

**Characterization of misfolded protein stress inducers in the different cell types of *C. elegans***

Various stressors can cause proteins to misfold and aggregate, often leading to cell damage and even cell death. To manage this stress, the cell employs mechanisms that mitigate misfolded protein levels, including the presence of chaperones, proteins that help other proteins fold properly. The nascent polypeptide-associated complex (NAC) is an a/b heterodimeric chaperone that assists in the folding and localization of nascent polypeptides to the Endoplasmic Reticulum (ER) during translation. Depletion of the NAC in *C. elegans* results in numerous cell-specific phenotypes, including a strong induction of the ER-specific unfolded protein response (UPR), which can either save the cell or initiate cell death by apoptosis. To better understand the nature of the stress induced in the ER when the NAC is depleted, we are comparing phenotypes seen in NAC-depleted *C. elegans* to those observed in worms treated with ER stressors with known mechanisms of action, i.e. thapsigargin and tunicamycin. We are observing stress effects in fluorescently marked hypodermal cells, specifically the viability and number of hypodermal cells in embryos. We will perform similar analyses in neurons to determine cell-specific responses to NAC depletion. Misregulation of the NAC has been associated with numerous diseases; a better understanding of the NAC’s role in protein stress management may help us understand why.

**P5** Lauren Pope, Jillian Breault, Dr. Jonathan Monroe

**Quaternary Structure of β-Amylase 2 in *Arabidopsis thaliana***

*Arabidopsis thaliana* contains nine β-amylase genes (BAMs) that play various roles in starch degradation. Former reports show BAM2 is catalytically inactive, yet we have contradictory evidence that it is active in the presence of high, yet physiological levels of salt (KCl). Once we found that BAM2 is active, we proceeded to characterize BAM2 with enzyme activity assays, which displayed sigmoidal kinetics and cooperativity with a Hill coefficient of 3.4. BAM2’s cooperative kinetics led us to believe that it is an oligomer, and through Multi-Angle Light Scattering (MALS) analysis we revealed that recombinant BAM2 is a tetramer. By studying a homology model of the hypothesized BAM2 tetramer, we identified several highly conserved residues on the surface of BAM2 that appear to bind to adjacent monomers. To test the accuracy of the homology model, we used QuikChange mutagenesis to generate a D490R mutant of BAM2 in one of these conserved regions. MALS data showed that the BAM2 D490R mutation disrupted tetramerization. We conclude that the D490R mutant probably altered BAM2’s secondary structure in this region, preventing a stabilizing interaction that would usually allow for tetramerization. This mutant was also catalytically inactive, leading us to conclude that BAM2 requires its tetrameric quaternary structure to be catalytically active.

**P6** Graham Price, Anne Masters, and Dr. Marquis T. Walker

**Identifying the genetic locus and characterizing the pathophysiology of *Drosophila* retinal degeneration mutant, rdgI**

The *Drosophila melanogaster* visual system has long been used as model to study light-dependent visual signaling. Our laboratory is interested in identifying novel factors that are essential in photoreceptor homeostasis. Here we are studying a *Drosophila* mutant called Retinal degeneration I (RdgI). The RdgI mutant line was isolated in a large scale mutagenesis screen focused on identifying eye-specific genes. As the name indicates, these mutants display severe retinal degeneration in adult flies. Currently the locus of the RdgI mutation in the genome is unknown. To target this site we are using a strategy of deletion mapping to determine its location and whether it corresponds to a known eye-specific gene. In addition we are measuring rhodopsin protein expression and imaging eye morphology in developing adults to characterize the photoreceptor pathology in RdgI mutants. Through this study we will identify the gene disrupted in RdgI flies and uncover its necessary role in photoreceptor maintenance.

**P7** Madeline Henwood, Shaw Camphire, Dr. Terrie Rife

**Characterizing tau in the nucleus**

A major hallmark of Alzheimer’s disease is the aggregation of hyperphosphorylated tau into neurofibrillary tangles. Tau (tubulin-associated protein) has recently been shown to bind to purine-pyrimidine repeats in DNA and stabilize them into Z-DNA. Evidence from our lab suggests that the binding of tau to purine-pyrimidine repeats causes transcriptional changes of the NOS1 gene, which has a purine-pyrimidine polymorphism in its promoter region. Six major isoforms of tau exist, and an imbalance of those isoforms coincides with changes in NOS1 expression and a build up of Z-DNA in severe AD patients. Based on studies of mouse tau isoforms and our lab’s previous work, we hypothesize that the 4R tau and 3R tau isoforms will be found in equal ratios in the normal nucleus of human neuronal cells, and that both 4R and 3R tau will bind directly to the purine-pyrimidine polymorphism to regulate NOS1 transcription. Two studies will be completed to test this hypothesis. The first study will determine the isoforms of tau normally found in the nucleus of human SK-N-MC cells. To test this, western blotting of collected nuclear proteins will be done and antibodies to specific tau isoforms will be used to identify which isoforms are present. A second study will determine the transcription factor(s) that bind to the NOS1 polymorphism. This study will be conducted using electrophoretic mobility shift assays (EMSA). We hypothesize that the 1N tau isoform binds predominantly to this repeat. The findings of these studies will further understanding of tau’s role in the nucleus.

**P8 g** Elizabeth Rogers, Stephanie Sharpes (co-first authors), Dr. Kyle Seifert

**The Effects of Spacer Variation and Hydrocarbon Tail Length on the Antibacterial Activity of Novel Tris- and Bis-Cationic Amphiphiles**

Approximately 2 million people are infected annually by bacteria resistant to commonly used antibiotics, resulting in increased hospital-associated morbidity and mortality and placing a large economic burden on patients and hospitals. Amphiphiles are a well-studied class of compounds with documented antibacterial properties, and their antibacterial activity relies heavily on both electrostatic and hydrophobic interactions with the bacterial membrane. To study the relationship between head group charge and the antibacterial activity of amphiphiles of different tail lengths, three series of bis-cationic amphiphiles and three series of tetra-cationic amphiphiles were synthesized and tested against seven different bacterial strains. For tetra-cationic amphiphiles, but not bis-cationic amphiphiles, antibacterial activity relied heavily on the number of carbons separating positive charges in the head group as well as on the size of the hydrophobic region. Understanding the effects of spacer variation and how the placement of positive charges within the hydrophilic head region changes antibacterial activity is important for the development of more effective compounds that may be used as novel disinfectants to mitigate the problem of antibacterial resistance and disease transmission.

**P9** Justin Rissmiller, Shannon Palmer, Dr. Bisi Velayudhan

**Effect of accelerated feeding and estrogen treatment on Brunner’s glands and submucosal thickness in the duodenum of diary calves**

Brunner’s glands are found mainly in the submucosal layer of the duodenum which helps to neutralize the acidic chyme by their alkaline mucus secretion. The goal of this study was to determine whether different diets and exogenous estrogen can influence the amount of Brunner’s glands and submucosal thickness in the duodenum. One week old Holstein calves were randomly assigned to two dietary treatments. The restricted group (R) received a 20% protein and 20% fat milk replacer, fed at a rate of 450g per day. The enhanced group (EH) received milk replacer with high protein (28%) and high fat (25%) fed at a rate of 1.2 kg per day. A subset of these calves (n=6) were sacrificed upon weaning at 8 wk to assess the effect of diet on duodenal morphology. The rest of the calves were given either a placebo or an estrogen implant for 2 wk (n=6), creating four total treatments; R, EH, EH+Es (enhanced diet with estrogen), and R+Es (restricted diet with estrogen). Calves were euthanized and duodenal samples were harvested for histological evaluation. Brunner’s gland area and total submucosal thickness were measured using ImageJ software and data were analyzed using Mixed model procedure of SAS. Submucosal thickness was significantly increased in estrogen treated animals who received the enhanced diet (P=<0.0001). We did not find a significance difference in the Brunner’s gland area between treatment groups. Overall, our data show that the duodenal submucosal thickness of young calves can be altered in response to enhanced feeding and estrogen treatment but further research is needed to determine the significance of these results.

**P10** *poster cancellation*

**P11** Marissa Reilly, Dr. Bisi Velayudhan

**Quantification of the enteroendocrine cells in the small intestine of dairy cattle in response to accelerated feeding**

Enteroendocrine cells are solitary epithelial cells that make up less than 1% of the cell population of the gastrointestinal tract. Although they make up a small percentage, enteroendocrine cells produce hormones that are essential for normal digestion, appetite, and metabolism. The purpose of this experiment was to evaluate the effect of diet on the duodenum enteroendorcine cell population of estrogen treated dairy heifers. A total of 12 animals were randomly assigned to two treatment groups, enhanced or restricted diet. The enhanced diet sample group was fed a milk replacer at 1.13 kg/d (28% crude protein, 25% fat) and the restricted diet sample group was fed a milk replacer at 0.45 kg/d (20% crude protein, 20% fat). All animals received estrogen implants at 8 weeks. Small intestinal samples were harvested after 2 weeks of estrogen implantation and routine histological processing was conducted to prepare the tissue samples for analysis. Hematoxylin and Eosin stained duodenal samples in duplicates were observed using a Zeiss Axioscope A. 1 at a 63x objective under oil immersion. A total of 20 images per animal were taken. A total of 3000 cells were counted per sample and percentage of enteroendocrine cells was calculated. Statistical analysis of the data showed no significant difference in the percentage of enteroendocrine cells between estrogen treated restricted diet (0.53± 0.4 %) and estrogen treated enhanced diet (0.70 ± 0.4 %). Our data provide no evidence that diet has an effect on the enteroendocrine cell population of the duodenum when treated with estrogen.

**P12** Patricia Brown, Dr. Kevin Hatala, Heather Dingwall, Dr. Brian Richmond, Dr. Roshna

Wunderlich

**Foot Structure and Function in Habitually Unshod Children**

The aims of this study are to present ontogenetic data on habitually unshod feet and to compare unshod foot structure and function to those of shod children and habitually unshod adults. We ask whether low hindfoot and high hallux pressures seen in adult unshod Daasanach are present throughout ontogeny or whether they develop only in the absence of footwear. 71 unshod Daasanach children (2-13 years) in Ileret, Kenya walked across a plantar pressure mat at self-selected speeds. Using RSscan footscan software, we measured foot length and width as well as dynamic plantar pressure, force, contact area and impulse for 10 anatomical regions. Results show that habitually unshod children’s feet are unlike their adult counterpart, with average peak pressures undistinguished in the hallux and higher in the heel, a distribution similar to shod children. In contrast, unshod children are similar to unshod adults in their widened distribution of pressure across the forefoot. The results of this experiment show that foot shape and function may be initially different in unshod children than adults and that the distinctive shape and function of the foot in unshod and shod individuals may develop in the context of footwear and potentially other factors such as substrate.

**P13** Danielle Orlandi, Mark McGowan, Dr. Anthony Tongen, Dr. Roshna Wunderlich

**The Effects of Cage Size in Captivity on Physical Activity Levels in *Propithecus coquereli***

The extent to which captive environments allow primates to have physically similar experiences to wild primates is important to primate health, captive primate husbandry, and to experimental research on captive primates. The purpose of this experiment was to assess similarity of locomotor activity and energy expenditure in lemurs in caged enclosures and natural habitat enclosures (NHE’s) at the Duke Lemur Center (DLC) to those in the wild. Accelerometry can be used not only to quantify activity but also to identify specific locomotor patterns and as a proxy for energy expenditure during animal movement. Using a custom-designed datalogger attached to the backs of two *Propithecus coquereli* (Pc), we collected three-dimensional acceleration for 4 hours on each of 2 days in 2 settings: 1) Indoor-outdoor cages at the DLC (multiple indoor cages 10’H x 7.5’W x 7’L per cage and multiple outdoor cages, 10’H x 7.5-11.5’W x 14’L), and 2) NHE (1.5-14 acres) in the Duke forest. We used continuous focal animal sampling to ground-truth the data. We quantified number of leaps and overall dynamic body acceleration (ODBA) for each period of measurement and compared these data to data of wild *Propithecus verreauxi* at Beza Mahafaly Special Reserve in southwest Madagascar. Our preliminary results illustrate considerable variability in leap count and ODBA depending on time of day, age of subject and activity. Our preliminary results cannot reject the hypothesis that *Propithecus* in captive environments exhibit similar locomotor patterns and similar ODBA to *Propithecus* in the wild. The preliminary results of this study suggest that lemurs in the captive enclosures studied here display locomotor behaviors and energy expenditure commensurate to those of lemurs in the wild. Nevertheless, there is considerable variability across animals, time of day, and activity that needs to be addressed methodically and with additional data collection. We also demonstrate the effectiveness of accelerometry as a tool for analyzing locomotor behavior and energy expenditure in captive and wild primates.

**P14** Matthew Morrissey, Guy Stewart, Dr. Heather Griscom

**The effects of varying environmental conditions on American chestnut hybrids in an Appalachian Cove Forest in West Virginia**

The American chestnut, *Castanea dentata*, was once a dominant canopy species in the eastern United States. Due to a fungal blight, *C. dentata* is now a rare understory species with low reproductive success. Given its decline in population, The American Chestnut Foundation (TACF) produced potentially blight-resistant hybrids (“Restoration Chestnuts 1.0”), with the hope of reintroduction. To test their success, an experimental study was designed in an Appalachian cove system of West Virginia. Four small (40% light) and four large gaps (60% light) were created by cutting canopy trees and clearing all vegetation. In March 2014, hybrid seeds were planted and inevitably preyed on. One year later, 50 hybrid chestnut seedlings were planted within each of the same plots. To test for competition and rodent girdling, 50% of saplings within plots were randomly assigned a tree shelter and 50% of the planting rows were outfitted with landscape fabric. After two growing seasons, saplings were not significantly different in height or diameter between gap sizes. However, the difference in light did yield significantly higher survival numbers in smaller gaps, with small gaps having an 80% survival rate, compared to 50% in large gaps. Landscape fabric and shelters produced significantly taller saplings, with p values of .002 and <.001 respectively. The fabric also produced significantly greater diameter values, with a p value of <.001. The significant difference in survival can most likely be attributed to the lack of *Rubus* canopies in the small gaps, which shaded and physically crushed the saplings. The shelters and fabric appear to be minimizing this crushing and preventing herbaceous competition. Continued monitoring is necessary to determine how successful the fabric and shelters will be as the saplings continue to get larger.

**P15** Molly Campillo, Dr. Joseph Harsh

**Expertise in "seeing" in visual data: An eye tracking and think aloud using graphical representations**

In science and our everyday lives, graphs are the most common data visualization used to communicate information. However, literature has shown that students of all ages struggle with making sense of graphs. To better understand how participants “see” graph data, this ongoing study used eye tracking (ET) and cognitive interviewing (CI) to compare the processes by which individuals of varying scientific expertise read and interpret graphs. Participant eye movements, focused on the parameters of focal attention and visual search, were recorded as they completed a series of graph-based tasks. Following the ET session, CI was conducted to gain insight to participants’ general graphing experience and decision-making processes after reviewing video playback of how they directed their attention during task-completion. Data were collected from 36 participants of varying educational backgrounds, including nonscience majors (n=15), early science majors (n=8), advanced science majors (n=4), science graduate students (n=5), and science faculty (n=6). Early findings highlight a level of variation in how individuals of varying STEM backgrounds direct their attention when completing graph-based tasks. Experts were more likely to use a directed search pattern focused on relevant graph information (e.g., variables), whereas, novices lacked alignment in their perceived and actual search patterns focusing on cues (e.g., answers) in completing the graph-based tasks. Given that research on the transition from novice to expert is important in designing curricula, we feel this study has implications for the advancement of strategies to aid in the teaching of data analysis skills.

**P16** Bryan Scalf, Jenny Russel, Dr. Tracy Deem, Dr. Chris Lantz

**Assessment of Faithful Interleukin-3 Production by Transgenic Fluorescent**  **Reporter Mice**

Interleukin-3 (IL-3) is an immunoregulatory cytokine secreted by CD4 T cells and other immune cells following their activation. Although our laboratory has recently shown that IL-3 influences immunity to blood-stage malaria infection, we do not yet know the identity of IL-3-producing cells or the kinetics of IL-3 production during the course of infection. To answer these questions, we paid a core laboratory at the University of North Carolina to generate novel transgenic mice using CRISPR/Cas technology that contain an endogenous fluorescent reporter gene (ZsGreen) for IL-3 expressing cells. In these reporter mice, transcription of the IL-3 gene results in the production of separate IL-3 and fluorescent ZsGreen protein products. As a first step to confirm that ZsGreen expression is accurately marking cells with the capacity to secrete IL-3, spleen cells were isolated from heterozygous reporter and wild-type mice and cultured *in vitro* under conditions that promote CD4 TH2 cell development (IL-2 and IL-4). At day 4 of culture, cells were activated for 6 hours using plate-bound anti-CD3 and soluble anti-CD28 antibodies, stained with fluorescent anti-CD4 antibodies and examined by flow cytometry. We found that significant numbers of CD4 T cells expressed the fluorescent ZsGreen protein whereas control cells from wild-type mice did not. These results indicate that IL-3 reporter mice represent a valuable tool for examining IL-3 production in IL-3-dependent disease processes such as malaria.

**P17** Andrew Ayad, Rohan Bamania, Elizabeth Dunlap, Mason Earp, Robert Hacker, Dr. Michael Renfroe

**The effect of stressors on beta-carotene production in *Dunaliella bardawil***

*Dunaliella* *bardawil* is a green algae that produces beta-carotene which is used commercially as an antioxidant, a precursor to Vitamin A, and a yellow pigment utilized in the cosmetic and food industries.  Since the production of beta-carotene from algae is more environmentally friendly and sustainable than traditional techniques of producing synthetic dyes, it is important to optimize the production of beta-carotene.  *D.* *bardawil* cells have increased production of beta-carotene when they are grown under stressful conditions.  *D. bardawil* were grown on a modified Johnson’s medium. Compared to the control, the two stress treatments that were designated for this experiment were high NaCl and high pH.  The effect of the two stress treatments were compared to the control for their production of beta-carotene.

**P18** Wendy Chen, Marlene Flores, Matthew Lowry, Hannah Smith, Morgan Smith, and Dr. Michael Renfroe

**Inducing the production of astaxanthin in *Haematococcus pluvalis***

Astaxanthin is a commercially important pigment used in the cosmetic and food industries with a market value in excess of $447 million.  This chemical is produced naturally by the alga *Haematococcus pluvialis*.  It is a freshwater, unicellular green alga belonging to the Order Chlamydomonadales in the Phylum Chlorophyta. This alga commonly grows in ponds and streams under low light conditions. To optimize vegetative growth we cultured H. pluvialis in two different media: Bold’s Basal Medium and Optimal Haematococcus Medium.  For astaxanthin production, a two-stage culture system was employed: a vegetative growth phase followed by a stress phase to induce pigment production.  Results will be beneficial to industrial producers of astaxanthin.

**P19** Andrew Ayad, Rohan Bamania, Wendy Chen, Elizabeth Dunlap, Mason Earp, Marlene Flores, Robert Hacker, Matthew Lowry, Hannah Smith, Morgan Smith, and Dr. Michael Renfroe

**Effect of synthetic seed storage conditions on emergence of *Saintpaulia rupicola***

*Saintpaulia rupicola* is a critically endangered species endemic to. In an effort to mass propagate *S. rupicola* for conservation, we investigated synthetic seed storage technology. During storage,synseeds may lose moisture decreasing their viability over time. We hypothesized that alternative storage conditions for synseeds would promote diffusion of oxygen but also keep the seed hydrated longer. The three conditions investigated were storage in water, storage in a mixture of Hoagland’s medium and glucose, and storage in air. These treatments were compared to a control treatment which was planted immediately without storage. Synseeds were prepared by encapsulating shoot primoridia in alginate beads. Synseeds were either planted immediately or were stored in air, submerged in water, or half strength Hoaglands + 1.5% glucose contained in 1.5 mL cryovials . Cryovials were stored for 0, 2, 4, 6, or 8 days. Ten seeds per storage condition per time period were planted on a Murashige and Skoog basal medium and data were collected on emergence of the synseeds 14 day after the last planting. We found that synthetic seeds stored in water are viable for growth for up to eight days while seeds stored in a Hoagland’s and glucose mixture or air had a loss of viability. Improved storage in water may be the result of water preventing desiccation of synseeds. These findings can be applied to conservation efforts of *Saintpaulia rupicola.*

**P20** Anjali Batra, Phoebe Cook, Dr. Idelle Cooper

**Color variation within the Hawaiian damselfly, *Megalagrion calliphya***

This study measures color within the Beautiful Hawaiian damselfly, *Megalagrion calliphya*, and compares variation in color in individuals over time and in populations over elevation. We conducted a mark-recapture study and scanned live damselflies in the field to quantify thoracic color using Photoshop. We identified repeatable areas of the thorax using landmarks and averaged the color values in that area to calculate hue, saturation, and brightness for each individual each time it was captured. There are two female morphs in this species, one of which is more similar to the male color (red) and one of which is green. Both red males and red females become redder over time, and populations at high elevation are redder as well. Green females, however, do not show significant color change over time or elevation. The similar color variation between red female morphs and red males suggests similar biological processes in response to their environment.

**P21** Taylor Derby, Monique Waldman, Dr. Marta Bechtel

**Characterizing Decellularization Methods in the Rabbit Cornea Scaffold**

The ability of the cornea to heal and maintain transparency has paramount importance in preserving eyesight. Corneal blindness due to trauma or disease affects over 10 million individuals worldwide, and corneal transplants are currently the only treatment. Tissue engineering aims to resolve this crisis through development of a bioengineered cornea. One current strategy being investigated uses decellularized xenograft scaffold tissue, in which cells and proteins are stripped from cornea tissue scaffold of one species and then human cornea cells are embedded into the xenograft tissue. However, problems with xenograft tissue rejection have hampered progress with this approach. An alternative strategy employs an allogeneic graft, in which cells are stripped from a human donor cornea and the remaining scaffold is embedded with donor cells from the patient receiving the transplant. This allograft model system eliminates problems associated with xenograft transplants, and employs a naturally-derived tissue scaffold. The rabbit cornea is a model commonly employed for cornea tissue research. This study aims to characterize and compare two methods for rabbit cornea decellularization, NaCl, or SDS, and their impact on subsequent recellularization. To evaluate the relative success of each method, overall tissue appearance, including the maintenance of transparency, was recorded, and histological analysis of the cornea tissue obtained from each decellularization method was conducted. Recellularization studies are ongoing.

**P22** \*Toma Matveeva, Jared Martin, Dr. Kimberly Slekar

**Investigating the role of ß-NAC on stress response in *Saccharomyces cerevisiae***

Molecular chaperones are proteins that interact with other proteins to ensure their proper folding and cellular localization. The nascent polypeptide-associated complex (NAC) is a protein complex composed of an alpha and beta subunit that is thought to play a role as a protein chaperone, acting on newly emerging proteins at the ribosome. Research using genetic model organisms, including *Saccharomyces cerevisiase* (baker’s yeast), has also indicated a potential role for the NAC in stress response. Many metabolic and cellular pathways are highly conserved and therefore we can learn about basic cellular functions using the yeast model that may be informative for understanding human cells. It is known that proper protein folding and response to stress are important for maintaining a healthy cell, and that defects in these processes have been linked to some human degenerative diseases such as Alzheimer’s, Huntington’s and Parkinson’s disease. NAC mutations cause embryonic lethality in mice, flies and nematodes. Our work in yeast will allow us to shed light on the role of the NAC in a single-celled eukaryote. We are conducting experiments with yeast strains that have a deletion of the yeast gene encoding ß-NAC, to determine whether the yeast NAC mutant has an altered response to stress. We are testing for potential growth defects of the mutant strain in response to heat stress and oxidative stress, and also whether the mutant strain has an altered lifespan. Our results will be used to determine whether detrimental effects occur in a yeast cell that does not contain a functional ß-NAC, which will inform future directions of the research in our laboratory.

**P23** Amanda Crandall, Kristianna Bowles, Rhiannon English, Cole Roberts, Nathan Robinson, Dariia Yerahova, Dr. Bruce Wiggins

**A Comparison of Riparian Characteristics and Resulting Water Quality in Restored Agricultural Systems**

Agronomic land use and urbanization are the leading causes of water quality decline within streams of the Shenandoah Valley. Implementation of riparian buffer zones is a common, beneficial approach to initiate restoration of negatively affected waterways; in the state of Virginia, the Conservation Reserve Enhancement Program (CREP) assists landowners in repairing natural habitat through the provision of cattle fencing and reintroduction of hardwood trees, native warm season grasses, and shrubs. We analyzed seven CREP restored sites of varying time since restoration (5-15 years) to determine the effects of time, land use, and riparian zone characteristics on water quality. The Virginia Stream Condition Index (VA-SCI), Hilsenhoff Biotic Index (HBI), and Shannon Weiner Diversity Index (H) were used to quantify water quality through the use of site-specific benthic macroinvertebrate identification. The percent forest, agricultural land, and impervious surfaces in watershed and 100 meter buffer areas for each site was calculated through GIS analysis. Riparian characteristics were determined through in-field assessment of overhanging vegetation, amount woody debris, number of riffles, average number of woody specimens (per m2), and average diameter-at-breast height (DBH). Single variable regressions showed no correlation or statistical significance between the resulting macroinvertebrate index scores and tested variables with the exception of woody debris presence. The amount woody debris was shown to significantly predict VA-SCI and H scores, with R2 values of 0.669 and 0.513 respectively; a lower amount woody debris predicted higher water quality. Through stepwise, multiple variable linear regression tests, we found that all combinations of time, riparian buffer characteristic groupings, and land use were significant predictors of macroinvertebrate index scores, all with adjusted R2 values above 0.750. Though these results were consistent with our predictions, it should be noted that the sample size of this study was small; an increased sample size and longer monitoring period may provide more substantial results in the future.

**P24** Kristianna Bowles, Amanda Crandall, Rhiannon English, Megan Moore, Cole Roberts, Nathan Robinson, Dariia Yeharova, Dr. Bruce Wiggins

**Using LiDAR to generate digital elevation models for watershed delineation and canopy height measurements**

The objective of this study was to determine if vegetation height had an influence on stream health as measured by various macroinvertebrate indices. The indices used are the Hilsenhoff Biotic Index, Shannon Diversity Index, VA Stream Condition Index, and total abundance. Vegetation height was compared to various vegetation metrics such as average diameter at breast height, number of riffles, number of woody specimens, and average number of trees, to determine if any relation exists. Light Detection and Ranging (Lidar) was utilized to determine the relative canopy height of watersheds and stream buffers. Lidar derived digital terrain models (DTM) were generated using ArcGIS’s model builder application to automate the process. Lidar derived digital surface models (DSM) were also generated in ArcGIS using a variety of Lidar data exchange and raster based tools. The DTMs modeled the elevation of the bare earth an where subtracted from the DSMs which depicted the elevation of vegetation. This subtraction yielded the relative height of vegetation in a given area. Nine sample sites where macroinvertebrates were collected had their watersheds and stream buffers measured using these Lidar methods. There was no significant relation between the vegetation height of these watersheds and buffers with any stream indices or vegetation metrics using linear regression. Increasing the sample size of the watershed and stream buffers may yield more informative results.

**P25** Kristianna Bowles, Amanda Crandall, Cole Roberts, Rhiannon English, Dariia Yeharova, Nathan Robinson, Dr. Bruce Wiggins

**Effects of Riparian Restoration and Land Use on Water Quality in the Shenandoah Valley, Virginia**

The Shenandoah Valley encompasses some of the highest agricultural producing regions in Virginia, many of which are large contributors to non-point source pollution. To help reduce this, the Conservation Reserve Enhancement Program (CREP) aids participating landowners in fencing out cattle and planting saplings to restore riparian buffers. The study was undertaken to determine how spatial and temporal factors impact the effectiveness of riparian buffer restoration by measuring overall water quality through macroinvertebrate biotic indices (HBI, VA-SCI, and Shannon Diversity). GIS analysis was also employed to calculate land use characteristics (land use, canopy cover, slope, relief, road density and impervious surface) for watersheds and for 100-meter buffers areas at each sampling site. Single variable, multiple variable, and linear regressions were performed separately within the watershed and buffer zones. Multiple linear regressions revealed that HBI had a negative trend towards more sensitive organisms and that VA-SCI and Shannon Diversity had positive trends towards more diversity and higher water quality when compared to restoration time. Despite not being statistically significant, these regression trends are indicative of improved water quality of the streams in CREP.

**P26** Dominic Sales, Michael Pamonag, Dr. Kristopher Kubow

**Influence of fiber orientation and fiber movement on cell polarization and migration**

Our current study aimed to further our understanding of cell migration within the dynamic relationship between cells and the extracellular matrix (ECM). Cell migration plays a driving role in many important processes, such as cancer cell invasion and regeneration of damaged tissues. The ECM is a network of fibrillar proteins, proteoglycans, and other molecules which connect the spaces in between the cells of tissues. As observed in tumor cell invasion, cells can follow aligned fibers, essentially using them as tracks to direct cell migration. Migrating cells have also shown the ability to manually align the fibers within the ECM, suggesting a positive feedback system in which cells both follow and align fibers. A previous study characterized a novel, photoactive methacrylated type I collagen which was found to have a quantifiable increase in storage modulus upon UV irradiation; however, cells’ ability to manipulate the fibers of these gels is unknown. Collagen hydrogels of varying levels of crosslinking were formed to observe patterns in cell invasion. In order to observe matrix invasion from a set point, we polymerized a small collagen gel (100uL), containing HT-1080 cells, along with an encasing, larger methacrylated collagen gel (300uL). Through gel contraction assays, we determined that cells in the crosslinked gels were unable to contract the matrix to the same degree with respect to the non-crosslinked gels. Currently, ongoing experiments are being conducted to observe migration patterns into gels with varying levels of crosslinking. Observations from these experiments could provide insights of how cell durotaxis can be altered with these crosslinkable hydrogels and how the positive feedback system between the cells and the surrounding fibers can be controlled.

**P27** Reef Buckhalter, Jessica Cornell, Peyton Coady, Dr. Marta Bechtel, Dr. Kristopher Kubow

**Development of a Cell Culture System to Study the Effects of Physical Confinement and Spatial Arrangement on the Maintenance of Chondrocyte Phenotype**

Articular cartilage, a frictionless connective tissue found in articulating joints, is responsible for reducing the mechanical stresses of movement on the body. This stress can often cause damage to the cartilage over time and lead to chronic degradation. Such damage can cause chondrocytes to lose their normal, lacunae-enclosed phenotype and become more fibroblast-like, exhibiting behaviors similar to wound healing situations. Prevention of this chronic condition requires stimulating chondrocytes to repair the cartilage tissue while maintaining their phenotype. Unfortunately, little is known about how chondrocytes produce cartilage matrix and about how chondrocyte phenotype is maintained, in part because chondrocytes do not maintain their phenotype in standard, two-dimensional cell culture dishes and are difficult to study in 3D matrices.   
Our goal is to develop a system that enables us to study chondrocyte phenotype in a 3D environment that is easy to image and has controllable parameters. Ultimately we want to study how altering chondrocyte confinement (simulating lacunae) and the molecular composition of their environment affects their phenotype. We therefore have developed tools to stamp “microwells” (wells big enough for one or several cells) into different gel materials. Using these wells, we can modify cell confinement, shape, and the stiffness and molecular composition of the environment. The stamps for the wells were produced using photolithography and molded in silicone (PDMS). Large wells formed correctly; however the wells sized for one to two cells were overall too shallow to be used effectively. Additional optimization is currently underway to increase the depth of the smallest microwells. The silicone stamps with larger wells were successfully used to produce wells in agarose and in PDMS. Cells cultured in agarose wells maintained a rounded phenotype similar to being seeded into 3D alginate gels as opposed to a fibroblast-like morphology in 3D collagen. In addition to preparing correctly formed single-cell wells, our future projects include producing wells in more physiologically accurate materials such as collagen I (simulating wound healing) and in adding surface functionality in the form of specific cell receptors to otherwise inert PDMS gels. All of these models will be used to study how the microenvironment affects chondrocyte phenotype.

**P28** Kristin Sammons, Lexi Deak, Dr. Corey Cleland

**Selective Stimulation of A-delta Nociceptors in Rat Hind Limb and the Resulting Nociceptive Withdrawal Response**

Rats rapidly withdraw their hind limb in response to heat or other noxious stimulation, which is known as the Nociceptive Withdrawal Response (NWR). Two types of nociceptors may mediate the NWR: C-fibers and Aδ nociceptors. Among the differences between these two types of nociceptors, C-fibers have large receptive fields while Aδ nociceptors have much smaller receptive fields. Previous studies have shown that the direction of the NWR does not depend on stimulus location. However, these experiments used a method of heating that may have predominantly stimulated C-fibers. If C-fibers were stimulated, we might expect no dependence on stimulus location due to their larger receptive field compared to Aδ nociceptors. Therefore, it remains possible that Aδ nociceptors could mediate a response that is dependent upon stimulus location. The specific aim of our ongoing experiments is use preferential stimulation of Aδ nociceptors using high intensity, short duration (100ms) pulses of heat to determine if the NWR depends in stimulus location. The length of the pulse was determined based on previous research showing that Aδ nociceptors are selectively activated with short, high intensity pulses of heat. Because Aδ fibers have small receptive fields, we hypothesize that the selective stimulus will result in a response that is dependent on stimulus location. Five small (1 mm) spots (three aligned rostral-caudal, three aligned lateral-medial) were blackened on the plantar surface of the left hind paw. These spots were stimulated in a randomized sequence and the initial and final positions of the paw were recorded with a camcorder (60 fps @ 1080p) placed underneath the rat. When stimulated, the rat picks up its paw and rapidly places it back down on the glass. The difference between the initial and final positions represents the NWR movement response vector. Unexpectedly, stimulus location still did not have an effect on the direction of the NWR. Rather, preliminary results (n=6) suggest that the direction of the response is determined only by the initial position of the paw, as observed in previous experiments where C-fibers were presumably stimulated. These results further substantiate our findings that the NWR is organized around initial posture rather than the details of the noxious stimulus.

**P29** Seerat Mavi, Mary Pegelow, Diana Grigoryan, Corey Dr. Cleland

**The effect of noxious tail simulation on the nociceptive withdrawal response of unrestrained rats**

When animals are exposed to noxious stimuli, they can have varying responses based on the intensity of the stimuli or environmental conditions. Previous studies have observed that when the tail of a restrained rat is stimulated using a heat stimulus, the tail moves in the opposite direction from the stimulation site. However, there is no research that evaluates the body movements of rats that are unrestrained. Earlier studies in unrestrained lizards have shown that as the stimulus location moves closer to the base of the tail, the animal tends to walk forward instead of moving the tail. This study aims to determine whether an unrestrained rat will move its body when its tail is stimulated using a noxious heat stimulus. Rats were anesthetized to mark the tail, feet, and body. The animal was then centered on a glass table, where a heat stimulus was delivered to one of the five stimulus locations on the tail. Movement was recorded through a video camera placed below the center of the glass table. In addition to tail withdrawal, we observed that concomitant body movement always occurred. The direction of body movement consisted of both forward translation and rotation away from the stimulus. The timing of body movement, based on initial foot movement, lagged tail movement by only 167 ms (median), however, the timing was not dependent on the stimulus location (p=0.37). The initial foot movement did depend on stimulus location; when the base of the tail was stimulated, the back left foot moved first, but when the tip was stimulated the front right paw moved first. These results indicate that while the tail movement occurs similarly to previous studies, body movement also plays an integral part in their nociceptive withdrawal responses.

**P30** Don Afful, Aubrey Siebels, Ariel Childs, Alex Zeher, Corey Cleland

**Sensory Mechanisms Underlying the Escape Response to Looming Stimuli in Crickets**

Animals respond to aversive stimuli with escape or withdrawal responses. In crickets, wind, which might normally be produced by an approaching predator, has been shown to evoke an escape response in which the cricket turns 180 degrees from the wind and then runs or jumps away. We have shown that crickets (Acheta domesticus) largely utilize the same turning strategy for looming stimuli, which provides both wind and visual sensory cues. However, there is lack of literature on the sensory modalities that underlie the escape. Our specific aim is to identify and compare the role of visual, cercal, filiform, and antennal sensory cues in the escape of the cricket from looming stimuli. Crickets (n=90) were stimulated with a 2.5” ball (1 m/s) projected at 45 degrees using an air cylinder and stopped 20 mm from the initial position of the cricket. Criclets were stimulated at 8 angles, in 45-degree increments around the body in random order. Above the platform was positioned a high-speed video camera (650 fps) and a LED ring light. Prior to stimulation, a primary sense organ (eye, cerci or antenna) or a source of sensory information was ablated or removed to test necessity, or isolated to test sufficiency for the escape response. Crickets were blinded with lacquer nitrocellulose, or deprived of cercal or antenna information via lesions applied at the base of the appendage. A glass panel was placed between the animal and the stimulus to block wind cues to intact crickets. Crickets’ compound eyes are only receptive to light below red wavelengths so 660nm red light was used to eliminate visual cues in intact crickets. Finally, a white ball against a white background was used to eliminate visual but retain wind cues. Results showed that both eyes and cerci, but neither filiform hairs not antennae, contribute to the escape response. The contributions of both eyes and cerci to the escape depend largely on the direction of incoming stimuli. The eyes mediate escape responses to anterior looming stimuli while the cerci mediates escape responses to posterior stimuli. In addition, our results showed that escape responses mediated by the cerci had a shorter latency but similar magnitude as compared to escape responses mediated by the eye. Taken togerher, our results suggest that crickets use both vision and wind to programs escape responses, and that the resulting movement differ in at least latency.

**P31** Ashley Ballengee, Paulina Bauer (co-first authors), Dr. Corey Cleland

**Skin temperatures in the nociceptive withdrawal response of the rat tail**

Rats rapidly withdraw their tail in response to noxious heat stimulation, which is known as the nociceptive withdrawal response (NWR). Previous studies from our laboratory have shown that stimuli placed 2 cm apart along the tail result in different tail movements. However, it is possible that heat applied to one spot spreads to adjacent spots, thus confounding the results. The specific aim of our research is to measure the distribution of temperature along the tail in response to localized heat stimuli delivered by an infrared laser. Preliminary results demonstrate that spread is minimal. At one cm away from the stimulated site, temperature increase is only about 10% of the increase 1 mm away from the stimulated spot. Thus, heat spread does not appear to be a problem for studying the heat-evoked NWR of the tail.

**P32** Courtney Neumeyer, Rebecca Livermore, Gregory Steffensen, Dr. David McLeod

**A Review of Overwintering Strategies in Three Species of North American Amphibians**

In temperate regions many organisms, especially ectotherms like amphibians and reptiles, hibernate during the winter months. Though it is well known that there are different strategies used by amphibians to survive freezing temperatures, many species-specific questions remain unanswered. The aim of this paper is to review the current research on amphibian overwintering strategies and apply it to three Virginia species: *Lithobates sylvaticus* (wood frog), *Lithobates palustris* (pickerel frog), and *Notophthalmus viridescens* (Eastern newt). Three different methods of overwintering in Virginia include freeze tolerance (*L. sylvaticus*), cavernous hibernaculum overwintering (*N. viridescens*), and underwater overwintering (*L. palustris and N. viridescens*). These overwintering methods require specific behaviors and physiological processes to prepare them for survival. Each is unique in the cost it has on amphibians, but they are overall beneficial to the survival of each species. We summarize the previous research conducted on freeze tolerance, cavernous hibernaculum overwintering, and underwater overwintering in temperate amphibians. Furthermore, we address the gap in our knowledge of these types of overwintering. The purpose of this review is to provide a basis from which to expand the understanding of overwintering in specific species of amphibians.

**P33** Jenna Salter, Dr. Bryan Cage, Dr. David McLeod

**Revolutionary Methods of Telemetry for the Ecological Study of Eastern Box Turtles**

Radio telemetry is a commonly used tool in the fields of ecology and wildlife biology. While effective it remains limiting in its expense and that its delivery of data only when the receiver is in proximity to the tagged animal. The goal of this project is to use the relatively inexpensive and flexible Arduino technology to collect data remotely and continuously. The successful use of Arduino microprocessors in remote data collection could revolutionize the way telemetric studies are conducted as data could be relayed to one’s computer or cell phone without having to track the animal or physically go to the site. This study investigates the potential for using this type of technology in remote data collection. A prototype unit for relaying geospatial and environmental information associated with Eastern Box Turtles (*Terrapene carolinensis*) is under development as is a graphical web-based interface to receive and disseminate these data. Results from the development and initial field-testing of the prototype are presented here.

**P34** Shelby Snowden, Dr. Kerry Cresawn

**An Exploratory Study to Understand Elementary School Students’ Conceptions of Food Chains**

Research has shown that elementary school is a critical time to pique children’s interest in

science. However, many enrichment activities known to pique this interest in young children

are not available to students of low socioeconomic status, English Language Learners, racial

minorities, and students with disabilities. This has encouraged many higher education

institutions to develop STEM outreach programs. Because of the cognitive gap between STEM

professionals and young children and the logistics of implementing student-centered activities

in heterogeneous classrooms, programs usually consist of activities that impress students with

“sophisticated” science but are beyond the cognitive levels of most students and do not create

lasting interest in science or facilitate learning. These activities can also be unattractive for

teachers, as they do not have the resources to carry out expensive, complex lessons and are

already rushed to cover material required by the Virginia Standards of Learning. Development

of a sustainable K-5 outreach model that piques interest while aligning with foundational

standards requires an understanding of the extent to which a short-duration enrichment

lesson can enhance conceptual understanding of topics. To assess Madison Discovery, a novel

K-5 outreach model to make science enrichment accessible to all children, we collected data

representing children’s conceptions of food chains through visual representation. This study

gathered baseline data that can be used to further develop the lesson, to serve as a model for

assessing other Madison Discovery lessons, ultimately leading to an outreach model that

serves K-5 communities as well as higher education throughout the country.

**P35** Chelsea Romanchuk, Ashley Warrington, Brooke Thompson, Dr. Conley K. McMullen

**Updating floristic surveys of the Edith J. Carrier Arboretum and the Smith Creek restoration area**

This research project aims at providing an updated account of specimens collected throughout the Edith J. Carrier Arboretum and the Smith Creek restoration area. This updated inventory reflects any changes in the taxonomic nomenclature of species that were collected, identified, and mounted during previous years of this study. A digital database with a description of GPS location, family name, species name, authority, and collection date was created to determine existing arboretum species as well as progress at Smith Creek following the implementation of riparian buffer restoration in 2006. The database for the arboretum contains 326 specimens. There are 174 different species within 123 genera and 69 families. The Smith Creek database contains 493 specimens comprising 269 different species within 173 genera and 60 families. GPS coordinates of each specimen will be added to a map in ArcGIS to provide the location of flora within the arboretum and to monitor the extent of riparian restoration at Smith Creek.

**P36** Sam Hetrick, Lilly Nelson, Romie Powell, Alexander Schmidt, Dr. Justin Brown

**The Role of Brain Stem 5HT1A and GABA-A Receptors in the Thermoregulatory Response to Hypoxic Stress**

Sudden Infant Death Syndrome (SIDS) is a leading cause of infant mortality (1). Alterations in brainstem development of Serotonin (5HT) and GABA are linked to its cause (2). The sympathetic premoter neurons located in the Nucleus of the Raphe mediate protective cardiovascular responses to environmental stress (3). It is hypothesized that alteration in these receptors at the NRP will also impair protective thermoregulatory responses to hypoxic stress such as hypothermia. Using aseptic techniques, male and female Sprague-Dawley rats (225-325g) were instrumented with radiotelemetry probes to non-invasively measure core temperature (Tc). Using a stereotaxic device, a steel cannula was inserted into the brainstem which allowed microinjection at the NRP. After recovery (1 week), rats were housed in a thermal gradient which allowed them to select their ambient temperature (STa) and thereby facilitated behavioral thermoregulation. Once acclimated to the gradient and to handling, 30mM of either a 5HT1A agonist (8OH-DPAT or “DPAT”), antagonist (WAY100635), a GABA-A agonist (Muscimol), antagonist (Bicuculine) or ACSF (control vehicle) was then microinjected into the NRP immediately before exposure to 6% O2 for 60 min. In rats injected with ACSF, Tc decreased by 1.8° C while the Tc of those injected with DPAT and WAY decreased by 3.8° C and 2.8° C respectively. Those injected with Muscimol and Bicuculine exhibited similar hypothermic responses to control in that the Tc dropped by 2.0° C and 1.9° C respectively. There were mild decreases in STa of control group rats (4.3° C) which was exacerbated in DPAT injected rats (8° C). Importantly, the STa responses to hypoxic stress helped facilitate Tc changes suggesting coordination between behavioral and autonomic thermoregulatory mechanisms which facilitated the protective hypothermic response. Rats injected with WAY seemed to reverse this trend initially with an increase in STa (3°C) which quickly faded. Muscimol and Bicuculine seemed to have minimal effect on STa responses. These preliminary data suggests that GABA-A receptors have minimal role in the thermoregulatory response to hypoxic stress. However, activation of the inhibitory 5HT1A receptor exacerbates the hypothermic response to hypoxic stress and may facilitate this protective response. Alterations in 5HT neuronal development may cause inadequate behavioral (STa decrease) and autonomic (Tc decrease) heat loss responses to hypoxic stress and may be a significant factor in the etiology of SIDS.

**P37**  Madison Frongello, Gabriela Constancia, and Dr. Marquis T. Walker

**Characterizing the role of the TRPML cation channel in *Drosophila* innate immunity**

Transient receptor potential mucolipin 1 (TRPML1) is a cation channel that is localized to the late endosome/lysosome in all cells. TRPML1 functions to regulate lysosomal pH and ion homeostasis. Loss of function mutations in the gene that encodes TRPML1 (MCOLN1) results in a devastating childhood neurodegenerative disease known as Mucolipidosis type IV (MLIV). TRPML1 is an evolutionarily conserved protein with homologs in *Drosophila* (TRPML) and *C. elegans* (CUP-5) . In *Drosophila*, trpml knockout flies exhibit neurodegeneration and motor defects similar to MLIV patients. Surprisingly, restoring the expression of wild-type trpml+ in phagocytic blood hemocytes rescues motor deficits in flies. This restored expression of TRPML rescued hemocyte-dependent removal of apoptotic neurons and slowed the progressive neuronal cell death. Similar to hematopoietically-derived macrophage in humans, fly hemocytes have a critical role in innate immune responses. These phagocytic cells rapidly recognize, target, and engulf foreign and infectious organisms to degrade and remove them from the host. Using the fly we will test whether innate immune function is also impaired in TRPML deficient flies. Through this study we intend to improve our understanding about the cellular requirement for TRPML, and how the loss of function could affect the overall health of MLIV patients.

**P38 g** Matthew Harris, Amanda Crandall, Mark Petre, Chloe Wines, Dr. David McLeod

**Ultrasonic Vocalization in Southeast Asian Frogs**

Ultrasonic calling was previously thought to occur exclusively in mammals and select invertebrates. Recent studies, however, have identified two amphibian species that also emit calls in the ultrasonic range; the Hole-in-the-head frog (*Huia cavitympanum*)*,* and the Concave-eared torrent Torrent frog (*Odorrana tormota*). Ultrasonic detection was assumed impossible among amphibians due to the lack of a middle ear, which prevents frequency detection above 5-8 khz. Despite this anatomical limitation, both *H. cavitympanum* and *O. tormota* possess the ability to emit and detect a wide range of frequencies, including sounds in the ultrasonic range, with *H.* *cavitympanum* calling entirely in ultrasonic frequencies.  Reasons ultrasonic calls evolved in amphibians include mate attraction, establishing territories with other males, localization acuity, and to signal in noisy environments. Some drawbacks of this vocalization are that it is energetically costly and the signal’s increase in range can attract parasites and predators.

**P39** \*Esraa Aldkheil, Dr. Nathan Wright

**Characterization of the Titin ZIg9/10-Obscurin Ig58/59 Complex**

Obscurin is a cytoskeletal protein implicated in organization and signaling within myocytes. While this giant protein is multifaceted, one of its main functions is to link the contractile cytoskeleton to the surrounding membrane structures. One way it does this is through the specific interactions between obscurin domains Ig58/59 and titin ZIg9/10. Here, we continue structural studies on titin ZIg9/10-obscurin Ig58/59 regions. The high-resolution structure of the obscurin Ig58/59 region has been previously solved in our lab, however molecular mechanism of how obscurin forms a complex with the titin ZIg9/10 region has not been examined thoroughly. To better understand the atomic etiology of this binding event, here we present initial data mapping the titin – obscurin binding interface.

**P40** \*Rachel Barborek, Kendyl Combs, Adam Fischel, Althea Neighbors, Will O'Connor, Osna Samady, Ryan Samuel, Rebekah Tenney, Dr. Susan Halsell

**Identifying the molecular components of cold nociception in *Drosophila melanogaster***

Nociception refers to an organism’s perception and reaction to potentially damaging noxious stimuli. While this response is essential, humans suffer from chronic pain in which the pain signals abnormally persist months after trauma, injury or infection. This study aims to better understand the molecular mechanisms of pain by researching the potential role of eight individual *Drosophila* Innexin gap junction proteins in cold nociception. Similar to mammalian Connexins, some of these proteins are hypothesized to be involved in the electrical synapsis between neurons. The expression level of each protein is knocked down by cell specific expression of innexin RNAi constructs in the dendritic arborization sensory neurons that mediate nociception.   
Wild type third instar *Drosophila* larvae exhibit a characteristic “cringe” response when exposed to noxious cold. Knocked-down larvae are subjected to the cold behavior assay, and their behavior is videotaped and analyzed to quantify the “percent cringe” value in order to identify the number of “cringers” for statistical analysis. By comparing the proportion of cringers between the knocked-down, experimental larvae and the wild type, the involvement of the knockdown protein in the cold nociceptive signaling pathway can be inferred. Controls utilizing Oregon-R wild type larvae and larvae in which tetanus toxin is expressed specifically in da neurons will be described.   
To date, the ogre and shaking-B Innexins have been tested with no significant change in cringing for either (p>0.1, Two-Tailed Homoscedastic TTest and Bonferroni correction). Additional Innexins will be tested and a pan-neuron driver will also be used to drive RNAi expression.

**P41 C** Andie Gargiulo, Ashton Holub, Melika Rahmani-Mofrad, Dr. Ray Enke

**RNA sequencing analysis of the human retinal transcriptome**

The retina is a layered neuronal tissue lining the posterior portion of the eye that converts photons of light into visual images in the brain. Rod and cone photoreceptors are highly specialized light sensitive neurons that initiate this process of phototransduction. Though they are very similar cell types, rods and cones have distinct functionalities, synaptic connection, and are affected by different blinding disease alleles. In order to better understand the distinct molecular function of human photoreceptors, cone rich central macula and rod rich peripheral retina samples were biopsied from post mortem human donor eyes in biological triplicate. Total RNAs were extracted from retinal samples using a Qiagen AllPrep Mini Kit. RNAs were validated for quality and sent for mRNA-Seq transcriptome analysis using the Illumina NextSeq 500 sequencing platform. Raw sequence quality assessment and high precision mapping to the Human 2013 Hg38 genome assembly were achieved using FastQC and TopHat software respectively. Differentially expressed genes (DEGs) between macula and retina were determined using CuffDiff software analysis of TopHat mapping outputs between the two sample groups. DEGs were filtered for biological and statistical significance yielding 2,240 genes significantly upregulated and 2,387 genes significantly downregulated in the peripheral retina compared to the macula with a p-value of <0.05. DEGs were then crossed referenced with evolutionarily conserved DNA binding sites for the transcription factor CRX*.* Current analysis focuses on identifying critical regulatory elements upstream of DEGs in an effort to characterize cell-specific mechanisms of gene regulation in photoreceptor neurons.

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**P42** Emily Miller, Ellen Jones, Faith Hartley, Dr. Joseph Harsh

**A closer look at interactions and activities within undergraduate research**

**experiences through weekly journals.**

Undergraduate research experiences (UREs) are growing in popularity within the STEM field to augment student skills and prepare them for research-related careers. However, despite the merit of prior research that has commonly relied on post-experience survey data to recognize an assortment of cognitive and affective outcomes conferred to URE participants, the nature of students’ day-to-day research activities - that contribute to these outcomes - have been largely unexamined. Using data collected as part of a large mixed-methods project investigating how UREs benefit students in the physical science, this study analyzed weekly electronic journals (i.e. short surveys) in which participants (n=110, 49% female) provided information about their research activities, interactions with members of the scientific community, and perceptions of the weekly events and their work over a 8-10 week summer research experience. Students experienced a wide-range of interactions, including varying mentorship models and availability, participation in research meetings, and lab group dynamics. Notably, student satisfaction in the research experience was correlated with the amount of time spent with a mentor, and research group meetings were regularly identified by students as a key learning opportunity. By focusing on participants’ weekly research practices, this study begins to provide a finer-grained understanding to how these experiences benefit students - which we feel will be useful to faculty and administrators for programmatic refinement in support of participant learning.

**P43** John Nguyen, Jonathan Popham, Dr. Joseph Harsh

**The Influence of Kinesthetic Games on College Students' Mental and Physical Well- being**

Recent innovations over the past ten years have increased the commonality of kinesthetic technologies (e.g., Wii Fit, Fitbit, Pokemon Go) that promote physical activity. Despite their growing popularity, few systematic studies have been conducted to investigate the effect of kinesthetic technologies on one’s well-being. Drawing on the recent cultural phenomenon of Pokemon Go!, a game that integrates physical and digital environments in which players are required to complete tasks that involve outdoor physical activity (i.e. walking), the present study examined the potential influence of this technology on college students’ physical, mental, and social health. For comparative purposes, survey data were collected from college-aged students that did and did not play Pokemon Go (n = 83; 64% female, 54% Pokemon Go players). Respondents were asked questions about their mental and physical well-being as well as their social interactions and interest in spending time outdoors. Quantitative and qualitative data lend support for a modest effect of Pokemon Go on respondents’ physical activity and time spent outside during and after game use. Additionally, notable changes in social interactions were observed as nearly one-third of respondents reported they spent more time with their friends/family because they played Pokemon Go. Early findings from this exploratory study highlight the conferred benefits of playing Pokemon Go for college students’ health, and we propose that a broader understanding to the impacts of kinesthetic technologies may uncover new opportunities to support one’s well-being and promote healthy lifestyles.

**P44** Kathryn McGee, Dr. Patrice Ludwig

**The Effect of Artificial Substrate on Initial Eastern Oyster Growth**

Ocean acidification is posing a new challenge in the effort to restore eastern oyster (*Crassostrea virginica*) populations. Calcifying organisms such as oysters create their shells by sequestering environmental calcium, however, their calcium carbonate shells are subject to dissolving in the acidified ocean. Restoring oyster reefs is important both ecologically and economically. Oyster serve as keystone species and ecosystem engineers building substrate for a diverse array of organisms. They are also economically important as a food source. The purpose of the restoration experiment described here was test the effect of artificial substrates on initial oyster growth. The experiment had 3 substrate treatments (n=4). We created two types of artificial substrates from molds of single oyster shells made with normal concrete (made with silica sand) and “special” concrete (made with limestone sand) and compared them to a natural oyster shell control. Limestone sand increases the calcium carbonate in the artificial substrate and could act as a localized buffer against acidification. We assume that the growth of the oyster on a particular substrate is a proxy for preference and eventual fitness of the oyster. After growing for 16 weeks from July 2016 to October 2016, all recruited oyster spat were photographed with a ruler for size reference. Photographs were taken and annotated using the app SnapChat and individual oysters were measured in cm from the extremes in height (hinge to beak) using the software ImageJ. The effect of substrate on growth will be measured statistically using an ANOVA; the results are pending.

**P45** Sarah McGrath, Charles Holmes, Connor Keelan, Dr. Morgan Steffen

**Effect of nutrient amendments on lake microbial communities**

Microbial communities drive biogeochemical cycling and form the basis of food webs in aquatic ecosystems. These communities are subject to wide fluctuations in environmental conditions, particularly in freshwater systems, which may experience major fluctuations in temperature and availability of key nutrients such as nitrogen and phosphorus. This study investigated the impact of nitrogen amendment on the bacterial community of Lake Shenandoah, a shallow artificial lake in Rockingham County, VA. This system receives unnatural influxes of anthropogenically derived nutrients due to surrounding urban development. In order to determine how nutrient input drives bacterial community composition in this freshwater system, microcosms of water from Lake Shenandoah were incubated for 48 hours at 26 °C with both 10 μM and 50 μM concentrations of urea, nitrate, and ammonium. After extracting DNA from nutrient-amended and control samples, the 16S rRNA gene was amplified via PCR before Illumina sequencing and targeted metagenomic analysis using QIIME. Preliminary results indicate levels of Cyanobacteria comparable to the control after a 10 μM NH4+ amendment, while there was an approximately 80% decrease in Cyanobacteria after amendment with 50 μM NH4+. OTUs assigned to Microcystaceae, a family which includes the toxic cyanobacterium *Microcystis aeruginosa*, were present in all control and amendment samples except the 10 μM NO3- amendment. OTUs assigned to Nostocaceae, a family which contains nitrogen-fixing *Nostoc* spp., were nearly ubiquitous in all but the 10 μM NO3- amendment.

**P46** Megan Coceano, Guadalupe Reynoso, Emily Walsh, Dr. Morgan Steffen

**Characterization of freshwater microbes relevant to cyanobacterial harmful algal blooms**

Cyanobacterial harmful algal blooms are ubiquitous in freshwater systems in the United States and around the world. From decades of research, we know that factors such as nutrient loading and temperature impact the success of organisms capable of forming toxic blooms in lakes. Only recently, however, has it been recognized that cyanobacteria may have a partnership with heterotrophic bacteria in the phycosphere, a region surrounding cyanobacterial cells analogous to the rhizosphere. From research conducted on eukaryotic marine algae, it appears that nutrient exchange within the phycosphere may promote the success of both partners. No research has yet been conducted on any freshwater phycosphere systems. To help understand the dynamics of microbial populations within freshwater blooms, we have established a consortium of phycosphere partners. Bacteria from cultures of *Microcystis aeruginosa* NIES 843 and the diazotrophic cyanobacterium, *Cylindrospermopsis*, were isolated and are currently being identified via 16S rRNA gene sequencing. Gram staining was conducted on the samples of lake water to determine the Gram status of the organisms and purity of our cultures. Additionally, we are working to establish the first culture of a *Cylindrospermopsis* isolate from the state of Virginia via enrichment culture techniques for diazotrophic cyanobacteria.

**P47** **C** Sophie Jurgensen, Charles Holmes, Dr. James Herrick

**Whole-genome epidemiology of environmental *Salmonella*: optimization and integration into an undergraduate course**

Approximately 9.4 million cases of foodborne illness occur annually in the United States, with *Salmonella* infections comprising about 1 million of these cases. Furthermore, environmental reservoirs of *Salmonella* adjacent to agricultural production may contribute greatly to the dissemination of these potential human pathogens, though they are not well characterized. Thus, the surveillance of environmental *Salmonella* becomes a critical step in monitoring the incidence of potentially pathogenic *Salmonella*. After obtaining sediment and poultry litter samples from streams and poultry houses in the Shenandoah Valley, modified FDA BAM methods were used to isolate and confirm the identity of environmental Salmonella isolates, with the notable addition of a PCR for the *Salmonella*-specific invA gene and Rep-PCR fingerprinting of isolates using BOX primers. To date, 23 *Salmonella* have been isolated from stream sediment (18) and poultry litter (5). These isolates will have their whole genomes sequenced by the FDA, after which bioinformatic analyses will be performed, including serotyping and evaluating the presence of medically relevant genetic elements, such as antimicrobial resistance genes and mobile genetic elements. These procedures will be incorporated into portions of Bacterial Discovery, a new laboratory class which follows the CURE model.

**P48** \* Sean Gay, Roxana Behrooz, and Dr. Mark Gabriele

**Double-labeling neuroanatomical staining protocols for differentiating auditory midbrain neuronal subpopulations**

Helpful in defining micro-organizational features of complex brain structures is the ability to stain specific neuronal subpopulations. To effectively identify functional compartments, it is often necessary to label multiple neuronal markers in a single tissue section. The present study aimed to design and test a double-labeling protocol in mouse pairing histo- and immunocytochemical approaches in individual slices. Nicotinamide adenine dinucleotide phosphate-diaphorase (NADPH-d), a metabolical marker, and calretinin (CR), a calcium-binding protein, were simultaneously labeled and observed in the lateral cortex of the inferior colliculus (LCIC), intercollicular nuclei, and superior colliculus (SC). Single-labeled neurons dominated the LCIC, whereas double-labeled neurons were evident in the more rostral regions, including the intercollicular nuclei and SC. Knowledge gained from this protocol provides a foundation for ongoing studies in the lab that aim to correlate neurochemical marker patterning with that of guidance molecule expression and afferent-efferent connections.

**P49** Joy Zhang, Dr. Terrie Rife

**The effect of alpha-synuclein on gene expression in SK-N-MC cells**

Neurodegenerative diseases are characterized by misfolded proteins that aggregate within the cell. One common neurodegenerative disease is Parkinson’s Disease (PD), which is diagnosed during an autopsy by noting the presence of -synuclein aggregates called Lewy bodies.  Alzheimer’s Disease (AD) is interpreted and defined by the neurofibrillary tangles made up of hyper-phosphorylated tau protein.   Although these proteins are synonymous with having the diseases, the cellular role of these proteins is not entirely understood.  Current literature has noted that both alpha- synuclein and tau are found in the nucleus and function to bind DNA.  It is hypothesized that the tau and alpha-synuclein proteins may assume the function of transcription factors. Chromatin Immunopreciptation for alpha-synuclein and Microarray data of what genes altered when exposed to high levels of alpha-synuclein and tau are publically available.  We are using this data and other Bioinformatics Techniques to identify possible transcriptional targets of alpha-synuclein and tau. To verify that these gene targets are transcriptionally regulated by tau or alpha-synuclein, SK-N-MC cells will be grown with varying levels of tau and alpha-synuclein. RNA will be harvested from these cells and used for quantitative PCR analysis of the selected genes.   Determining the potential effects of transcriptional activity of tau and alpha-synuclein on other genes, will further the understanding of Parkinson’s and Alzheimer’s disease.

**P50** Rebecca J. Livermore, Ranjit Z. Virk, Dr. Katrina Gobetz

**A comparison of diet between fossil and modern equids using plant remains from dental plaque**

The dual aim of this study is to 1) identify plant remains in the dental calculus (calcified plaque) of fossil equids to determine diet and make generalizations about habitat, and 2) do a tandem analysis of a reference sample of modern horses to compare appearance and abundance of masticated plant remains with a fossil sample of horses. Calculus, which begins as a microbial mat on a tooth and becomes cemented over time, acts as a trap for minute plant fibers and cells. We are using calculus to gauge diet (hardwood-browser, grazer, mixed-feeder) and habitat (montane, xeric, arid, lowland, etc.) of fossil species, as it provides direct evidence of plants consumed over the life of an individual. We sampled calculus for this study from 5 fossil horse genera ranging in size and geologic age from the tiny Eocene horse, *Orohippus*, to the nearly modern-type, larger Miocene equid, *Hipparion*. After processing these samples to isolate phytoliths (silicified plant cells) and xylem tissue fibers, we identified plant remains and compiled them for each fossil sample. We compared the prevalence of woody taxa or grasses among compiled fossil sample groups to elucidate diet and ecosystem. Towards our second aim, we compared modern and fossil calculus to confirm that the plant remains in the fossil horse teeth are similar to the known appearance of chewed plants in modern horse samples. We then conducted a statistical analysis to ascertain whether there were significant differences in abundance of plant material in modern vs. fossil equid sample groups.

**P51** \*Cecilia Rogers, Dr. Heather Griscom

**Investigating the ecology and habitat modeling for *Solanum conocarpum* on St. John, USVI**

Approximately two thirds of St. John is National Park territory. However, the island has been threatened with tourism and the installation of mega marinas, greatly impacting the island biodiversity. One species that may become extinct due this degradation is *Solanum conocarpum. S. conocarpum* (marron bacora) is a rare shrub, endemic to the dry forests of St. John, USVI. This plant is a species of conservation concern and is one of very few native and endemic plants on the island. Very little is known about the ecology and reproduction of *S. conocarpum*. Most plants are found clustered around John’s Folly and Nanny Point. Recently it has been noted that the greatest recognized threat to it is a lack of natural reproduction, possibly due to suboptimal habitat conditions. This study will investigate the ecology and habitat of *S. conocarpum* in order to construct a habitat model. We aim to find a relationship between plant population health and habitat conditions that will help improve conservation strategies. Data will be taken from the 11 natural populations on the island. Each individual will be counted, measured in height (m) and stem diameter (cm). Additionally, all fruits and flowers will be counted, if present at all. At each site, slope, aspect, sunlight, and elevation data will be taken to create a habitat model. 100g of soil taken at 10cm will be obtained from each population site and analyzed for contents including nitrogen, carbon, and moisture. General observations will be taken in efforts to see what species interact with *S. conocarpum*. We predict that the largest populations are still at Nanny Point and on regions with drier, poorer quality soils. We predict optimal habitat to be around Nanny point, and on the opposite side of the island at Brown Bay. Restoring this rare plant would help with preservation of the St. John National Park lands, giving service members a stronger argument to preserve the island. Species restoration is integral to saving St. John from further degradation in the midst of the sixth global mass extinction.

**P52** Anna Nordseth, Dr. Heather Griscom

**Coffee carbon stocks, pests and diseases under varied shade management: a review**

Coffee agroforestry systems have received increased attention in recent decades because of their capacity to improve agricultural sustainability. Coffee (*Coffea arabica*), one of the most economically important crops, is widespread throughout the tropics and can have serious environmental impacts. To ensure sustainable coffee production, it is critical that coffee systems are maintained to maximize carbon storage and minimize susceptibility to pests and diseases. This study reviews the history of coffee production, from forested coffee systems to industrial coffee monocultures. We describe the five classifications for coffee systems, and use them as a framework to compare aboveground carbon stocks across management regimes and site conditions with a specific focus on coffee tree carbon stocks. Finally, we synthesize literature on coffee pests and diseases under varied shade management and investigate how these relationships may be altered with future climate change. Although no direct relationship was found between levels of shade management and coffee carbon stocks, site conditions such as precipitation and temperature appear to influence coffee carbon stocks depending on whether the coffee is grown in sun or shade. Additionally, the relationship between shade management and the prevalence of pests and diseases was unclear. Increasing our understanding of how site conditions and system shade management affect coffee carbon stocks and the prevalence of pests and diseases will allow for improved land-use planning, greater resiliency of coffee systems, and increased potential for agroforests to play a role in climate mitigation.

**P53** \*Charles Holmes, Dr. Heather Griscom

**Variation in mycorrhizal colonization of american ginseng (*Panax quinquefolius*) from varying soils and forests: a preliminary study**

American ginseng (*Panax quinquefolius*) is one of the most culturally and economically important non-timber forest products (NTFPs). *P. quinquefolius* has been previously observed to be colonized by arbuscular-vesicular mycorrhizal (AM) fungi, endosymbionts which provide nitrogen and phosphorus in exchange for carbohydrates, but variation in this symbiosis due to environmental factors has been poorly characterized. This study was a preliminary investigation of variation in AM colonization due to plant size, forest composition, and soil type. After collecting root samples from wild-simulated *P. quinquefolius* at various sites in West Virginia and Virginia, roots were cleared and stained to visualize mycorrhizae before quantification using a light microscope. Arbuscular colonization of large plants was approximately twice that of small plants; the inconsistency of this result with previous research may be due to differing nutrient availability to noncommercial populations of *P. quinquefolius* than farm-raised American ginseng. While sample sites varied in the presence of ginseng-associated trees, all plants sampled were within 10m of at least one of these tree species, and *P. quinquefolius* associated with the greatest proportions of these species displayed the greatest arbuscular colonization. In contrast with previous studies, AM colonization was not correlated with soil phosphorus, as the degree of colonization varied among different sites which had comparable available phosphorus. This study, while limited by a low sample size due to the low availability of noncommercial wild-simulated ginseng, provides direction for future research into the role of AM fungi in the conservation of this valuable forest herb.

**P54** Shelby Saroka, James Collins, Dr. Chris Lantz

**Establishment of Genotyping Protocol for Interleukin-3 Transgenic Fluorescent Reporter Mice**

Interleukin-3 (IL-3) is a hematopoietic growth factor and immunoregulatory cytokine that is secreted by various immune cells during the host response to malaria infection. However, the in vivo cellular sources and the kinetics of IL-3 production during malaria infection is unknown. Using CRISPR/Cas technology, we have generated heterozygous IL-3 fluorescent reporter mice that produce IL-3 along with the fluorescent protein ZsGreen (ZsG). Identification of IL-3 producing cells can thus be examined in these mice using fluorescent microscopy or flow cytometry. To obtain wild-type controls (ZsG-/ZsG-) and reporter mice homozygous for the ZsG gene (ZsG+/ZsG+) requires mating heterozygous mice, genotyping the offspring, and subsequently expanding breeding colonies. To genotype mice, we extracted DNA from mouse tails and established a PCR protocol that allowed the identification of homozygotes and heterozygotes based on size differences in PCR products visualized by agarose gel electrophoresis. We found that our breeding scheme generated the expected frequency of the various genotypes. Out of 58 offspring, 15 pups (25%) were found to be ZsG+/ZsG+, 26 pups (45%) were identified as ZsG+/ZsG-, and 13 (22%) were ZsG-/ZsG-. We are in the process of expanding our colonies of wild-type and homozygous reporter mice to provide sufficient numbers of animals for future experiments.

**P55** Marina Barmanova, Dr. Tim Bloss

**Role of BTF3/ICD-1 in control of cancer-associated gene expression in *C. elegans*** Cells constantly undergo misfolded protein stress, thus triggering unfolded protein response (UPR), which can decrease misfolded protein levels or initiate apoptosis depending on stress levels. The nascent polypeptide-associated complex (NAC) is heterodimeric chaperone that prevents misfolded protein stress in the ER; depletion of the NAC triggers a strong UPR. When not in complex, the individual subunits of the NAC are thought to regulate gene expression; in particular, the beta subunit of the NAC (βNAC) is thought to affect the expression of Epherin receptor 2 (EPHB2), a gene associated with cancer. I want to determine the putative regulation of the EPHB2 homologue Vab-1 by the bNAC homologue ICD-1 in *C. elegans*, with the hopes of gaining insight into the roles βNAC and EPHB2 in the development of cancer. The gonad of C. elegans provides a cancer cell model, and was characterized in vab-1 knockouts with wild type and depleted levels of ICD-1. The depletion of ICD-1 from vab-1 knockout worms generated mutant gonad phenotypes that differ from vab-1 knockouts alone: primarily the growth of the gonad past the vulval midline of the worm, indicating continued gonadal growth throughout adulthood, abnormal for a wild type *C. elegans*. This phenotypic difference between treated and untreated vab-1 knockouts indicates ICD-1 may not be the only factor controlling the expression of Vab-1 in *C. elegans*.

**P56** Sydney Ashton, Dr. Rocky Parker

**Searching for sex differences in snake skin**

Sexual dimorphism in expression of steroid hormone receptors in garter snake skin

The production of many secondary sexual signals, including pheromones, is controlled by sex hormone action at the sites of signal synthesis. The red-garter snake (*Thamnophis sirtalis parietalis*) is an ideal vertebrate for studying the interaction between steroids and sexual signals: males exclusively rely on skin-based female pheromones during courtship and pheromone composition is augmented by treatment with sex steroids (e.g., males produce female pheromone if implanted with estrogen). But how do steroid hormones promote pheromone expression at the molecular level in snake skin? Feminizing effects of estrogens on sexual signals are known to result from activation of estrogen receptors α (ESR1) and/or β (ESR2), while masculinizing effects of androgens arise from androgen receptor (AR) activation. We hypothesized that ESR1, ESR2, and AR are expressed in garter snake skin but their expression is sex-dependent with female skin expressing higher levels of ESRs and males higher expression of AR. To test this, red-sided garter snakes (n=10 males, n=10 females) were collected in the spring mating season, and mRNAs from skin and control tissues (liver, gonad, kidney, intestine) were extracted and used to synthesize cDNAs. Primers were designed using the available T. sirtalis genome (NCBI) and tested in real-time PCR reactions. While all three receptor types were expressed in male and female skin, ESR1 was more highly expressed in female skin while AR showed greater expression in male skin. We thus attribute the feminizing effect of estrogen on pheromone phenotype in males to their lack of circulating estrogen and subsequently dormant ESRs. Further, activated AR may have an antagonistic role in pheromone expression since testosterone is a known inhibitor of pheromone production in male garter snakes.

**P57** Monifa Williams, Zachary Schuhmacher, Patrick Kilkenny, Klebert Feitosa, Dr. Roshna

Wunderlich

**Characterization of intrinsic foot muscles using novel Polydimethylsiloxane**

**PDMS methodology and EMG measurements**

The intrinsic foot muscles (IFM) include twenty muscles that originate and insert within the foot. They are responsible for flexion, extension, abduction, and adduction. Literature suggests that the IFM are active during walking, support the longitudinal arch, and assist in balance. Weakness and atrophy of IFM due to conditions such as diabetic neuropathy can lead to foot deformities, plantar fasciitis, ulceration, and falls in elderly populations. Current techniques characterize IFM using flexion measurements, but functions and activation of IFM have not been thoroughly studied, partially due to challenges in measuring strength during toe abduction. To validate IFM function, we have measured IFM activation during activities using electromyography (EMG) of six intrinsic foot muscles. Challenges in strength measurements were overcome by using Polydimethylsiloxane (PDMS) gel as a force transducer for characterizing the strength of IFM abductors. Pairing EMG with the PDMS measurements, along with other techniques used in literature allows us to determine when muscles are active and interpret function. Our probe into new aspects of IFM functions/activation may directly lead to the development of IFM strengthening techniques to improve function, prevent injury, and enhance performance. This project is a collaborative effort between the animal movement lab in Biology and the soft complex materials lab in Physics and Astronomy.

**P58** Elizabeth Rogers, Stephanie Sharpes (co-first authors), Dr. Kyle Seifert

**The Antibacterial and Biofilm Disruption Activity of Novel Tris-Cationic Amphiphiles**

Approximately 2 million people are infected annually by bacteria resistant to commonly used antibiotics, resulting in increased hospital-associated morbidity and mortality and placing a large economic burden on patients and hospitals. The formation of bacterial biofilms poses a unique threat by increasing resistance to antibiotics and chemical removal, leading to increased risk of transmission and re-infection. Amphiphiles are a well-studied class of compounds with documented antibacterial properties. Four novel series of tris-cationic, double-tailed amphiphiles were synthesized for use in this study. Derivatives with 12 carbons per tail showed the most efficient bactericidal activity against seven different species of bacteria (two Gram-negative and five Gram-positive) for all series with MIC values as low as 2 μM. Additionally, the most effective compounds disrupted up to 68% of established *P. aeruginosa biofilms* at concentrations comparable to the antibiotic tobramycin or common disinfectant benzalkonium chloride. Amphiphiles offer a potential route for the prevention of the spread of common nosocomial infections. The continued research in this field with the development of more diverse and novel of amphiphiles may be very useful in decreasing the growing threat of antibiotic resistance and nosocomial infections.

**P59** Morgan Hennessy, Dr. Oliver Hyman, Dr. Elizabeth Doyle, Dr. Ray Enke

**Integration of Geospatial Analysis into Biology Undergraduate Curriculum**

Integration of geographic information systems (GIS) and geospatial analysis into undergraduate biology curriculum benefits students by enhancing more traditional biology concepts and techniques. Here we present a modular series of adaptable geospatial laboratory activities that can be woven into a broad spectrum of biology curriculum to enhance learning outcomes. Modules include the use of freely available online geospatial tools and a mobile smartphone application for use in field activities. As proof of principal, we report the integration of these modules into an upper level undergraduate genomics course to augment a DNA Barcoding course-embedded research module. Course assessment data demonstrate that geospatial activities provided students with a novel experience using geospatial analyses in biology. Preliminary data have prompted further widespread implemented of these geospatial modules into the JMU Foundations in Biology core curriculum in which all JMU Biology and Biotechnology majors enroll in.   
Acknowledgements: This work was supported by a JMU Geospatial Mini Grant.

**testosterone is a known inhibitor of pheromone production in male garter snakes.**

**BIOSYMPOSIUM 2017 - Author Index**

**Numbers refer to (T)alks, (P)osters**

T1-T14: TH 1-4:45pm *Biosci 2007*

T15-T25: FRI 9-noon *Biosci 2007*

P1-P21: TH 11-1pm *2nd Fl. Foyer and hallway*

P22-P59: FRI 2-4pm *2nd Fl. Foyer and hallway*

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