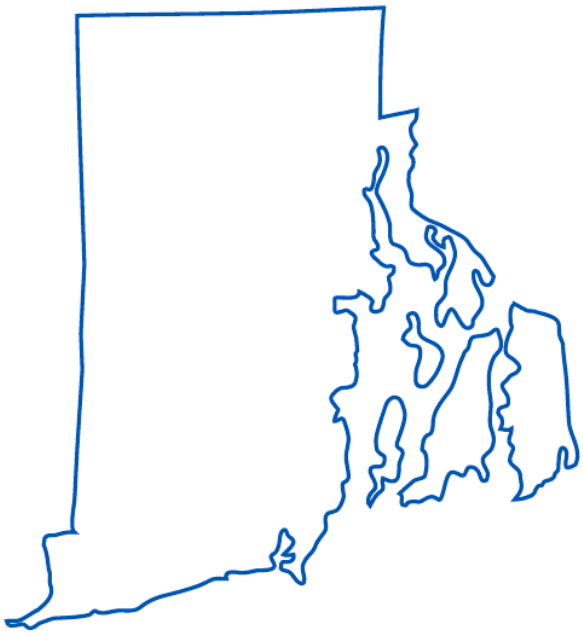
2022 Rhode Island Summer Undergraduate Research Symposium



Friday, July 29, 2022

University of Rhode Island

Center for Biotechnology & Life Sciences
Fascitelli Center for Advanced Engineering
Paramaz Avedisian '54 Hall, College of Pharmacy

Sponsored by

RHODE ISLAND INBRE
Biomedical Research Excellence

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Coastal Ecology Assessment Innovation & Modeling

URI MARC U*STAR Program

RI-INBRE, RI NSF EPSCoR C-AIM & MARC U*STAR

2022 Rhode Island Summer Undergraduate Research Symposium

8:00 – 9:00 AM

Check-in & Continental Breakfast

- CENTER FOR BIOTECHNOLOGY & LIFE SCIENCE

Poster Set-up

- FASCITELLI CENTER FOR ADVANCED ENGINEERING
- PARAMAZ AVEDISIAN '54 HALL, COLLEGE OF PHARMACY

9:00 – 9:30 AM

Welcoming Remarks

- CENTER FOR BIOTECHNOLOGY & LIFE SCIENCE

Samantha Meenach, RI-INBRE Training Core Director

President Marc Parlange, University of Rhode Island

Provost Sean Reid, Providence College

Provost Rupendra Paliwal, Bryant University

Jim Lemire, RI C-AIM Undergraduate Research Coordinator

9:30 – 11:00 AM

Poster Session A

- FASCITELLI CENTER FOR ADVANCED ENGINEERING
- PARAMAZ AVEDISIAN '54 HALL, COLLEGE OF PHARMACY

11 AM - 12:30 PM

Poster Session B

- FASCITELLI CENTER FOR ADVANCED ENGINEERING
- PARAMAZ AVEDISIAN '54 HALL, COLLEGE OF PHARMACY

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POSTER PRESENTATION SCHEDULE

*** PLEASE NOTE: Posters are to be set up prior to the welcoming remarks and should remain up until 12:30 PM. Posters are to be presented according to the schedule below.*

Session	Presentation Times
A	9:30 – 11:00
B	11:00 – 12:30

Poster numbers	Location
1-23 (A & B)	Fascitelli Center for Advanced Engineering (1st floor)
24-51 (A & B)	Fascitelli Center for Advanced Engineering (ground floor)
52-76 (A & B)	Paramaz Avedisian '54 Hall College of Pharmacy

POSTER SESSION A

9:30 – 11:00 AM

Fascitelli Center for Advanced Engineering, 1st Floor
A-1 to A-23

Fascitelli Center for Advanced Engineering, Ground Floor
A-24 to A-51

Paramaz Avedisian ’54 Hall, College of Pharmacy
A-52 to A-76

Exploring the microbiology of quiescence in the coral *Astrangia poculata* as a model for understanding microbiome interaction with environmental disturbance

Meriel McGovern[1], Alicia Schickle[1], Anya Brown[2], Casidhe Hughes[1], Emma Place[1], Nikki Rosa[1], Amy Apprill[3] & Koty Sharp[1]

[1]Marine & Natural Sciences, Roger Williams University, Bristol, RI
[2]Evolution & Ecology, University of California, Davis, CA
[3]Marine Chemistry & Geochemistry, Woods Hole Oceanographic Institution, Woods Hole, MA

Coral microbiomes mediate the health of tropical corals and their survival of environmental disturbance. The local temperate coral *Astrangia poculata* is a valuable experimental organism for studying the dynamics and diversity of coral microbiomes because its microbiome composition is similar to that of many tropical corals and shifts predictably across seasons. During winter months, *A. poculata* undergoes a period of dormancy, or quiescence, likely triggered by cold temperatures (<5^{o}C), in which the polyps fully retract, become unresponsive to stimuli, and cease feeding. We propose that changes in the *A. poculata* microbiome during the winter-spring transition may be a model for coral microbiome recovery from environmental stress and/or disturbance. Here we designed an aquarium system to induce quiescence in *A. poculata*. Replicate aposymbiotic *A. poculata* colonies from Rhode Island and Massachusetts were held in individual tri-pour beakers with independent seawater flow and divided into two tanks (n=10 per geographic origin, per tank). The ambient (control) treatment was held at 19^{o}C. The quiescent treatment started at 19^{o}C, ramped down to 5^{o}C over 1 week, held at 5^{o}C for 4 weeks, and then ramped up to 19^{o}C over 1 week. Replicate seawater and coral mucus samples were collected during the ramp-down and ramp-up; during quiescence; and for two weeks post-quiescence. These samples were processed for 16S rRNA gene sequencing. Results suggest that the community composition of the mucus microbiome shifted depending on the quiescence vs ambient treatments, on the population, as well as during, and emergence from quiescence. Dispersion (inter-individual variability) of microbiome composition in *A. poculata* colonies is similar before and during quiescence but decreases during emergence from quiescence. *A. poculata* microbiome diversity decreased during quiescence but increased after emergence from quiescence. These patterns were consistent with previously reported results from wild *A. poculata* colonies sampled around quiescence. However, the taxonomic composition differed between tank and wild colonies with at least one exception: a *Sulfitobacter* sp., increased in proportional abundance in *A. poculata* colonies as they emerged from tank-induced and wild quiescence. This study highlights the community composition changes associated with quiescence and provides clues to the patterns and taxa that may play a role in recovery from an environmental disturbance.

This work was supported by RI NSF EPSCoR C-AIM

Tracking the Fate of Polyethylene Microbeads and Microbead-Associated Microbes in Exposure Assays of the Coral, *Astrangia poculata*

Emma Place, Alicia Schickle, Nicole Rosa & Koty Sharp

Biology, Marine Biology & Environmental Science, Roger Williams University, Bristol, RI

Microplastics (<5mm diameter) are detrimental to marine life and are found in even the most remote oceanic regions. Microplastics are consumed, often unintentionally, by a variety of marine organisms, including *Astrangia poculata*, a temperate heterotrophic coral. *Astrangia poculata*, a suspension feeder, feeds on zooplankton and other particles in the water column making it vulnerable to incidental microplastics ingestion, either directly from the water column, or via contaminated prey. This study aims to use *A. poculata* and its copepod prey, *Pseudodiaptomus pelagicus*, as an experimental system to track the fate of microplastics and microplastics-associated microbes. In this study, we developed methods to reliably coat polyethylene microbeads with GFP-tagged *Escherichia coli*. Also, *P. pelagicus* was fed GFP-E. coli-biofilmed UV-fluorescent polyethylene microbeads, and the copepods were subsequently fed to *A. poculata.* Fluorescence imaging was used to image the microbeads and *E. coli* cells. Data demonstrated that *A. poculata* ingested microplastics via contaminated *P. pelagicus*. *Astrangia poculata* was also directly fed GFP-E. coli biofilmed microbeads and imaged via epifluorescence microscopy. Imaging is ongoing to localize GFP-*E.coli* cells resulting from this ingestion; however, initial findings suggest that *E. coli* is transferred into *A. poculata* via direct ingestion and indirect ingestion (via *P. pelagicus*) of the microbeads. These findings will provide important insight and new methods for exploration of the fate of microplastics and their associated microbes throughout food webs, specifically via indirect ingestion of microplastics in marine organisms across multiple trophic levels.

This work was supported by RI NSF EPSCoR C-AIM

Determining the origin of potential coral probiotic *Pseudoalteromonas rubra* from laboratory and wild samples.

Casidhe Hughes[1], Kira Bernabe[2], Meriel McGovern[3], Emma Place[3], Nicole Rosa[1], Alicia Schickle[3], David Nelson[4] & David Rowley[5]

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[4]Biology, University of Rhode Island, Kingston, RI
[5]Pharmacy, University of Rhode Island, Kingston, RI

The microbiome of tropical corals plays a significant role in coral survival and fitness, benefitting the metaorganism through processes such as nitrogen fixation, digestion of complex molecules, energy by photosynthetic production, and protection against pathogens. Generally, coral microbiome composition is distinct from the seawater microbiome, suggesting that corals actively recruit and regulate specific bacterial associates. *Pseudoalteromonas* sp. is a genus of bacteria widely documented to be associated with tropical corals, and recent work demonstrated that bacteria from this group produce a variety of compounds with antimicrobial properties, likely in a protective role for the coral host. Strains of *Pseudoalteromonas rubra* isolated from tropical corals have been shown to produce secondary metabolites, including N-acyl homoserine lactones, which can both facilitate and interfere with bacterial quorum sensing, and prodigiosins, which appear to have antimicrobial activity and can induce larval settlement in at least one coral species. *Pseudoalteromonas rubra* KB322 was isolated from seawater in a recirculating tank system holding the temperate coral *Astrangia poculata* in the Roger Williams University Wet Lab. *Pseudoalteromonas rubra* KB322 inhibits the growth of the deadly coral and shellfish pathogen, *Vibrio coralliilyticus*. Here we present work to identify the origin of *P. rubra*, both in the RWU Wet Lab and in the wild. A sequence-specific PCR primer (Pr1283R) targeting *P. rubra* KB322 was designed and tested. A PCR-based assay with Pr1283R was used to survey the presence of this species in various locations in the Wet Lab, including multiple seawater sources, *A. poculata* surface mucus layer, and crustose coralline algae (CCA) growing in the *A. poculata* system. PCR with Pr1283R was also used to test for the presence of *P. rubra* in wild-collected seawater and CCA. To date, the surveys only detected the sequence in the Wet Lab CCA. Bacterial strain CH007, with colony and cell morphology consistent with KB322, was isolated from Wet Lab CCA. Identity of CH007 and of Pr1283R PCR amplicons from the RWU Wet Lab CCA are currently being confirmed using 16S rRNA gene cloning and sequencing. Ongoing work includes testing for potential of KB322 and CH007 to induce *A. poculata* larval settlement. Our research provides key insights into where this potential bacterial probiotic exists, and how it may interact with native temperate corals.

This work was supported by the National Science Foundation

Exploring indicator displacement assays for phosphate detection in seawater

Quade Oser, Paul Eyo, Francis Radics & J.J. Breen

Chemistry & Biochemistry, Providence College, Providence, RI

Indicator displacement assays are based on the optical signal modulation of a noncovalently bound indicator upon dissociation by an analyte species. Our work has focused on exploring the lower detection limits for fluorescence displacement assays for inorganic phosphate in seawater using complex ions containing two di(2-picolyl)amine ligands (also called DPA or bis(2-pyridylmethyl)amine), each coordinating a zinc cation. We have been exploring the use of the well-established $[Zn_2(H\text{-}bpmp)]^{3+}$ ligand system with two fluorescent dyes that absorb and emit visible wavelengths; salicyl fluorone (SF) and a dihydroxy bodippy dye (Dh-BD, or 10-(3,4-dihydroxyphenyl)-5,5-difluoro-1,3,7,9-tetramethyl-5H-dipyrrolo[1,2-c:2',1'-f][1,3,2]diazaborinin-4-ium-5-uide). To date, our limits of detection for a simple fluorescence assay with their dyes is on the order of a single ppm's for SF and single ppb's for DH-BD both in the presence of 0.1 M NaCl.

This work was supported by RI NSF EPSCoR C-AIM

Exploring Rhodamine B Hydrazides for Nitrite (and Nitrate) Fluorescence Detection

James Bateman & J.J. Breen

Chemistry & Biochemistry, Providence College, Providence, RI

A rhodamine B hydrazide fluorescent probe was prepared from rhodamine B and phenyl hydrazine in a two-step reaction following the work of Lee and coworkers (Bull. Korean Chem. Soc. 2013. Vol. 34, No.2). The rhodamine B hydrazide (RB-H) product was characterized by absorbance, and emission spectroscopy as well as NMR. Dilute solutions of RB-H in a pH 4.0 Acetate: MeOH (1:1) buffer were exposed to nitrite in the presence of chloride and without. Preliminary results show a limit of detection of 500 ppb (1.0×10^{-5} M) with 0.18 M NaCl and 50 ppb (1.0×10^{-6}M) without. Experiments are ongoing to improve detection limits, ease of use, and to extend into detect nitrate following reduction to nitrite with zinc metal.

This work was supported by RI NSF EPSCoR C-AIM

Alternative Technologies for Assessing Fish Populations Using Environmental DNA

Grace Foltz[1], Matthew Rock[2] & Jeffrey Markert[1]

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[2]Environmental Biology, Providence College, Providence, RI

The Narragansett Bay forms New England's largest estuary and is home to thousands of species of fish, plants, and other wildlife. Human activity including climate change, industrial waste, commerce, and fisheries all impact the Bay in various ways. In order to determine how fish communities change in response to human activity, we focused on a new way to survey fish based on environmental DNA (eDNA) - the DNA found in water that comes from shed cells or tissues. Our pilot work aimed to establish a protocol that would estimate fish populations using a quantitative polymerase chain reaction (qPCR) from water samples where our target species resided. These species and their target gene region include: Tautogs *Tautoga onitis*, mtD-loop; Winter Flounder *Pseudopleuronectes americanus*, mtD-loop; and Bluefish *Pomatomus saltatrix*, COI. Our project used manual fish surveys conducted in collaboration with Dr. David Taylor's team from Roger Williams University to see whether environmental DNA concentrations reflect the number of fish counted. A total of 65 500 mL water samples were collected near the shore of three Rhode Island locations: Festival Pier in Pawtucket, Goddard State Park and Conimicut Point Park in Warwick. Throughout the course of the project, we have developed protocols for filtering our water samples, created primers and probes for the qPCR machine, and optimized the conditions for the qPCR runs. Our initial analysis of qPCR data using the Tautog and Winter Flounder probes suggests eDNA quantities roughly correlate with census counts for target species within habitats across time. However, differences in baseline signal between habitats suggest that the tool must be calibrated differently for each habitat surveyed. Developing these alternative technologies for assessing fish populations may ultimately be cost-effective, efficient, less labor intensive, and less invasive. Our data will hopefully create a baseline for surveys to detect changes in fish populations as the environment continues to change.

This work was supported by RI NSF EPSCoR C-AIM

Mt. Hope Bay Sediment and Martian Regolith Simulant Nutrient Flux Impact on Algal and Diatoms Oxygen Production

Carly Ferreira[1], Stephen O'Shea[1] , Isabel August[2], Teagan Bellitto[2] & Maggie Stefanowicz[2]

[1]Chemistry, Roger Williams University, Bristol, RI
[2]Roger Williams University, Bristol, RI

Sediment nutrient anions (NO_2^-, NO_3^-, PO_4^{3-}, NH_4^+) and cations (Cu^{2+}, Fe^{3+}, Zn^{3+}, Ni^{2+}) fluxes play an essential role in the requirements of algae (*Tisochrysis lutea*) and diatom (*Thallassiosira weissfloggi*) growth. Sediment fluxes have been used to characterize Mt. Hope bay and Mars regolith simulant water profiles and give insight into the nutrient levels and easily extractable metals released in the water column. The research investigated the impact sediment fluxes had on the algae and diatom oxygen production and variation in nitrate and nitrite concentration in the above water profiles sediments in BOD microcosms. The underlying sediment substrates were collected from sites in the estuarine Mt. Hope Bay in RI and contrasted with Martian regolith simulant. The extent of these fluxes was gauged by the O_2 production of algae and diatom communities by fluorescence-quenching optodes. Spectrocolormetric HACH® spot tests determined the suite of nutrients and the concentration of metal cations was determined by ICP OES from filtered samples.

This work was supported by RI NSF EPSCoR C-AIM

Investigation of Phosphorus Deposition in Mount Hope Bay from Freshwater Inputs

Maggie Stefanowicz[1] , Isabel August[2] , Teagan Bellitto[3] , Carly Ferreira[4] & Stephen O'Shea[4]

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[2]Biochemistry, Roger Williams University, Bristol, RI
[3]Marine Biology, Roger Williams University, Bristol, RI
[4]Chemistry, Roger Williams University, Bristol, RI

The importance of elemental phosphorus (P) bioavailability as a limiting nutrient within marine biological cycles has recently been refocused to quantifying the sources and sedimentary sinks within estuarine environments. Sedimentary P is an ecologically conserved critical nutrient for successful plant growth but can lead to eutrophication on the release of high anthropogenic concentrations. This study encompassed the environmentally exposed Mount Hope Bay (MHB) watershed spanning Rhode Island and Massachusetts state lines. MHB watershed is fed by four freshwater inputs (Taunton, Lee, Cole, and Kickemuit rivers) exiting out through the East Passage and the Sakonnet River to the Atlantic Ocean. Twenty-four surface (~5cm) sedimentary sampling sites were investigated across the watershed. The concentrations of nutrients (PO_4^{3-}, NO_3^-, NO_2^-, NH_4^+) were evaluated using Hach® spectrocolormeteric tests, using various chemical extraction methods: Total P (TP) (Koroleff 1983), Inorganic P (Ruttenberg 1992), Water Soluble Phosphorus (WSP), Readily Desorbable Phosphorus (RDP), Algal Available Phosphorus (AAP), and Bicarbonate Extractable Phosphorus (Olsen-P), (Zhou. *et al*). Confirmatory evidence for the presence of orthophosphate in extracts was also shown by P31 nuclear magnetic resonance. Each of these sites was further characterized by its carbon and carbonate content, granular size, pH, orp, salinity and acid extractable metals. This research demonstrated the various extraction techniques followed the same phosphorus content profile with an increasing concentration gradient transect from the river's source to the river's mouth paralleling increasing salinity.

This work was supported by RI NSF EPSCoR C-AIM

Microplastic Pollution in Aquatic Ecosystems of Rhode Island

Logan Beattie[1], Sarah Davis[2], Andrew Davies[2] & Coleen Suckling[2]

[1]College of Engineering, University of Rhode Island, Kingston, RI
[2]College of Environment & Life Sciences, University of Rhode Island, Kingston, RI

As global plastic production continues to grow, increasing amounts of microplastic (MP) pollution has been found in the environment. MPs have been found in every studied environment on the planet, and are known vectors for pollutants, disease, and invasive species, which can lead to negative outcomes for animals, ecosystems, and humans. There has been a global effort to quantify their presence and impacts and provide the information needed for waste and ecosystem management. However, little work has been done to quantify MPs in the aquatic environments of Rhode Island. The goal of this work was to design and operate a filtration device that could quantify MPs in freshwater sources, an important introduction source to the coast region. Three stainless steel filters were used in an innovative pump sampling system: 280 µm, 100 µm, and 10 µm. The sampled filtrate was then processed to isolate and extract MPs. First, samples were sonicated to remove particles trapped in filter pores. Then chemical digestion with potassium hydroxide and citric acid was used to dissolve organic matter. Density separation using potassium formate (d = 1.520 g*cm-3) was then applied to separate MPs. Lastly, a polycarbonate filter (5 µm mesh size) and a vacuum were used to isolate MPs for imaging. MPs were imaged using an automated Olympus BX63 Microscope in the Suckling Lab and were then characterized using Raman Spectroscopy. Throughout the sampling and processing procedure, strict contamination control and procedural controls were followed. The work conducted by our lab lays the framework for further work to be done studying the presence of MP pollution.

This work was supported by RI NSF EPSCoR C-AIM

Effect of Size on Foraging Behavior of the Green Sea Urchin *Strongylocentrotus droebachiensis*

Marguerite McNamara, Tara Plee & Coleen Suckling

Fisheries, University of Rhode Island, Kingston, RI

Strongylocentrotus droebachiensis, the Green Sea Urchin, is a cold water species, found in both the Northern Atlantic and Pacific oceans. Green Sea Urchins are commercially important in the United States (primarily in Maine), Canada, Japan, and parts of Northern Europe, commonly harvested from the wild. Unfortunately, the green sea urchin fishery in Maine has been overexploited causing increased regulation such as an upper size limit (test diameter (TD) 2 1/16" to 3" = 52-76 mm) which may be impeding population growth due to conspecific competition. More specifically, size restrictions could be creating competition between larger individuals, preventing smaller individuals from accessing high value food sources, such as kelp, therefore preventing their ability to grow into larger, harvestable sized animals for the market This pilot study was conducted to determine the veracity of a possible link between abundance of urchins in particular size classes and competition while foraging. Green sea urchins were separated into size classes [Large and Small], placed alone into separate tanks with one piece of kelp (*Saccharina latissimi*) on the opposite end of the tank. Their behavior was filmed for 1.5 hours, and the time taken to reach the kelp, the amount of time associated with kelp, and the amount of kelp consumed. Small urchins were found to be much more efficient foragers in all respects. Further study is needed to determine if this is representative of trends at a larger scale,and how foraging behavior is affected by the presence of other urchins.

This work was supported by RI NSF EPSCoR C-AIM

Working towards sustainability in green sea urchin (*Strongylocentrotus droebachiensis*) production

Zachary Robinson[1], Chris Boylston[1], Tara Plee[2] & Coleen Suckling[2]

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[2]College of Environment & Life Sciences, University of Rhode Island, Kingston, RI

Strongylocentrotus droebachiensis, also known as the green sea urchin, is a cold water species commonly found in the Gulf of Maine. In recent years, due to global warming, the Gulf of Maine has been experiencing an increasing number of heat waves, which is when a specific area of the ocean warms to temperatures above the 90th percentile temperature range for 5 days in a row. Stress priming, a process by which animals are subjected to short periods of stressful environmental conditions, has shown to be successful in enhancing tolerance to field heterogeneity. In this study, we began to test this concept through a pilot trial to determine whether exposure to increasing seawater temperatures can be tolerated by hatchery-raised juvenile green sea urchins. This trial will set up the baseline for what the maximum temperature juvenile urchins can be exposed to before mortality occurs. Future research will determine if we can make adult green sea urchins more resilient to heat waves by exposing them to elevated temperatures as juveniles in a controlled environment.

This work was supported by RI NSF EPSCoR C-AIM

Stakeholder engagement on the Woonasquatucket through community programming and undergraduate research

Rola Jiang, Andre Gomes & Daniel B. Hewins

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The Woonasquatucket (Algonquian: where the saltwater ends) River stretches nearly 26 km with a watershed area of 130 km2 in rural and urban central RI before joining the Providence River and draining into Providence Bay. Its historical legacy as a part of the industrial past of RI has led to it being designated an American Heritage River, while more recently the river has undergone a revitalization to create green spaces for the people living along its banks. To understand the needs of stakeholder organizations and communities in the Woonasquatucket River Watershed, we developed a two-dimensional approach to generating stakeholder feedback and implement basic research in support of community organizations. Currently our work is a collaborative effort with Revive the Roots (Revive) a non-profit organization dedicated to sustainability and community engagement through a diverse set of programs at the headwaters of the headwaters of the 'Woony'. Our collaboration with Revive is allowing us to deploy supporting communications to the ecology of the watershed, the fate and transport of plastic pollution, while generating a network of community members to provide feedback and lead the next generation of programs along this waterway. Many of our communications are interpretive and include QR codes that encourage community members to communicate with our team. In addition, Revive has become a starting point for understanding the needs of organizations and how basic ecosystem research can be leveraged to support their aims as a nucleus of stakeholder engagement. We plan to work with the Revive Land Stewardship board to generate a better understanding of how the agricultural past of RI affects plant and soil ecology, and how contemporary land use and recreation affects plastic pollution along the Woonasquatucket River by sampling soils and vegetation across a range of ecosites. In this poster we highlight recent and planned community engagement projects, and proposed basic research planned to begin in Autumn of 2022.

This work was supported by RI NSF EPSCoR C-AIM

Long-term annual variations in the abundance of juvenile finfish and portunid crabs relative to increasing water temperatures in Narraganset Bay, RI

Lydia Furtado[1], David Taylor[1], Anna Gerber-Williams[2] & Conor McManus[2]

[1]Biology, Roger Williams University, Bristol, RI
[2]Division of Marine Fisheries, RI Environmental Management, Jamestown, RI

This study examined the effect of increasing water temperature on the estuarine biota of Narraganset Bay, Rhode Island (RI), with a specific focus on three juvenile finfish and portunid crab species: bluefish, summer flounder, winter flounder, blue crab, green crab, and lady crab. Data supporting this study were derived from the RI Department of Environmental Management, Division of Marine Fisheries (DMF) long-term seine survey (1988-2021). Sampling of target species was performed monthly from June to October using a beach-seine set (61 × 3.1 m) at 15 fixed sites throughout the bay. Target species were enumerated, and variations in annual abundance [Log(CPUE)] were examined relative to year and ambient temperature via linear regression analyses. Mean monthly temperatures significantly increased in Narragansett Bay ($R^2 = 0.15$, $p < 0.05$) at a rate of ~ 0.03 ^oC per year, i.e., 1 ^oC increase from 1988 to 2021. During this time series, bluefish abundance remained relatively constant ($R^2 = 0.08$, $p = 0.10$). Conversely, summer flounder and blue crab abundances significantly increased over time ($R^2 = 0.31$-0.38, $p < 0.0001$-0.0005), with summer flounder abundance unrelated to temperature ($R^2 = 0.005$, $p = 0.69$) and blue crab catches positively correlated with warming waters ($R^2 = 0.15$, $p < 0.05$). Winter flounder and green crab abundances significantly declined across years ($R^2 = 0.34$-0.56, $p < 0.0001$-0.0005), with catches inversely related to temperature ($R^2 = 0.15$-0.26, $p < 0.005$-0.05). Similarly, the lady crab population decreased over time ($R^2 = 0.78$, $p < 0.0001$), but annual abundances were independent of temperature ($R^2 = 0.06$, $p = 0.20$) and negatively correlated with increased blue crab numbers ($R^2 = 0.18$, $p < 0.05$). These results suggest that Narragansett Bay has undergone significant changes in species composition over the last three decades, with seasonal water temperatures partially explaining annual variations in the abundance of select organisms (e.g., winter flounder, green crabs, and blue crabs). Moreover, putative biotic interactions among estuarine organisms may regulate intra-specific abundances, e.g., lady crabs potentially displaced by the influx of blue crabs via predator-prey and/or competitive interactions. Further analysis of the expansive RI DMF data set relative to other biotic or abiotic factors may provide additional insights into the mechanisms underlying temporal variations in the community composition of the bay.

This work was supported by RI NSF EPSCoR C-AIM

Spatial and temporal variations in the total mercury concentration of bivalves from Rhode Island estuarine and marine waters

Jayden Schoeps[1], David Taylor[1], Patrick Barrett[2] & Conor McManus[2]

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Shellfish, including bivalves, support substantial commercial and recreational fisheries in Rhode Island (RI), and, therefore, may be an important dietary source of contaminants for human consumers. Mercury (Hg), for example, is a pervasive contaminant in estuarine and marine environments, which often bioaccumulates in the tissues of resident biota. In this study, total Hg concentrations were measured in five bivalve species sampled from RI inshore and coastal waters (Mar-Aug, Nov-Dec, 2006-2022), and results were examined relative to the geographic location and time of year individuals were collected. Mean Hg concentrations varied significantly across bivalve species, with the highest concentrations measured in the hard shell clam *Mercenaria mercenaria* (mean ± SD Hg = 0.21 ± 0.14 ppm dry wt; n = 212), followed by the soft shell clam *Mya arenaria* (0.15 ± 0.10 ppm; n = 86), horse mussel *Modiolus modiolus* (0.14 ± 0.04 ppm; n = 30), and ribbed mussel *Geukensia demissa* (0.14 ± 0.10 ppm; n = 165), and lowest in the blue mussel *Mytilus edulis* (0.10 ± 0.05 ppm; n = 217). The Hg concentrations of bivalves were significantly higher in the upper Narragansett Bay relative to the lower bay (0.17 ± 0.14 ppm and 0.11 ± 0.04 ppm, respectively), with spatial differences in contamination likely reflecting species-specific distribution patterns and a north-south gradient in anthropogenic pollution sources. Bivalve Hg concentrations were also generally higher during the late fall and early spring (Nov-Dec, Apr: 0.21 ± 0.13 ppm) compared to summer months (Jul-Aug: 0.10 ± 0.07 ppm), which may be caused by seasonal patterns in environmental conditions (e.g., temperature and/or dissolved oxygen) and prey availability. Finally, for commercially utilized bivalves of regulation size (blue mussel, hard and soft shell clam), all individuals had Hg concentrations below the U.S EPA threshold level of 2.1 ppm dry weight (converted from 0.3 ppm wet weight using 86% moisture content of bivalve tissue). These results suggest the consuming bivalves from the Narragansett Bay pose minimal Hg-related risks to human health.

This work was supported by RI NSF EPSCoR C-AIM

Mercury contamination and bioaccumulation trends in commonly consumed sunfish (*Lepomis* spp.) from southern Rhode Island

Brianna Lotti[1], David Taylor[1], Melanie Hedgespeth[2] & Mark Cantwell[2]

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Mercury (Hg) is a widespread environmental contaminant that may poses a risk to human health, with human exposure occurring primarily through fish consumption. This study examined Hg concentrations and bioaccumulation trends in commonly consumed freshwater sunfish, including the bluegill (*Lepomis macrochirus*), redbreast (*L. auritus*), and pumpkinseed (*L. gibbosus*). Sunfish were collected from the Pawcatuck River and Kedniker Island Pond (Grills Preserve, Bradford, RI) in June 2022 using an electrofisher and rod & reel. Dorsal muscle tissue was excised from each individual and measured for total Hg content (ppm dry weight) using atomic-absorption spectrometry, after which results were analyzed relative to fish species, body size (cm total length) and estimated age (years). Mean Hg concentrations did not differ among sunfish species (mean ± SD: Bluegill = 0.83 ± 0.37, n = 26; Redbreast = 0.92 ± 0.27, n = 11; Pumpkinseed = 0.88 ± 0.36, n = 9). All sunfish experienced increased Hg levels at larger body sizes. Moreover, there was no significant difference in Hg bioaccumulation rates, i.e., Hg-age relationships, between bluegill and redbreast sunfish. The absence of inter-specific differences in sunfish Hg content and bioaccumulation rates is attributed to the similar life history characteristics and trophic ecology among species. From a human health perspective, 11% of the adult sunfish analyzed in this study exhibited Hg concentrations exceeding the U.S. EPA threshold level of 1.36 ppm dry weight. Therefore, only very frequent consumption of sunfish from the study area (> 1 meal per week) are anticipated to pose a risk to human health owing to Hg exposure.

This work was supported by RIDEM

Sensitive Electrochemical Nitrite Sensing Based on Carbon Black gold Nanoparticles modified Screen Printed Electrode

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Nitrite salts are widely used in industrial manufacturing and overfertilization can lead to the contamination of surface water and groundwater. For example, nitrites can cause the transformation of normal hemoglobin to methemoglobin, leading to loss of hemoglobin's ability to transport oxygen and can also lead to accelerated algae growth and eutrophication of large bodies of water. There is an unmet need for portable, reliable and economical sensor for nitrites due to its ubiquitous nature and toxicity. Herein, we demonstrate that a screen-printed electrode (SPE) can be utilized for the detection of nitrite in aqueous solutions. Coating the sensor with carbon black gold (CB/AuNP) nanoparticles creates an increase in sensitivity and signal amplification. The sensing of nitrite can be done at the micro molar level via cyclic voltammetry, differential pulse voltammetry and square wave voltammetry. The sensor demonstrated a wide linear range of concentration detection from 0.1 to 70 mM at pH 3 with a detection limit of 0.1 mM nitrite in water sample. The practicality of this sensor approach was shown by measuring the concentration of nitrite ions in tap and spring water samples. The disposable CB/AuNP SPE offers great potential for a low cost, portable and economic approach for nitrite detection in seawater samples.

This work was supported by RI NSF EPSCoR C-AIM

Flow Electrochemical Sensor based on Carbon black-Gold Nanoparticle (CB-AuNP) Polylysine Nanostructured Sensor for Electrochemical Detection of Phosphate in water samples

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Phosphate is an important part of sea ecosystems as plant life needs it to survive, however it can lead to eutrophication, an overgrowth of plant life in seawater (algal blooms). This results in reduction or elimination of dissolved oxygen that is crucial for fish and other sea creatures. Herein, we report on a highly sensitive electrochemical sensor for detection of phosphate levels in seawater samples. The method is based on measuring phosphomolybdate complex formed by a reaction between phosphate and molybdate which is subsequently detected on the electrode surface. To enhance the sensitivity of the sensor and lower the detection limit, a modified screen-printed electrode was used. Screen-printed electrodes modified with carbon black decorated with gold nanoparticles (CB-AuNP) successfully increased the detection of phosphomolybdate complex reduction at + 64 mV vs. Ag/AgCl. Analytical figures of merit including reagent concentration, working potential, flow rate and concentration of CB-AuNP on electrode surface were optimized. Results show a dynamic range at low phosphate concentrations from 0.05 – 50 μM with detection limit of 0.05 μM phosphate, calculated as three times the standard deviation of the blank divided by the slope of calibration curve. Accuracy was also assessed using recovery studies and by measuring phosphate in real water samples with good agreement with the spectrophotometric method. Results show great promise for a simple, low-cost method for real-time, on-site detection of phosphate in seawater samples.

This work was supported by RI NSF EPSCoR C-AIM

The Effects of Pollutants on Developing Vertebrates

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Plastic pollutants are an ongoing problem in our oceans and waterways, in particular, microplastics – plastic fragments that do not exceed five millimeters in diameter. Microplastics present an emerging threat to aquatic ecosystems. Narragansett Bay alone contains roughly 40 – 4.6 million particles/100 g of sediment. The impact of plastics on the environment starts at the lower trophic levels with primary consumers, and inevitably works its way up to larger marine mammals. Many organisms are exposed to microplastics through a variety of avenues, such as habitat disruption and contaminated food sources. In addition, chemicals that are insoluble in water are released into the environment by the sorbing and desorbing properties of microplastics. The general consensus of most studies point to the conclusion that microplastics are detrimental to the health of marine organisms. Previous findings suggest that toxins from plastics can have fatal impacts on developing embryos, in some cases reducing survivability and driving developmental abnormalities. However, the specific cellular/molecular pathways affected in the presence of microplastics and their chemicals remain unknown. This lack of understanding makes it important to take a closer look at how microplastics and their chemicals affect gene expression during embryonic development in aquatic organisms. For this study, we looked at the affect microplastics have on the embryonic development of Atlantic silverside (*Menidia menidia*) – a wild species native to the Narragansett Bay. Atlantic silversides play an important role in the Narragansett Bay ecosystem because not only are they primary and secondary consumers, feeding both on producers and small marine creatures, but they are also an important food source for larger fish and coastal birds. Using zebrafish embryos as a baseline, we look to assess the molecular effects of Polyethylene microplastics on silverside embryos in a controlled laboratory environment. Using in situ hybridization and qPCR we will monitor the development of early forming tissues and gene expression in developing embryos. These experiments will provide us with a better understanding on how microplastics, and the chemicals associated with them, affect vertebrate development in aquatic environments, and give us insight into the effects that they will have on the entire ecosystem.

This work was supported by RI NSF EPSCoR C-AIM

Cloud Storage for SimpleChartsRI.com website

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SimpleChartsRI.com is a web-based tool that helps Rhode Island high school teachers and students use data visualization in education. We added individual user accounts along with a login feature so users could save any charts they created to their account. Saved charts can be viewed from anywhere the user is logged in and downloaded as .png files. Additionally, when registering users are prompted for "Student", "Teacher" or "Other" status. When users are logged in the website offers features tailored specifically for "Teacher", "Student" or "Other" users.

This work was supported by RI NSF EPSCoR C-AIM

Simple Charts for Everyone: SimpleChartRI UX Redesign

Shun Huang

Graphic design, Rhode Island School of Design, Providence, RI

This poster describes our user experience(UX) redesign for SimpleChartsRI, a web-based data visualization tool. The redesign process was divided into three parts. Firstly, we conducted a UX audit to find the problems in the existing design. Secondly, we used user analysis to clarify the target user, understand user needs and behavior. Finally, we built a prototype to organize all the insights into the effective design solution. The new UX design enhances the usability, accessibility and aesthetic of the tool.

This work was supported by RI NSF EPSCoR C-AIM

The Effect of Raw Materials on the Pore Structure of Ceramic Water Filters and its Relationship with End User Performance Metrics

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Contaminated drinking water leads to a variety of diseases that cause sickness and death; however, an estimated 884 million people live without access to clean drinking water. One technology focused on providing clean drinking water to under-served communities is ceramic water filters (CWFs). CWFs are a point-of-use technology that are low cost and use locally available clay and burnout materials during construction. The goal of this project is to relate manufacturing processes (selecting and processing clay and sawdust) to the pore structure and determine the effect of the pore structure ceramic strength and bacterial removal. Clays sourced from filter factories at 9 locations around the world and hardwood sawdust were used in the construction of the filters. The porosity of the filters was measured by soaking in water and weighing to determine void volume. Compositional analysis for the clays was performed using X-ray diffraction (XRD). Clay minerals were extracted from the bulk material, and both were analyzed using XRD. Compressive and flexural strength were used as metrics for ceramic strength and bacterial removal was quantified using E. coli as the model organism. Compressive strength was shown to decrease with an increase in porosity and initial testing shows that the ceramics have a high flexural strength. Bacterial removal testing was performed but due to limitations with the tests, conclusive results are yet to be determined. This study will demonstrate how the quality of raw materials used for CWF construction influence performance metrics that are important to the users. This information will assist CWF factories in choosing materials and processes that can be used to manufacture high quality filters.

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Two-Dimensional Tracking of Quadri-Flagellate *Ulva* Zoospores

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A critical stage in the development of anti-biofouling materials is testing under controlled and real conditions. For these tests, reproductive quadri-flagellate zoospores from the marine algae *Ulva*, are commonly used as a model biofouling organism. Tracking the movement of these zoospores enables us to visualize and understand Ulva zoospore movement patterns and the biofouling process. This experiment aims to test a two-dimensional particle tracking method for zoospore tracking applications. The experiment used a Lecia DMi8 Inverted Optical Microscope with a magnification of 100x and a Teledyne Photometrics Prime 95B high-speed camera at a speed of 100 frames per second and a pixel size of about 1 micron. These instruments were used to record videos that were then run through a MATLAB particle tracking algorithm capable of mapping the movement of individual zoospores contained within a volumetric slide made of one glass slide, three coverslips, and epoxy. A diluted solution of seawater, gametes, and zoospores was pipetted into this volumetric slide, recorded, and then run through the algorithm delivering a graph of particle paths that show visual similarities to known movement patterns of *Ulva* zoospores. Of these known patterns and the most visible from a top-down two-dimensional perspective was “spinning” shown as a continuous circular plot on the graph created by the algorithm. The spinning occurs in the last stage in a zoospore’s progress towards attaching to a surface, becoming a biofoulant. The results of this experiment show that tracking and analyzing zoospore movement is possible with this method, however, the five other known movement patterns; orientation, wobbling, gyration, hit & run, and hit & stick are difficult to definitively pick out in the data. Optimization is needed before this method can be considered completely viable for zoospore tracking and analysis.

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Optimization of ImageJ Cell Counting of *Ulva* spp. Zoospores

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Ulva spp. is a type of algae known for causing biofouling of marine sensors. One method of studying the ability to remove *Ulva* spp. spores from a surface is to count and compare their zoospores after being treated by a pressurized waterjet. On June 6th and 20th, 2022, *Ulva* spp. were collected from Mackeral Cove, Jamestown RI. They were combined with sterile seawater to stimulate the release of *Ulva* spores due to temperature shock. Next, the quadriflagellated zoospores were identified with a microscope. The slides underwent 20psi from the water jet in order to remove loosely adhered spores. For each sampling, 12 control samples and 12 water jet samples were imaged with 7 fields of view taken for each. An upper threshold of 144 and a lower threshold of 255 were applied, which highlighted the *Ulva* separate from other particles. Likewise, a 14.5-infinity pixel size was also set in order to properly count cells based on their size. When compared to manual counts, the mean percent error was 2.28% and the range was 0 to 4.96%. There was a large reduction in the adhered cells after the water jet test. Both the cell count and area distribution on the slide saw a major decline.

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Identifying the Effect of N-terminal Acetylation on Thymine DNA Glycosylase Activity in Chromatin

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Thymine DNA glycosylase (TDG) is an enzyme that initiates the first step of base excision repair (BER) on DNA. TDG can identify and excise mismatched thymine, uracil, 5-fluorouracil, among other modified bases. TDG contains a long, unstructured N-terminus with multiple known acetylation sites. N-terminal acetylation is an irreversible protein modification present in 80% of proteins in the human proteome and produces a myriad of effects that are highly dependent on the protein that it is affecting. One challenge that glycosylases face in eukaryotes is when DNA is packaged into chromosomes, which gives rise to issues with steric interactions between the enzyme and the histone core, accessibility issues depending on how the DNA is oriented relative to the histone core, and even changes in the periodicity of DNA affecting substrate recognition. However, repair is still observed in the most basic unit of chromosomes, the nucleosome core particle (NCP). Using site-directed mutagenesis to convert lysine residues to glutamines as acetylation analogs, we observe the effects of N-terminal acetylation on thymine DNA glycosylase's ability to identify and excise lesions in DNA that is packaged into NCPs to elucidate how this modification affects glycosylase activity.

This work was supported by RI-INBRE

Image Analysis for Pharmaceutical Compound Characterization

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Pharmaceutical sciences are using polymeric micro- and nanoparticles to reduce the toxicity and side effects of drugs[1]. The size and shape of the particles are critical for their pharmaceutical efficiency. Electron microscopy is often used to visualize these particles and quantify their morphology. However,electron microscopy is costly and time-consuming. Another drawback of electron microscopy is that generally the particles are only characterized in 2D, and thus the 3D information of the particle shape and volume quantification is lost. In contrast, X-ray microscopy is a 3D technique that can analyze hundreds of particles in a single run with minimal sample preparation and human interaction.XRM can be used to quantify the particle size distribution and shape of micro- and nanoparticles for inhalation drug formulations, but its use is not yet popular in the pharmaceutical literature[2,3,4]. In this work, we compared the analysis of particle size and shape using scanning electron microscopy(SEM) and X-ray microscopy(XRM) and developed the image analysis protocols to measure particle volume using XRM. NanoComposite microParticles(nCmP) of acetalated dextran were synthesized. We analyzed the particle morphology by XRM using different acquisition settings.Image analysis protocols on software Dragonfly were developed to quantify the particle size and shape of the microparticles, including a systematic comparison of the effects of image filters applied to the obtained data. Our findings indicate that XRM is difficult to use for particles below 5μm diameter, and we were not able to visualize the individual nanoparticles within the nCmP.

[1.] Wim H De Jong and Paul JA Borm, Drug delivery and nanoparticles:Applications and hazards,International Journal of Nanomedicine,(2008)3,133-149
[2.] Hrishikesh Bale, Robert Hammond, Permesh Gajjar, Ioanna Danai Styliari, Philip Withers, Phillip Withers, and Darragh Murnane, Unlocking secrets of inhalation blends through X-ray computed tomography and microscopy,Microscopy and Microanalysis,(2021)27,292-295
[3.] Edward Yost, Pascal Chalus, Shawn Zhang, Stefanie Peters, Ajit S. Narang, Quantitative X-ray microcomputed tomography assessment of internal tablet defects, Journal of Pharmaceutical Science,(20219)108,1818-1830
[4.] Joseph P. Neilly, Leilei Yin, Sarah-Ellen Leonard, Paul J.A. Kenis, Gerald D. Danzer, Ashtamurthy S. Pawate, Quantitative measures of crystalline fenofibrate in amorphous solid dispersion formulation by X-ray microscopy

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Ac-Dex Spray-Dried MP Project

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Lung cancer is the deadliest form of cancer, as more than half of patients diagnosed with the disease perish within a year. The conventional treatment for lung cancer is chemotherapy administered either orally or via IV. However, potent chemotherapeutics delivered via these routes of administration typically result in undesirable side effects, potentially hindering overall treatment efficacy. The pulmonary route of administration offers a more localized drug delivery platform which can result in an increase in drug concentration at the target site, thus leading to lower required doses and a reduction in off-site toxicity and related side effects. Acetalated dextran (Ac-Dex) is a biodegradable, hydrophobic polymer prepared via a one-step reaction by reversibly modifying dextran with acetal groups, therefore changing its solubility. Ac-Dex has many properties suitable for controlled drug release and was used as the excipient in the production of dry powder aerosol particles.

In this investigation, aerosolized microparticles loaded with the lung cancer chemotherapeutics doxorubicin (DOX) or paclitaxel (PTX) were produced via spray drying.The spray drying conditions including flow rate, inlet temperature, drug loading, and overall feed concentration were varied to produce microparticles with varying morphologies and aerosol dispersion properties. Following microparticle fabrication, the thermal phase transitions, crystallinity, size, and morphology of the systems were evaluated, in addition to drug loading, drug release, and aerosol dispersion properties. Our results demonstrated that microparticles with a higher drug loading of DOX show a more spherical morphology while microparticles with a higher drug loading of PTX show a more collapsed and wrinkled morphology. Raising the pump rate and lowering the inlet temperature during spray drying produced microparticles with a more spherical morphology with both drugs, however it also resulted in a bumpy surface on the particles. Introducing water as a cosolvent with ethanol to PTX-loaded microparticles resulted in mixed morphology, with some particles exhibiting a spherical, hollow, and bumpy structure and others exhibiting a collapsed and corrugated structure. The investigation is still ongoing as conditions for perfectly spherical microparticles are being optimized.

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Cell Membrane Coated Nanoparticles for Treating Pulmonary Fibrosis

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Pulmonary fibrosis (PF) is a progressive, incurable, terminal illness that occurs when lung tissue becomes damaged, thickened, and scarred, leading to decreased lung function, and ultimately death. Idiopathic pulmonary fibrosis (IPF) incidences, in which the cause of the disease is unknown, are significantly rising in the US and are expected to affect over 17 per 100,000 people in the near future. Currently, the conventional methods of treating IPF have faced many challenges including limited treatment options, repetitive treatment regimens, lack of inhalable or particle-based formulations, and imprecise lung-specific delivery and tissue targeting. The short half-lives, frequent dosing, and systemic side effects of current therapeutics make treating PF difficult. Administering aerosolized therapeutics through the pulmonary route is an attractive alternative for treating IPF as it offers increased drug concentration at the target site, less frequent dosing, reduced cytotoxicity, and minimized adverse effects. The goal of this study is to create cell membrane-coated nanoparticles (CMCNP) capable of intentionally interacting with PF-related lung tissue to slow the progression of the disease. CMCNPs are capable of attenuating pulmonary fibrosis through enhanced transepithelial delivery of NP, increased retention of NP in lung tissue, and cell-specific targeting via the cell membrane coating. Long term, CMCNP have the potential to improve the quality of life for patients with pulmonary fibrosis.

Acetalated dextran (Ac-Dex) is a pH-sensitive dextran derivative polymer prepared via a one-step reaction by reversibly modifying dextran with acetal groups. Due to its effective drug encapsulation and acid-triggered drug release, this polymer is suitable for controlled drug release. CMCNPs were fabricated by producing Ac-Dex NP loaded with the model drug curcumin, which were then coated with cell membrane vesicles. Following particle fabrication, the CMCNPs were incorporated into cell membrane-coated nanocomposite microparticles (CMC-nCmP) via spray drying, after which their size, morphology, and drug loading was investigated. Current results demonstrate that CMC-nCmP encapsulate drugs and have a collapsed and corrugated morphology with uniform spherical nanoparticles embedded within them. This research is still ongoing as CMCNP drug release, internalization, and cell uptake are yet to be investigated.

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Is the posterior parietal cortex necessary for response renewal in novel and familiar contexts?

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The Posterior Parietal Cortex (PPC) integrates multisensory information and aids in decision making. Recent evidence suggests PPC involvement in the renewal of an extinguished conditioned fear response in novel contexts but not familiar ones. It is unknown if context-dependent renewal is limited to fear-based memories or whether renewal would also occur with positive conditioned stimuli. Using chemogenetics and stereotaxic brain surgery, a virus was injected into the dorsal or caudal PPC of experimental rats. During behavioral testing, the virus will be fully expressed through an IP injection of Clozapine N Oxide (CNO) before renewal, which will temporarily inactivate the PPC. Control rats, who received craniotomies but no virus injections, were trained, extinguished, and tested for renewal of positive and negative associations in both familiar and novel contexts. After the IP injection (which should have no behavioral effect), control rats demonstrated renewal in both familiar and novel contexts for both positive and negative stimuli. Experimental rats will be tested in the same paradigm and are expected to renew both negative and positive associations in a familiar context, but not a novel context as they cannot fully integrate their surroundings without PPC activity. If the PPC controls context-dependent renewal beyond the fear renewal paradigm, these results may have implications for the use of positive reinforcement in pharmaceutical treatments of addiction and phobias in humans.

This work was supported by RI-INBRE

Gender moderates the relation between opioid symptom severity and positive emotional avoidance

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Intro: Positive emotional avoidance is characterized by attempts to alter the form, frequency, and context of positive emotional experiences (Hayes et al., 1996). Positive emotional avoidance can elicit negative interpretations of previous positive emotions, which can result in increased distress and further avoidance of positive emotions, including through alcohol and drug use (Weiss et al., 2018). Expectations of emotional responses vary across men and women, linked to gender norms established by society (Parker et al., 2021). For example, positive emotional avoidance has been shown to occur more often in men than women (Schick et al., 2020). Research suggests that among men, positive emotional avoidance may stem from a socialized expectation of limited emotional expression and avoidance of situations that could elicit a charged emotional response (Schick et al., 2020). Therefore, we hypothesize that gender will moderate the relation between opioid use disorder (OUD) symptom severity and positive emotional avoidance, such that men who experience positive emotional avoidance will have significantly higher levels of OUD symptom severity compared to women.

Methods: Participants were 85 trauma-exposed community individuals (Mage = 43.45, 35% women, 54.1% white) who used opioids over the past 30 days. At baseline, participants were interviewed with a structured diagnostic assessment (SCID-5) to assess OUD. An empirically-validated self-report measures assessed emotional avoidance.

Results: Results revealed that gender moderated the relation between OUD symptom severity and positive ($b = -0.40$, $p = 0.03$), but not negative ($b = -0.08$, $p = 0.24$), emotional avoidance. Analysis of simple slopes revealed that OUD symptom severity was significantly positively associated with positive emotional avoidance among men ($b = 0.31$, $p = 0.03$), but not women ($b = -0.10$, $p = 0.45$).

Conclusion: Results from our study revealed that gender moderated the relation between OUD symptom severity and positive emotional avoidance, such that that OUD symptom severity was significantly positively associated with positive emotional avoidance among men, but not women. These findings suggest the potential utility of intervening on positive emotional avoidance—such as targeting an increase in positive emotional acceptance and willingness—among men to reduce OUD symptom severity.

This work was supported by RI-INBRE

The Longitudinal Relation between Borderline Personality Disorder Symptoms and Positive Emotion Dysregulation in Women Experiencing Intimate Partner Violence

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Introduction:
Borderline personality disorder (BPD) symptoms are a chronic mental health concern that commonly entail unstable emotions, low-self esteem, and problematic social relationships (APA, 2013). BPD affects roughly 3% of the population, making it one of the most common psychiatric disorders (Carpenter et. al., 2013). Those experiencing intimate partner violence (IPV) have a higher risk of developing BPD (Munro & Sellborn, 2020). Central to BPD is higher levels of emotion dysregulation (APA, 2013), including among women experiencing IPV (Ross, 2011). However, research is limited by a near-exclusive focus on the dysregulation of negative (versus positive) emotions. Positive emotion dysregulation is defined as the nonacceptance of positive emotions, difficulties engaging in goal-directed behaviors when experiencing positive emotions, and impulse dyscontrol in the context of positive emotions (Weiss et al., 2015). Research shows positive emotion dysregulation is linked to IPV (Simpson et al., 2015), and preliminary studies among adolescents indicate positive emotion strategies are related to BPD symptoms (Kim et. al., 2014). However, there is a need to assess the relation between BPD symptoms and positive emotion dysregulation among women experiencing IPV, a high-risk sample. This study will utilize a longitudinal design to examine the bidirectional relation between positive emotion dysregulation and BPD symptoms among women experiencing IPV. We hypothesize that positive emotion dysregulation will predict BPD symptoms, and in turn, BPD symptoms will exacerbate positive emotion dysregulation.

Method:
Participants were 172 community women (M age = 40.33, 40.7% white) experiencing IPV and using substances who participated in a baseline interview and then returned for a one-month follow-up session. Participants self-reported on BPD symptoms and positive emotion dysregulation. A cross-lagged panel model was run in Mplus Version 8 to examine the bidirectional relations between BPD symptoms and positive emotion dysregulation, while controlling for initial levels of symptoms.

Results:
Initial levels of positive emotion dysregulation were not associated with BPD symptoms at one-month follow-up, when controlling for initial BPD symptoms (B = -0.06, SE = 0.11, p = 0.60). However, initial levels of BPD symptoms were significantly associated with positive emotion dysregulation at one-month follow-up, when controlling for initial positive emotion dysregul

This work was supported by RI-INBRE

Elucidating the Active Site Environment and Mechanism of Human DNA Polymerase Theta

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DNA is constantly being damaged from a variety of factors, one of which being UV light. DNA polymerases are enzymes that play an important role in DNA replication and repair. Human DNA Polymerase Theta (Pol θ) is involved in DNA repair and helps to protect cells from damaging environmental factors like UV despite being an error-prone, low fidelity enzyme. Understanding the dynamic mechanism in which Pol θ incorporates the correct versus the incorrect nucleotide during repair is pivotal to understanding its low fidelity status. More importantly, we can compare incorporation rates and active site micro-environments during phosphodiester bond formation with cancer-associated variants of Pol θ to better understand how Pol θ may protect against DNA damage. To do this, we monitored nucleotide incorporation in real-time using a stopped-flow instrument, by visualizing changes in fluorescence between a 2-aminopurine fluorescence labeled DNA substrate and Pol θ's active site to explore the hypothesis. As expected, with WT there was a decrease in fluorescence signal due to pi stacking when the polymerase is shifting to integrate the correct nucleotide. Interestingly, we observed a little fluorescence change with cancer-associated variant, L2538R, suggesting that the variant has a different active site environment compared to WT implying altered DNA repair capabilities.

This work was supported by RI-INBRE

Nanoparticle Delivery of Anti-Inflammatory Medication as a Possible Treatment of Rheumatoid Arthritis

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Rheumatoid arthritis is an autoimmune disease that causes the body to attack healthy cells and leads to painful swelling around joints. There are five different types of treatments for rheumatoid arthritis, each one targets a different aspect of the disease and carries its own set of side effects. Currently there is no cure for rheumatoid arthritis, there is only means of mediating swelling and managing pain. The medication we are working with, 3-a-Anincholestane (3AC), inhibits the initiation of arthritis and reduces the ability for the disease to progress. Previous tests using 3AC, delivered through injection, lead to weight loss and death in live mouse models. A solution to this is to use nanoparticles to deliver the 3AC medication in a more targeted approach than traditionally medication delivery systems. We made nanoparticles from a biocompatible polymer called poly lactic-co-glycolic-acid (PLGA) which reduces the likelihood of the nanoparticles being targeted by the immune system. Our preliminary study looks at uptake and survivability of macrophage cells J7741A.1 when exposed to different types of nanoparticles.

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Cloning Danio Rerio DNA POLQ for Expression System Development

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The POLQ gene encodes for DNA Pol θ, a low-fidelity polymerase essential to various DNA repair pathways initiated by DNA damage. Mutations in POLQ have been detected in melanoma patients, and in vitro studies have observed decreased Pol θ activity with such mutations suggesting a potential role for Pol θ in cancer. To better understand the in vivo effects of mutant Pol theta, we aim to express the cancer-associated variants in the animal model zebrafish (Danio rerio). An initial BLAST alignment showed sequential similarity between human POLQ and zebrafish POLQ domains, which suggests zebrafish as a viable model to express POLQ cancer-associated variants. Despite this, we sought to express and purify Zebrafish Pol θ (zPol θ) to compare DNA polymerase activities prior to in vivo studies. This project describes the successful cloning and initial expression studies for zPol θ with the future goal of translating this work into a full animal model.

This work was supported by RI-INBRE

Synthesis of Functionalizable Cyclic Ester Monomers for the Preparation of Degradable Polymers for Disease Treatment

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Multi-drug resistance represents a major hurdle in disease treatment. The use of functionalizable polymers for new drug delivery methods is a promising proposition for the future of the biomedical field. We can apply polymers to drug delivery by appending the desired drug onto a polymer that contains a targeting moiety and is hydrolyzable. This allows the drug to enter the cell with the polymer. Once the polymer is inside the cell, it will be hydrolyzed to unmask the drug and release the drug into the body. Our lab is working toward the design of a new class of highly functionalized polymers that can be metabolized in vivo for use in drug delivery applications. This focus of this project is to develop modular synthetic routes toward functionalizable cyclic ester monomers that can be converted into hydrolyzable polymers for drug delivery.

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Quantifying the acid content of vanadium containing electrolyte solutions

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The vanadium redox flow battery (VRFB) relies on the reduction of V^{5+} to V^{4+} on the positive electrode and the oxidation of V^{2+} to V^{3+} on the negative electrode via the reactions below.

$VO_2+ + H_2O \rightarrow VO^{2+} + 2H^+$ (1)

$V^{2+} \rightarrow V^{3+} + e^-$ (2)

In both cases, the reaction is performed in high concentrations of H_2SO_4. During charge/discharge of the VRFB, protons shuttle across the membrane to maintain charge balance. Additionally, reaction (1) above produces H^+, further complicating knowledge of the H^+ concentration. We have previously shown that the H^+ concentration directly effects both the physio-chemical behavior of the cell and the electrochemical performance. 1,2 Thus upon running the cell, it is impossible to know for certain what the H^+ concentration is. To our knowledge, no simple/quick laboratory method has been developed to determine such. For example, titration with OH^- produces a host of V(OH)x precipitates which clouds the solution and makes it impossible to achieve a titration endpoint. This presentation will highlight our work to quantify the H^+ concentration in the presence of vanadium species.

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Optimization of Biogenic Manganese Oxide Nanoparticles for Environmental Applications

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Manganese oxide nanoparticles (MnONPs) have a high redox potential, and can oxidize organic compounds, such as endocrine disrupting chemicals (EDCs) and other industrial organic pollutants. The physicochemical properties of biogenic manganese oxide nanoparticles make them potentially applicable for water treatment. This study uses a naturally occurring microorganism, Pseudomonas putida GB-1, to biosynthesize MnONPs. The purpose of this project was to optimize the purification process of biogenic MnONPs from the bacterial biomass while preserving their reactivity and physicochemical properties. The P. putida was cultured in batch mode to produce MnONPs. After culture, an oxidative purification protocol was performed by treating the biomass containing MnONPs with 1.25% NaOCl for 8 hours at pH 7. The concentration of MnONPs was measured after the bacterial culture and throughout the purification process using inductively coupled plasma mass spectrometry (ICPMS). During the batch culture, the Pseudomonas converted 98% of the aqueous Mn (II) to Mn (III/IV) particles. Treatment with NaOCl or buffer (pH 7) released up to % 22 of MnONPs from the biomass. Major losses of MnONPs occurred during cleaning and washing stage and purification from biomass stage. After purification, the size of the particles in the supernatant were measured to be 129□26 nm using dynamic light scattering. The study showed us that purification process works and the synthesized nanoparticles by P. putida were recoverable, however the amount of MnONPs decreased with each step of the purification and cleaning process. The Future work will focus on improving the cleaning and purification procedures to reduce loses of MnONPs.

This work was supported by RI-INBRE

Characterization of Tracer Particle Dynamics Through Biomimetic Hydrogel Materials

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Transport of nanoparticles in viscoelastic materials is essential for applications ranging from 3D printing to pharmaceutics. In this work, we investigate how nanoparticles diffuse through biomimetic emulsions and how the dynamics change based on the emulsions volume fraction. The emulsions are created by dispersing cyclohexane into an index-matched water-glycerol solution through sonication to produce a transparent liquid. The emulsion viscosity is measured through rheology, and the particle dynamics are quantified with optical microscopy and tracking algorithms. When conducting optical microscopy, a fluorescent filter is used to illuminate the fluorescent particles and droplets, allowing us to track their movements. With this data, we compare the measured diffusivity to theoretical predictions using the Stokes-Einstein equation and relate this to the transport of nanoparticles through biomimetic tissues. At lower volume fractions, there is a greater volume of the continuous phase allowing the particles to diffuse more freely, whereas at higher volume fractions, the dispersed phase is greater and inhibits particle movement. For this reason, we find that the diffusivity of the emulsion droplets and nanoparticles decrease as the volume fraction increases. Future work will investigate how these dynamics depend on the strength of attraction between droplets, controlled by the addition of telechelic triblock copolymers. Our findings will be used to design novel 3D printing inks and synthetic tissue analogues that could be used in both clinical and industrial applications due to their structure and versatility.

This work was supported by RI-INBRE

Investigating Carbon Nanotubes as Long-Term Bioimaging Probes

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Near-infrared (NIR) fluorescence, sensitivity to environmental conditions, and biocompatibility are some properties of single-walled carbon nanotubes (SWCNTs) that enable their investigation as biological sensors. These novel optical properties and biological interactions must be explored in depth before they can be applied for reliable biosensing applications in vivo. The DNA functionalization of SWCNTs, which facilitates biocompatibility and has been shown to influence their intracellular behavior, is one focal point that demands extensive research. Here, we examine how differences in DNA wrappings are reflected in the NIR fluorescence intensities of SWCNTs within live cells. Two different DNA sequences ((GT)6 or (GT)30) were chosen to functionalize SWCNTs for use within two types of live cells (macrophages (RAW 264.7 cell line) or fibroblasts (3T3 cell line)). Next, each sample of SWCNTs was incubated with a flask of live cells, washed, and then placed into fresh media. The treated cells were imaged with a NIR hyperspectral microscope at regular intervals for 7 days following the initial exposure to SWCNTs. The images were subsequently processed to illustrate any differences in NIR fluorescence intensity as a function of cell type and DNA sequence. In this ongoing work, we are finding significant differences between the cell lines and DNA sequences at various timepoints after an initial SWCNT exposure. The results of this study have implications for the long-term imaging of SWCNTs in preclinical and clinical applications.

This work was supported by RI-INBRE

Furthering the Understanding of Gene Regulation, Chromatin Expression and Structure

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Chromatin (DNA complexed with protein) is purposefully organized in the nucleus of every cell. In general, chromatin exists in a mixture of two states: heterochromatin, where DNA is condensed and genes are mostly not expressed and euchromatin where DNA is more open and genes are mostly expressed. The genome is divided into chromosomes and each chromosome is further subdivided into discrete domains of chromatin interaction separated by linear DNA segments termed topologically associated domains (TADs). The organization of chromatin within TADs is essential for “normal” cell function and gene expression, in fact, mutations in the genetic code which alter chromatin organization often lead to a deregulation in gene expression. This makes understanding the mechanisms that cause deregulation in chromatin formation and gene regulation important. In particular, understanding of the mechanisms that control gene regulation will allow us a better understanding as to why certain genes are expressed and when. Using the zebrafish hox clusters as a model we identified regions of the hox genes that interact with circular chromosome conformation capture (4C). Hox genes are a family of evolutionarily conserved transcriptional activators important for metazoan growth, body plan development, and other regulatory processes in the cell. This model organism was also selected partially due to its orthologous genetic connection to humans. This study looked specifically at the connections made within the hox aa cluster. The aa hox genes are important in processes that define the first vertebra, development of the hindbrain and central nervous system. Libraries created using 4C will be sequenced to identify the points of connection to gain insight into the regulatory regions important for correct cluster organization.

This work was supported by RI-INBRE

Understanding Chromatin Organization and Gene Expression with Circular Chromosome Conformation Capture

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Chromatin (the complex of DNA and protein) organization is an important component of the transcription process. In particular, gene expression is often contingent upon whether the chromatin is open, exposed to the transcriptional machinery, or closed, hidden from these factors. Each chromosome is subdivided into domains of intimate chromatin interactions (loops) called topologically associated domains (TADs). The organization of chromatin within TADs is essential for "normal" gene expression within the cell. In fact, changes in the chromatin structure, such as alteration in looping, causes gene expression to be impacted heavily. This indicates that understanding chromatin organization can lead to the ability to better understand gene expression. Mutations in chromatin organization can lead to the incorrect cellular functions, or lack of crucial cellular functions. Using zebrafish as a model, we explored how chromatin organization regulates certain Hox genes. Recently discovered in various forms of cancer, Hox genes are crucial for the development of appendages such as the tail of zebrafish. Using circular chromatin conformation capture (4C), we are able to specifically extract and identify parts of looped chromatin. With this extraction, we are then able to identify specific loops of chromatin and analyze their individualized functions. Specifically, Hox gene b5b has been shown to aid in the development of vagal neural crest cells in zebrafish.

This work was supported by RI-INBRE

The Chromatin Organization Role in Gene Regulation

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Chromatin (a complex of DNA and protein) has a 3D organization structure inside the nucleus and is essential for gene regulation. Structurally each chromosome contains discrete domains termed Topologically Associating Domains (TADs). TADs are chromatin loops that are either loosely coiled (open) correlating with active gene expression or tightly coiled (closed) correlating with inactive genes. This organization within a TAD is essential for normal gene expression within a cell with changes in the chromatin structure observed in disease states such as cancer. The goal of our research is to better understand the mechanism that regulates chromatin organization. By understanding how loops are formed and maintained within TADs, we could be provided with more insight into how chromatin regulates genes.

As a model for our research, we used Zebrafish hox genes which are closely related to human HOX genes and are controlled by chromatin organization. Interestingly, HOX genes have recently been found in various cancers, though the role of HOX in carcinogenesis is unclear. Zebrafish possess seven HOX clusters, one of these the HOX ca cluster, is responsible for mutants related to the extension of lateral bones in some types of Zebrafish. Using circular chromosome conformation capture (4C) we collected 4C libraries of the chromatin connections made by hox genes. These libraries will then be sequenced using Illumina sequencing to identify genomic connections.

This work was supported by RI-INBRE

Investigating the Role of arhgap1 in Melanocyte Development in *Danio rerio*

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Melanoma is a rare but deadly form of skin cancer that can spread rapidly if untreated. Melanocytes, the cells that give rise to melanoma, originate in the embryonic neural crest. To leave their embryonic origin, neural crest cells (NCCs) undergo an epithelial-to-mesenchymal transition (EMT), in which changes in gene expression alter adhesion and increase motility. Paracrine signals and the local tissue environment guide NCCs to locations throughout the embryo. Malignant cells share many features with migratory NCCs, except that their behaviors are unregulated. Better understanding the mechanisms that regulate embryonic processes such as migration and differentiation may therefore provide new strategies to prevent tumor metastasis. To identify genes responsible for embryonic melanocyte migration, our lab utilized CRISPR/Cas9 to knockout genes in zebrafish. One intriguing candidate identified was Rho GTPase Activating Protein 1 (arhgap1). At 3dpf, arhgap1 KO embryos had a significant decrease in melanophores. To further investigate the role of arghap1 in melanocyte development, two mutant alleles were isolated; a 12 base pair (bp) deletion that removes 4 amino acids and a 6bp deletion resulting in a premature stop codon. To establish stocks for each allele, PCR and restriction digests were used to genotype carriers. To manipulate arhgap1 expression during development, Gateway cloning was used to produce two arghap1 expression plasmids tagged with green fluorescent protein (GFP). Confirmation of plasmid localization in zebrafish embryos was observed using fluorescence microscopy. Resources established by this work will facilitate future studies of arhgap1's role in melanocyte development.

This work was supported by RI-INBRE

Cryptic role of alx4b in eye iridophore development

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Melanoma is a deadly form of skin cancer that is prone to metastasis. Melanocytes, the cells responsible for melanoma, originate in the embryonic neural crest, a multipotent population of stem cells that gives rise to many adult cell types including smooth muscle, peripheral nerves, craniofacial bone, and pigment cells. As the embryo grows, the fates of these stem cells are progressively restricted, eventually resulting in differentiation as a specific cell type. Zebrafish (*Danio rerio*) are a model organism for studying cell specification and differentiation. They have three neural crest derived pigment cells, melanocytes, iridophores, and xanthophores. Previous studies suggest that melanocytes and iridophores share a bipotent precursor cell, but the mechanisms that promote one fate over the other are not well known. To better understand the mechanisms of pigment cell lineage restriction, our lab isolated loss of function alleles for homeobox transcription factors alx4a and alx4b, known to be expressed by differentiated iridophores, but not melanocytes. We found that alx4a mutants do not develop body iridophores whereas alx4b mutants were indistinguishable from wildtype, suggesting that alx4a but not alx4b is required for iridophore development. Interestingly, while alx4a mutants lacked all body iridophores, they still developed iridophores in their eyes. To test the hypothesis that alx4b can compensate for the loss of alx4a during eye iridophore development, we generated double mutants and quantified eye iridophore phenotypes. Understanding the role of alx4a and alx4b in iridophore development provides further insight on the gene regulatory network governing iridophore specification.

This work was supported by RI-INBRE

Temporal characterization of ptk2bb expression and its potential interaction with the kita signaling pathway during zebrafish melanocyte development

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Of the three main types of skin cancer, melanoma accounts for the smallest percentage of diagnoses but the majority of deaths. Melanocytes, pigment cells that develop into melanoma, are derived from neural crest cells during embryonic development. Malignant melanoma cells utilize similar signaling pathways as zebrafish embryonic melanocytes. During normal development, KIT receptor tyrosine kinase pathway promotes melanocyte differentiation, migration, and survival. KIT mutations are found in many mammalian cancer types including melanoma. Despite the abundance of studies on KIT function, the intracellular mechanisms that promote specific melanocyte behavior in response to KIT signaling are not well understood. As part of an ongoing CRIPSR/Cas 9 screen, our lab identified protein tyrosine kinase 2 beta, b as a potential downstream target of KIT signaling. Knock-out of zebrafish ptk2bb, which encodes a cytoplasmic protein tyrosine kinase, showed a phenotype reminiscent of kita loss of function mutants in which dying melanocytes are extruded from the skin. To further characterize the relationship between KIT signaling and ptk2bb, we used CRIPSR/Cas 9 to knock-out ptk2bb expression in wildtype and heterozygous kita embryos. To determine when and where ptk2bb is expressed during key stages of zebrafish embryonic development, in situ hybridization and reverse-transcriptase PCR were performed. Preliminary results suggest that ptk2bb is expressed early in development but at low levels. Characterization of ptk2bb expression during embryonic development is a vital milestone in understanding its role in melanocyte development and may lead to the development of novel gene therapies for cancer patients.

This work was supported by RI-INBRE

Sex Specific Differences in Behavior and Metabolism Due to Chronic Obesogenic Diet Exposure in Sprague Dawley Rodents

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Increased worldwide prevalence of obesity, insulin resistance and related metabolic complications has created a need for new possibilities to combat obesity. Diet-induced obesity resulting from overconsumption of calorie dense and highly palatable foods coupled with diminished energy expenditure is a major health risk factor for metabolic syndrome, decreasing life expectancy and burdening the economy. Studies show that chronic consumption of an obesogenic diet can disrupt the negative feedback system that regulates body-fat and metabolism. Notably, males and females develop metabolic syndrome and its comorbidities at different rates. These differences are attributed to innate differences in insulin sensitivity, as well as adipose, skeletal muscle and hepatic tissue physiology that confer male susceptibility towards metabolic dyshomeostasis, which is further altered by nutritional status. While most preclinical studies have investigated males regarding diet-induced metabolic dyshomeostasis, inherent sex differences occur that can mitigate or exacerbate these diseases. Accordingly, using a chronic obesogenic diet paradigm in Sprague Dawley rodents, we examined neurobehavioral and metabolic sex differences to test the hypothesis that males have inherent deficiencies in endocrine glucoregulatory mechanisms in tandem with diet-induced defects in anxiety and hedonic pathways, which together instigate peripheral insulin resistance, glucose dyshomeostasis, and promotes weight gain and adiposity. Overall, our results show that chronic obesogenic diet feeding caused males to gain significantly more weight than control males, in part to hyperphagia, in tandem with sex-specific neurobehavioral changes modified by dietary status.

This work was supported by RI-INBRE

A laboratory assay for detecting green tide toxicity

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Macroalgal blooms are large aggregations of algae that can affect water quality and negatively impact marine life. Bloom-forming *Ulva* species have been shown to produce and release compounds that impact surrounding organisms. This research is applicable to coastal cities and towns throughout the United States that rely on beach tourism and local seafood which can be negatively impacted by macroalgal blooms. The goal of our research is to determine when macroalgal blooms, specifically green tides, are producing harmful compounds by developing a standard laboratory assay using phytoplankton. To determine if *Ulva* blooms impact the growth of phytoplankton, we exposed phytoplankton (*Dunaliella tertiolecta* & *Skeletonema costatum*) to: 1) water from a lab culture comprised of *Ulva lacinulata* and *U. compressa* at a concentration of 5 g L-1; and 2) water collected in the field from within an *Ulva* bloom. Treatment groups included *Ulva* bloom water and pasteurized bloom water (to eliminate impacts of marine viruses), along with a natural seawater control and an artificial seawater control. A spectrophotometer was used to measure the growth of phytoplankton via optical density and samples were fixed to quantify cell concentrations. Results showed that the laboratory cultured *Ulva* bloom water did not negatively impact the growth of *Dunaliella tertiolecta*, but rather, after 6-7 days *Dunaliella tertiolecta* cultures performed better when grown with *Ulva* bloom water than in the controls. Research using field-collected *Ulva* bloom water and a second phytoplankton species, *Skeletonema costatum*, is underway to compare to our laboratory culture results and will be presented.

This work was supported by RI-INBRE

Impacts of sea lettuce compounds on survival and growth of marine isopods

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There has been an increase in macroalgal blooms across Narragansett Bay and surrounding areas. Macroalgal blooms occur when there is intense growth in free floating macroalgae that is driven by an abundance of excess nutrients. Blooms can alter seawater chemistry and harm the organisms that reside in affected waters. These blooms include *Ulva*, a green macroalgae that releases compounds that have been linked to the inhibition of macroalgal growth, as well as mortality of larval oysters. Based on this previous evidence, we investigated the lethal and sublethal effects of compounds released from both *Ulva* blade and tube on *Idotea balthica*, a marine isopod. *Idotea balthica* is essential to the health of ecosystems and is an important prey species for larger invertebrates and fishes. Using divided co-culture mesocosms *I. balthica* was observed for changes in growth and survival over the course of 4 weeks while being exposed to *Ulva* (either 3.5 g/L or 5.0 g/L) or *Gracilaria* (either 3.5 g/L or 5.0 g/L), a red macroalgae that is not known to release inhibitory compounds. Separate trials were conducted for *Ulva* blade and tube. *Idotea balthica* length was measured weekly via photographs to track if *Ulva* inhibited growth and mortality was tracked daily. By comparing the start and end length of isopods after exposure to *Ulva* and *Gracilaria*, we concluded that *Ulva* had no significant effect on isopod growth and survival.

This work was supported by RI-INBRE

The impact of sea lettuce on grass shrimp growth

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Macroalgal blooms, large accumulations of detached seaweed, can be found in low wave energy environments through Narragansett Bay. Green tides are macroalgal blooms comprised of different forms of *Ulva*. Macroalgae serve as a form of habitat for invertebrates and a nutrient source for marine life. Harmful macroalgae blooms can affect the marine life that resides within the water columns and coastlines. These blooms alter seawater chemistry and potentially release harmful toxins that inhibit the growth and behavior of invertebrates. Bloom conditions were mimicked within mesocosm experiments to see the full effect of toxins released within *Ulva* blooms on the growth and behavior of grass shrimp. Each experiment was composed of 25 individual mesocosm that contained two grass shrimp and five different treatments. The mesocosms were 2-liter buckets with a mesh insert that divided the space to separate the grass shrimp and the algae. The treatments consisted of blade-forming or tubular *Ulva* (3.5g or 5.0g/L), *Gracilaria* (3.5 or 5.0g/L), and a mesocosm control that contained only grass shrimp. *Gracilaria* is a red macroalgae that is not known to produce inhibitory compounds; we conducted separate experiments with two species of blade-forming *Ulva* (*Ulva compressa* and *U lacinulata*) and tubular *Ulva*. We found that exposure to blade-forming *Ulva* had made no significant effect on the growth of grass shrimp; analysis of the results of the experiment with grass shrimp and tubular *Ulva* is ongoing and will also be presented.

This work was supported by RI-INBRE

Expression of Rhodothermus marinus bS21 and bS22 in *Thermus thermophilus*

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Ribosomes are responsible for the protein synthesis of every living cell. Many antibiotics interfere with protein synthesis to exert their antimicrobial effect. The thermophilic bacterium *Rhodothermus marinus* (Rma) is resistant to many 30S subunit antibiotics, while the phylogenetically unrelated thermophilic bacterium *Thermus thermophilus* (Tth) is sensitive to these. We aimed to determine if either of two ribosomal proteins (bS21 and bS22) from *R. marinus* would function in *T. thermophilus* which lacks homologs of either of these proteins. We will determine if the *R. marinus* proteins are incorporated into *T. thermophilus* ribosomes and test the hypothesis that resistance is related to the presence of bS21 or bS22.

This work was supported by RI-INBRE

Establishing a *C. elegans* Model for Fanconi Anemia Neurological Syndrome

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Fanconi Anemia (FA) is a rare genetic disease with a frequency of 1 in 160,000 live births. Patients with biallelic mutations in any of 23 FA genes display an increased risk for progressive bone marrow failure, and cancer, and a shortened lifespan. Recently, Fanconi Anemia Neurological Syndrome (FANS) has entered the landscape as a phenomenon of study. Clinical observations have described abnormalities in the brain tissue of FA patients, though little is known about the relationship between FA gene mutations and nervous system structure. Preliminary data from the Howlett Lab and others have shown that the FA gene FANCD2 interacts with nervous system development genes, suggesting a mechanism for altering nervous system structure in FA. In order to test the hypothesis that nervous system development is altered in FA, the Barbagallo-Howlett collaboration assessed nervous system structure and function in the previously characterized *C. elegans* model of FA. Behavioral analysis shows changes in movement patterns of FA worms, while motor neuron function is indistinguishable from wild type suggesting changes in neuronal function upstream of the motor neurons. Early chemotaxis analysis suggests differences in odorsensory neurons. Together, these experiments are beginning to outline the molecular mechanisms of FANS. The end goal of this project is to elucidate the mechanisms underlying nervous system dysfunction in FA, allowing for insight into neurological disease progression in human patients.

This work was supported by RI-INBRE

Retinaldehyde Genotoxicity and Processing by ALDH1A1 in Fanconi Anemia

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Fanconi Anemia (FA) is a rare genetic disorder characterized by developmental abnormalities and increased risk for bone marrow failure and cancer. There are currently 23 known FA genes. A major role for the FA proteins in the repair of DNA interstrand crosslinks (ICLs) has been established. Cells from FA patients exhibit increased ICL sensitivity and general genome instability overall. However, it has yet to be determined if the FA proteins play additional roles in the repair of other types of DNA damage. Recent RNA-seq analysis from our laboratory has established that retinoic acid metabolism is dysregulated in FA-D2 (FANCD2-/-) cells. For example, we observed increased levels of the RDH10 and ALDH1A1 genes in FA-D2 (FANCD2-/-) cells compared to FA-D2 cells complemented with wild-type FANCD2. During retinoic acid metabolism, the RDH10 alcohol dehydrogenase converts retinol to retinaldehyde. Retinaldehyde is then converted to retinoic acid by the ALDH1A1 aldehyde dehydrogenase. Retinoic acid is transported to the nucleus where it binds to the RAR/RXR nuclear receptors that regulate the transcriptional activation or repression of many genes. Much like other aldehydes produced during metabolism, retinaldehyde is a potent genotoxin. We hypothesize that altered retinoic acid metabolism in FA cells may be a strategy to mitigate the genotoxic effects of retinaldehyde. To test this hypothesis, we exposed FA-A (FANCA-/-) patient cells and FANCA-complemented FA-A cells to retinaldehyde and analyzed levels of several DNA damage markers following exposure. Our results demonstrate that retinaldehyde exposure leads to increased DNA double-strand breaks (DSBs) and that the repair of these DSBs may be attenuated in FA patient cells. In this study, we have also examined the subcellular localization of ALDH1A1 in FA-D2 (FANCD2-/-) cells. Using both immunofluorescence microscopy (IF) and subcellular immunoblotting, we have determined that ALDH1A1 localizes primarily to the cytoplasm and that its subcellular localization is not altered upon treatment with retinaldehyde.

This work was supported by RI-INBRE

Evaluation of Aminoguanidine's Methylglyoxal Trapping Activity and Cytoprotective Effects Against Methylglyoxal Induced Cell Death

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Methylglyoxal (MGO) is a reactive carbonyl species that mediates the formation of advanced glycation endproducts (AGEs) via the Maillard reaction with protein and reducing sugars. As the precursor of AGEs, MGO can induce oxidative stress in human cells, which may lead to cell death. Although inhibitors of AGEs, such as aminoguanidine, are reported to show cytoprotective effects, aminoguanidine's cytoprotective effect against MGO induced cytotoxicity in human skin cells is still unclear. Therefore, we aim to validate a high-performance liquid chromatograph (HPLC) based method to evaluate aminoguanidine's MGO trapping activity. In addition, we characterized MGO induced cytotoxicity in human keratinocyte HaCaT cells, which can be used as a model to screen AGEs inhibitors for cytoprotective effects. The HPLC method for the MGO trapping assay was developed by measuring the peak areas of three MGO trapping reaction chemicals including 1,2-phenylenediamine (as the reactant), MGO derivation (as the product), and 2,3-dimethylquinoxaline (as the internal reaction standard). With this validated HPLC method, we showed that aminoguanidine had promising MGO trapping activity by decrease the MGO content by 40%. In addition, MGO (at 400 μM) induced significant cytotoxicity by reducing the cell viability of HaCaT by 69.7%, as compared to the control group. The future study of this project will be focus on the evaluation of aminoguanidine's cytoprotective effects against MGO induced toxicity in HaCaT cells.

This work was supported by RI-INBRE

Evaluation of the Inhibitory Effects of Phytocannabinoids on Elastase Activity

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Elastase, a serine-type protease expressed in the basal layers of the epidermis, breaks down the tissue connecting protein elastin. Elastin accounts for up to 4% of the total protein content of the skin tissue, which is crucial for maintaining skin's mechanical structures. Declined elastin level in skin tissue is linked to the loss of skin structural integrity and consequently result in the formation of skin wrinkle. Reported studies have shown that natural products from plants with anti-elastase effects may exert skin protective effects. Although published studies support the skin beneficial effects of phytocannabinoids from Cannabis sativa, it is unknown whether phytocannabinoids can inhibit the activity of elastase. Herein, we aim to evaluate a panel of phytocannabinoids including cannabidiol, cannabidiolic acid, cannabinol, and cannabigerol for their anti-elastase activities using in vitro and in silico assays. Data from the elastase enzyme inhibition assay showed that cannabidiol, cannabidiolic acid, cannabinol, and cannabigerol had a moderate anti-elastase activity with an IC50 value of 551.2, 295.0, 358.0, and 347.3 μM, respectively. In addition, the molecule docking method was used to explore the molecular interactions between test compounds and elastase protein. Findings from the current study suggest that some phytocannabinoids can inhibit the activity of elastase enzymes but further studies using cell-based models are warranted to confirm their effectiveness.

This work was supported by RI-INBRE

Development of a Thermoresponsive Photoink for 3D Printing/Bioprinting Applications

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Previous work in our lab has successfully explored poly(N-isopropylacrylamide) (PNIPAm; a thermoresponsive polymer) polymer-based microwell arrays developed using chemical crosslinking methods, for tumor spheroid culture. To the best of our knowledge, there are no commercially available thermoresponsive photoinks that are compatible with digital light printing (DLP) to provide a suitable, cytocompatible surface for the long-term culture of viable 3D cancer spheroids. PNIPAm experiences reversible changes in its physical properties once its lower critical solution temperature (LCST) is exceeded; like gel/well shrinking and switching from hydrophilic to hydrophobic in nature. This creates a microwell array that encourages cells to cluster and form spheroids within the cell culture incubator at 37^{o}C, and once removed from the incubator the gel/wells expand allowing the spheroids to be harvested easily. Early formulations, developed by us using Cellink's LumenX+ DLP printer, experienced bleaching, curling and layer peeling, but showed thermoresponsive properties including changes to hydrophobicity (as measured by contact angle testing) and a clearly defined LCST. In this work, we attempted to address the bleaching and other issues through the use of two full factorial optimization experiments. High concentrations of N,N'-Methylenebisacrylamide (Bis) were identified as a factor in bleaching. The gels behaved significantly differently to earlier chemically crosslinked microwell arrays. Formulations with comparatively higher LCSTs above 35^{o}C performed better and retained their structure in commonly used buffers and cell culture media. A549 cancer cells were successfully cultured as 3D cell spheroids on preliminarily optimized formulations.

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Development of 3D printed mini-tablets for pediatric delivery

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Despite tremendous strides in the field of advanced drug delivery systems over the past decade, there remains a lack of age-appropriate drug product commercially available for pediatrics. It is not uncommon for adult drug products are extemporaneously compounded into child-friendly dosage forms such as oral suspensions for ease-of-swallowing. However, this can lead to inaccuracies in dosing, thus posing major health risks to pediatric patients. To overcome limitations inherent to traditional batch manufacturing methods, additive manufacturing (AM), an emerging technology in the pharmaceutical industry, has been used to produce personalized medications built in a layer-by-layer fashion. One type of AM, 3D printing, has revolutionized the way in which pharmaceutical researchers approach developing novel drug delivery formulations. We hypothesize that developing mini-tablets with smaller dimensions than traditional formulations can help overcome these pediatric drug delivery obstacles, thereby leading to improved medication adherence and therapeutic outcomes. Mini-tablets were designed on Solidworks to have an immediate release outer layer and a sustained release inner core to achieve a dual release platform of a model compound dexamethasone. Preliminary in vitro dissolution studies of the drug-loaded solvent casted materials demonstrating showed greater than 80% drug release in 30 mins from the outside layer (immediate release layer) and greater than 80% drug release in 24 hours from the inner core (sustained release layer). A custom filament developed using the solvent casted materials will next be produced through hot melt extrusion and subsequently used to 3D print the final mini-tablet formulations.

This work was supported by RI-INBRE

Characterization and In Vitro Testing of Mannose-Modified BLZ Liposomes for Reprograming Tumor-Associated Macrophages

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Glioblastoma (GBM) is the most common and malignant primary brain tumor with an average overall survival of 15 to 21 months after first diagnosis and a 5-year survival rate of less than 5%. Currently, there is an urgent need for better therapeutic strategies against GBM. Glioma-associated microglia/macrophages play a pivotal role in tumor growth, cancer immunosuppression, and drug resistance, thus bleak prognosis.

The main objective of the present research was to develop a novel immunotherapeutic strategy based on BLZ-loaded mannose-modified liposomes to reprogram tumor-associated macrophages into tumor-inhibiting macrophages to block glioma progress. Mannose has been selected as the target ligand for tumor-associated macrophages. The effectiveness of BLZ-945 to reprogram cells, in order to assist in the body's immune response, was evaluated with antibody staining through flow cytometry. While the drug proved the ability to convert tumor-associated macrophages into different phenotypes, the repolarization capabilities of mannose-modified BLZ-Liposomes will be evaluated in future work.

This work was supported by RI-INBRE

Validation of an RNA-sequencing Pipeline by Comparison to a Published Study with Alternate Mapping, Transcript Assembly, and Visualization Methods

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For gene expression profiling, RNA sequencing is a valuable tool that comprehensively determines the transcriptome of a biological sample, the set of messenger RNA (mRNA) sequences that encode DNA's genetic information. The process involves reverse transcribing fragmented RNA into sequences of complementary DNA, which can be read by a machine upon modifications and subsequently be filtered, mapped to a reference genome, and assembled through various analysis approaches. This research project seeks to both create and ultimately validate an RNA-seq data pipeline, an algorithm whose output from one step becomes the input for the next. To do so, the Snakemake workflow was used to write a programming script for single-end RNA-seq data on the University of Rhode Island Andromeda cluster. To test the pipeline's accuracy via a case study, an RNA-seq dataset of six pancreatic stem cell samples was downloaded from the NCBI GEO database (GSE:136064). The samples' associated study used different software tools to map the reads, count the transcripts, and graph gene expression, and the samples were run through the pipeline before data visualization was performed in R. While there were missing gene annotations in the results as well as undisclosed parameters for the study's plots, the graphs that were generated, such as a volcano plot of differentially expressed genes, a PCA plot, and bar plots of specific genes, showed overall consistency with the study's corresponding graphs. This leads to the conclusion that additional analysis methods would improve the results but that the pipeline is nonetheless accurate.

This work was supported by RI-INBRE

Maternal exposure to PFOS alters functional development of the nervous system

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Over the last 40 years, the rates of neurodevelopmental disorders such as ADHD, autism, and epilepsy have become more prevalent, with 15% of children in the United States being affected. These disorders do not have a clear genetic cause, suggesting that environmental exposure could play a role in increased disease prevalence. PFOS is a common toxicant found in many non-stick products that is known to bioaccumulate in human tissues. Though the accumulation of PFOS in neural tissues is well documented, we know little about how it affects nervous system development. We hypothesize that maternal PFOS exposure alters nervous system development and function. We tested this hypothesis using the simple and genetically tractable nervous system of C. elegans as our model organism. C. elegans have a well-characterized series of behaviors that have been linked to the function of specific neuronal classes, allowing us to use behavior as an indirect readout of neuronal function. Behavioral analysis of three specific neuronal types, cholinergic (ACh), dopaminergic (DA), and GABAergic (GABA), show changes in body posture and movement within our model organism. This data suggests that there are impacts in the body posture as well as in the movement among the mutant and wildtype populations of C. elegans when exposed to different concentrations of PFOS, and later neuronal activity in progeny. These results indicate that maternal PFOS exposure alters the nervous system, supporting the need for further research examining the effect of environmental toxins on the function and structure of the nervous system.

This work was supported by RI-INBRE

Alpha-lipoic acid as a protective agent against PFOS-induced developmental defects in *Drosophila melanogaster*

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Perfluorooctanesulphonic acid (PFOS) is a highly toxic chemical known to present significant risk to human health. As a persistent organic contaminant, PFOS has been widely detected in the environment, wildlife, surface water, where it often accumulates. The high bioaccumulation, negligible elimination, and high toxicity of PFOS allow it to instill detrimental effects on human health, remaining undigested in humans for over 5.4 years. Therefore, there is great need to establish a potential therapeutic that can outcompete its harmful repercussions. Our study proposes alpha-lipoic acid (ALA) as a dietary supplement that could outcompete environmental PFOS absorption lowering risks of chronic metabolic disease. ALA is a structural analog of PFOS due to its similar eight-carbon backbone that could outcompete the chemical and reduce its likelihood of absorbance. To identify which concentrations of ALA were most optimal in reversing the effects of PFOS, we exposed *Drosophila melanogaster* to four ALA concentrations, 0.022%, 0.045%, 0.090%, and 0.18% in combination with two PFOS concentrations, 0.05 μM and 0.1 μM from the moment of their conception over the course of their two-week long lifecycle. Our developmental results showed no viability in the higher PFOS and ALA concentrations and no difference in terms of days to reach adulthood in the lower ALA concentrations even in combination with PFOS. However, despite reaching full adulthood the PFOS exposed F1 progeny exhibited lower survival rates and appeared feebler than those exposed to ALA only. We will continue to expand on these results by measuring their triglycerides, cholesterol, and glucose concentrations which are vital for proper metabolic function.

This work was supported by RI-INBRE

Maternal exposure to PFOS alters structural development of the nervous system

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In the past decade, ADHD prevalence has risen by over 1% and autism spectrum disorders have more than doubled. These disorders show no clear genetic link suggesting that the environment may be affecting the increase in these disorders among the population. PFOS is an environmental toxicant that can be found in food packaging materials, firefighting foam, and nonstick cookware, with measurable levels of PFOS, detected in 98% of human serum samples. Animal studies show that PFOS is able to cross the blood-brain barrier and accumulate in the brain tissues of adults and children. We hypothesize that maternal stores of PFOS are passed to progeny, altering nervous system development. We tested this hypothesis using the simple nervous system of *C. elegans* as a model for assessing changes in nervous system structure in the progeny of PFOS-exposed mothers. We assessed the structure and number of cholinergic, GABAergic, and dopaminergic neurons in the progeny of mothers exposed to 10uM, 20uM, or 40uM PFOS immediately prior to fertilization. These experiments reveal changes in the adult nervous system, but not when these changes occur, setting the stage for future experiments in which I will evaluate nervous system structure during each developmental stage to better characterize changes in gross neuronal structure. Taken together, these studies will allow further research examining the effect of environmental toxins on the structure and morphology of the nervous system.

This work was supported by RI-INBRE

The role of alpha-lipoic in mitigating the developmental effects of perfluorooctanesulfonic acid in *Drosophila melanogaster*

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The prevalence of metabolic diseases in humans is an epidemic, with around 37 million reported cases of obesity and diabetes in recent decades, where 85.2% of those with diabetes were also obese. Although nutritional intake, genetics and lifestyle are known factors that contribute to metabolic syndromes, the exact etiology is still unknown. This suggests that environmental toxins may be a contributing factor to the increase in cases. Perfluorooctanesulfonic acid (PFOS) is an environmental toxin found in a myriad of consumer products and is present in 98% of human serum samples. It is characterized by its high bioaccumulation, long half-life, and negligible excretion (Chen et al. 2014). PFOS is present in humans, but the impact of chronic exposure is not well understood. Using *Drosophila melanogaster* as a model system to better understand the impact of chronic PFOS exposure, embryonic development was tracked and biochemical assays and RT-qPCR were conducted. This research aimed to unveil the role of the dietary supplement, alpha-lipoic acid (ALA), in diminishing the risk of metabolic disease from preconception exposure to PFOS. Overall, we observed that the progeny of animals exposed to 0.05uM and 0.1uM PFOS had observable morphological abnormalities and females contained less triglyceride content than the control group. Groups exposed to 0.022% ALA with 0.05uM PFOS showed promising results in combating the negative effects of PFOS exposure on a developing embryo as adults were well developed compared to those exposed solely to 0.05uM PFOS. We plan to expand on this work by understanding how *Drosophila* insulin-like peptide expression is altered as a result of PFOS exposure. This is an essential next step in determining a strategy to prevent PFOS uptake into maternal oocytes and diminish the prevalence of metabolic diseases.

This work was supported by RI-INBRE

Characterizing motor, sensory, and cognitive functions in a rabbit model of cerebral palsy

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Cerebral Palsy (CP) is the most common motor disability in children. Previously studied rodent models of CP found that subjects could have an extensive injury to the CNS without prominent motor deficits, indicating that this model is not comparable to CP in humans (Cavarsan et al., 2019). This study aimed to determine the effect of prenatal hypoxia-ischemia (HI) on tactile sensitivity and motor function in neonatal rabbits to explore the effectiveness of a rabbit model of CP. Fetal HI in embryonic day 25 pregnant dams was induced via balloon catheterization at the bifurcation of the descending aorta, occluding blood flow to the lower body, including the uterine horns, for 40 minutes. Von-Frey filament test, Hargreaves' test, and Open-Field Assays were performed at P11 and P18 to evaluate tactile sensitivity, general motor ability, and fear/anxiety. Our results show HI kits have a decreased withdrawal latency in the Hargreave's Test at P11 compared to sham kits. In HI kits, the Von-Frey Filament Test showed a decrease in the paw withdrawal threshold at both P11 and P18 compared to sham kits. In the Open-Field Assay, there were no significant differences in the time HI kits spent in the center of the field compared to Sham kits at both time points. These findings suggest HI kits exhibit increased sensitivity to mechanical and thermal sensory stimuli. This is not accompanied by alteration in fear/anxiety behavior.

This work was supported by RI-INBRE

Investigation of Protein Biomarkers for Cerebral Amyloid Angiopathy In Different Disease Stages of the rTg-DI Rat Model

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Cerebral Amyloid Angiopathy (CAA) is a common cerebral small vessel disease of older adults and prevalent comorbidity of Alzheimer's disease (AD). CAA is characterized by progressive deposition of amyloid beta-peptide in the cerebral vasculature leading to cerebral microhemorrhaging, stroke, perivascular neuroinflammation, white matter damage, and severe cognitive impairment. Despite hallmark imaging characteristics of CAA, definitive diagnosis requires post-mortem confirmation, and no validated biomarkers have been established. The underlying mechanisms leading to CAA-related vasculopathies are poorly understood and currently have no effective treatment. Thus a better understanding of potential protein biomarkers of CAA, and underlying molecular mechanisms of disease progression, is vital to developing novel treatment strategies and diagnostic tools. In this study, we investigated the longitudinal expression of proteins identified as potential markers of CAA in the established rTg-DI preclinical model of CAA via protein mass spectrometry and immunoblotting. In both emergent and advanced CAA stages, the elevated expression of ANXA3, HTRA1, and APOE proteins was revealed in rTg-DI brains. Immunolabeling revealed strong colocalization between APOE and amyloid deposits in rTg-DI rat brain regions, while strong enhancement of ANXA3 and HTRA1 were associated with areas of amyloid deposition and severe vasculopathies. Hence, we explore important findings regarding these three potential markers of CAA in rTg-DI rats.

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Lessons from Community Engagement Activities for Cancer Survivorship Study & Intersectional Identities that Influence the Cancer Survivorship Experience

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There are several factors including the aging of the population, an increase in population size, and improved public health practice such as preventive screening leading to early detection, coupled with strong medical cancer care services that have resulted in a growing number of cancer survivors across the United States. However, disadvantaged groups referred by some as minority groups such as Black and Latinx populations, have worse cancer survivorship experiences compared to White populations. Previous research has demonstrated that Black and Latinx cancer survivors experience disproportionate negative effects of cancer, which are fueled by physical, emotional, social, and financial challenges. Thus, this highlights the fact that there is racial and ethnic disparities in cancer survivorship in the country. This study examined the role that sociocultural factors play in perpetuating the difference in cancer survivorship rates among Black/Latinx populations and Whites. Eight cancer survivors who identified as Black or Latinx were purposively recruited and interviewed using semi-structured interview approach. From the interviews completed and analyzed thus far, the following themes were identified: 1) Invisibility of survivors; 2) Understanding the needs of cancer survivors; and 3) Issues of empowerment. The findings may help inform policies for culturally relevant cancer survivorship interventions.

This work was supported by RI-INBRE

Lessons from Community Engagement Activities for Cancer Survivorship Study

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Cancer survivorship research has been expanding in the last few years. However, many of these cancer survivorship studies have not explored the complexities in the cancer survivorship experience of Blacks and Latinx people, hence, our study. There exists low participation of racial and ethnic minorities in medical research and mistrust between underrepresented communities and researchers. Part of our goal for this cancer research is to develop relationships with the community so that we may facilitate recruitment, identify relevant issues about cancer survivorship, and plan potential interventions. To develop these relationships, we partnered with "gateway consultants" including cancer support leaders and other local community leaders. We also found attended local community activities hosted by cancer survivors, to engage and actively listen, and share more about our research objectives. This helped build trust with the participants as we made sure to share our research in an environment where the participants feel safe and can share their experiences freely and without judgment. As a researcher, you must acknowledge and understand that you may be in a position of greater privilege, and understanding the implications of such power dynamics is critical in successfully engaging with a disadvantaged community/group. This part of our study will highlight some of the initial lessons learned from conducting a community-based participatory approach to cancer survivorship research in Rhode Island.

This work was supported by RI-INBRE

Investigating Metal Binding to KmtR Mutants

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Mycobacterium tuberculosis (*M. tuberculosis*) is the bacteria that causes tuberculosis. This bacteria kills about 2 million people per year, which makes it one of the leading causes of death per year. There is an increase in number of drug resistant strains of *M. tuberculosis*, and therefore targeting other pathways to kill these bacteria is important. The bacteria contain several metal transport systems which are necessary for its survival. The metalloregulator KmtR is one of two nickel and cobalt regulators. The focus of this research is to determine the different residues that are responsible for binding of cognate nickel and cobalt and noncognate zinc metals. The E101Q mutant was expressed and purified, and metal binding studies are being pursued.

This work was supported by RI-INBRE

Examining the Metal Binding Affinities of KmtR Mutants

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It is estimated that one-third of the world's population is latently infected with *Mycobacterium tuberculosis*, resulting in more than 9 million new cases and 2 million deaths each year. The survival of *M. tuberculosis* in its host can be attributed to its several metal transport systems. KmtR is the second metalloregulator in this bacterium that is associated with Ni (II) and Co (II) export. Metal binding studies are being conducted to identify the protein residues that bind to the cognate metals Ni (II) and Co (II) and the noncognate metal Zn (II). The H102Q mutant, among others, was expressed and purified.

This work was supported by RI-INBRE

Determining the Effect of KmtR Mutant, H111Q, on Metal Binding Affinites

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Mycobacterium tuberculosis is the second leading infectious killer after COVID-19. The bacteria utilizes several metal transport systems to help it survive in the host. The bacteria has two metalloregulators that are associated with nickel and cobalt export, NmtR and KmtR. The focus of this research is on KmtR. KmtR represses the expression of the genes, cdf (which encodes the export protein) and kmtR. The goal of our research is to identify the residues that are responsible for binding the cognate metals, nickel and cobalt, as well as the noncognate metal, zinc, to KmtR. Mutagenesis studies coupled with metal binding experiments will be used to determine how KmtR binds these metals. The H111Q mutant, among others, has been made, expressed, and purified in our lab. Data from our experiments involving this KmtR mutant protein will be discussed.

This work was supported by RI-INBRE

Analysis of Biofilm Formation of Enterohemorrhagic *E. coli* (EHEC) 0157:H7 Mutants on Biotic and Abiotic Surfaces

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Escherichia coli (*E.coli*) are Gram-negative, rod-shaped, facultative anaerobic bacteria, and cause about 300-500 million infections and 200,000 deaths per year. *E. coli* is transmitted to humans through the consumption of cattle and produce products, as well as through environmental exposure. Most of this environmental burden is due to a phenomenon called super-shedding (SS), in which the Enterohemorrhagic *E. coli* (EHEC) pathovar, O157:H7, has a uniquely high production of biofilms, a stronger adherence to produce, and a unique RSE phenotype. To investigate the increased biofilm formation by super-shedders, we are analyzing the formation of biofilms of both super-shedder and non-super-shedder isolates on biotic and abiotic surfaces, such as glass, plastic, and spinach green lysate. These data are critical because they would allow for the identification of the drivers for increased biofilm formation in super-shedders, and which proteins or genes are essential for biofilm formation in both super-shedders and non-super-shedders. Future work will focus on studying the transcriptional change in colonic epithelial cells in relationship to the changing interphase with EHEC during stages of adherence. Bioinformatics will be used to compare adhesins of interest to determine how conserved they are in gene presence and percent identification in other *E. coli* and other bacterial species. Additionally, RNA-Seq software will allow for the comparison of transcriptome profiles over time. This could further the understanding of the various aspects of the interface between EHEC and its host and to potentially help define a therapeutic target for the interaction between pathogen and host.

This work was supported by RI-INBRE

Analyzing the Mechanisms of Unique Adherence Phenotypes of Super Shedder *E. coli*

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Enterohemorrhagic *Escherichia coli* (EHEC) is a pathovar of *E. coli* that produces Shiga toxin that causes damage to the intestinal wall lining. This is commonly caused by consuming undercooked or contaminated meat, but an infection can also be caused by consuming contaminated water or milk, working with cattle, or even person to person. Symptoms of an EHEC infection include abdominal cramps, diarrhea, and nausea. Super shedding (SS) is a phenomenon that has been reported in some cattle, (SS) *E. coli* produces more biofilm than non-(SS) *E. coli*. EHEC attaches to the inner intestinal epithelial cells, in this study the goal is to analyse the ability of (SS) and non-(SS) to produce biofilm and determine the genes responsible for the adherence pattern. Firstly, an adherence timeline is to be determined to know the timeframe in which the EHEC cells adhere to the epithelial cells, and how long each adherence phase needs. The four attachment phases between the cells are searching, initial attachment, intimate attachment, and biofilm formation. When the timeframe for biofilm formation is known, RNA extraction and isolation is used to study which genes are possible adhesins. This is done through comparing the different regulation patterns of genes at the different adherence phases. Growth curves of two strains; SS17 (a representative SS isolate) and EDL933 (a non-SS isolate) were done and the results were plotted. The results of these studies have potential to help develop better therapeutics against EHEC and possibly other enteric bacteria.

This work was supported by RI-INBRE

Analyzing the Mechanisms of Unique Adherence Phenotypes of EHEC Mutants

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Enterohemorrhagic *Escherichia coli* (EHEC) is a pathovar of *E. coli* that is one of the leading causes for human foodborne illness and a leading factor in the global food burden. This pathovar directly affects human epithelial cells in the gastrointestinal tract. EHEC produces a number of virulence factors including Shiga toxin, which causes significant damage to blood vessels, resulting in subsequent damage to the intestinal wall and other organs, causing the symptoms of this infection which include nausea, fatigue and bloody diarrhea. EHEC can be transmitted through cattle, produce and other environmental exposures. The majority of environmental burden resulting from *E.coli* is caused by the super shedder isolates. This isolate has unique phenotypes that result in higher production rates of biofilms and stronger adherence to hosts, such as produce and human epithelial cells of the small intestine. To investigate the increased biofilm formation by the super shedder isolate, biofilm production was analyzed on both biotic and abiotic surfaces, including plastic, glass, and spinach lysate, to better understand its adherence to the vehicles in which EHEC causes human infection and transmission. Future work will focus on studying the transcriptional change of colonic epithelial cells in response to the changing interface of EHEC over an adherence timeline to Caco-2 cells. This will involve RNA extraction and sequencing of EHEC cells at five different adherence steps, searching, initial attachment, pedestal formation, micro-colony aggregation and biofilm formation. This is important in understanding the interactions between EHEC and its hosts to develop better therapeutic approaches.

This work was supported by RI-INBRE

Bacterial Shape and Structure: Cloning and Purification of the Cell Wall Synthesis Proteins PBP1b and LpoB

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Bacterial cells grow and reproduce through coordination of many proteins collectively known as the cell division machinery. This machinery facilitates the division and physical separation of one cell into two identical progeny cells. In *Escherichia coli*, the cytoskeletal protein FtsZ promotes the formation of a division septum at the cell center and assembles into a large dynamic protein structure called the Z-ring. Individual FtsZ proteins in the Z-ring form large linear polymers that assemble in a head to tail arrangement. Protein interactions between FtsZ in the Z-ring and additional cell division proteins direct peptidoglycan synthesis in the cell wall and facilitate septation of the cell. FtsN, which may trigger this septation event, is a bitopic membrane protein with a small cytoplasmic domain and large periplasmic domain containing a peptidoglycan-binding SPOR domain. FtsN interacts with other cell division proteins on both sides of the cytoplasmic membrane. Specifically, FtsN interacts with the FtsQBL complex in the periplasm and may help to recruit peptidoglycan synthetases such as FtsI and PBP1b. Recently, it was discovered that FtsN interacts with PBP1b to activate its transglycosylase activity. Thus, we investigated how FtsN interacts with PBP1b and an activator of PBP1b, called LpoB. We cloned PBP 1b and LpoB into expression vectors and then purified them by immobilized metal affinity chromatography. We are currently optimizing biochemical assays to monitor protein complex formation by FtsN, PBP1b, and LpoB.

This work was supported by RI-INBRE

Eradication of an established *Staphylococcus aureus* biofilm with synergistic combination of an anti-biofilm and an antibiotic agent

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BACKGROUND: Biofilms are intrinsically resistant communities surrounded by a protective extracellular polymeric substance (EPS) causing high recurrent infections.1,2 We aim to combine biofilm destabilizing agents, cellulase, and ascorbic acid with vancomycin or daptomycin in a novel strategy to eradicate established *Staphylococcus aureus* biofilms.1,3

METHODS: Four unique MRSA biofilms of varying strengths/stabilities were grown in tryptic soy broth with 1% dextrose, 12.5mg/mL magnesium, 25mg/mL calcium, and 6 log10CFU/mL bacterial inoculum. Biofilms were grown 20hrs in 96-well tissue culture treated polystyrene plates then gently rinsed with sterile water before 24hr lock treatment with anti-biofilm agents or antibiotics, monotherapy or in combination. Subsequently, plates were rinsed and dried overnight to fix biofilms to the well surface. Biofilms were stained with 0.1% crystal violet (CV) for 15 minutes and then rinsed before glacial acetic acid (33%) was added to resolubilize the remaining CV. BioTek plate reader at 570nm read the optical density of the remaining CV. Eradication was defined as readings of ≤0.09 optical density.

RESULTS: Cellulase minimum biofilm eradication concentration (MBEC) was 1.5-25mg/mL, while ascorbic-acid, vancomycin and daptomycin MBECs were greater than tested concentrations. Daptomycin with 2.5% cellulase lock caused an 76% biofilm reduction. Pre-locking with 2.5% cellulase lock for 4 hours before 5mg/mL daptomycin lock caused a 86% biofilm reduction. Ascorbic acid and vancomycin did not surpass cellulase or daptomycin therapies.

CONCLUSION: Anti-biofilm pre-locking before antibiotic was more effective than exposing both agents simultaneously. Daptomycin (5mg/mL) with 2.5% cellulase was the most effective biofilm eradication. Vancomycin (5mg/mL) with 2.5% ascorbic acid was the least effective

KEYWORDS: S. aureus, biofilm, antibiotic, MRSA, resistance

FUTURE WORK: The same quantitative assay will be performed with multiple *S. aureus* isolates to validate our findings.

This work was supported by RI-INBRE

Time is on My Side: Evaluating Genetic and Chemical Inactivation of Bacterial Autolysins

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Peptidoglycan (PG) is the major structural polymer in most bacterial cell walls. It helps to protect from environmental stress and preserve cell morphology throughout their life cycle. Additionally, the biosynthesis of peptidoglycan is an important regulator of bacterial cell growth and division. The backbone of PG is composed of alternating N- acetylglucosamine (GlcNAc) and N-acetylmuramic acid (MurNAc) linked via a β-(1,4)-glycosidic linkage. Adjacent glycan strands are cross-linked via a pentapeptide attached to MurNAc to form a 3-dimensional mesh. Bacterial autolysins are a broad class of enzymes that break down PG and are required for cell growth and separation of daughter cells following cell division. These autolysins are classified based on which bond they cleave in PG. Our group has developed a chemical probe, masarimycin, an inhibitor of the exo-acting N-acetylglucosaminidase (GlcNAcase) LytG and a bacteriostatic inhibitor of *Bacillus subtilis* growth. Using *B. subtilis* as a model organism, we are investigating the changes in autolysin expression under genetic and chemical inactivation. We hypothesize the chemical inactivation response will produce a unique molecular phenotype compared to genetic deletion. We have previously shown that deletion of the major autolysins LytC, LytD, and LytF resulted in increased sensitivity to masarimycin. To investigate this increased sensitivity, qPCR was employed to measure changes in autolysin gene expression in the presence of masarimycin. Preliminary results suggest that when LytC is deleted and the cells are exposed to masarimycin, a 50-100 fold reduction in expression of lytD, lytG, lytF is observed. Additionally, we have found that *B. subtilis* adapts to the deletion of LytC by up regulating the GlcNAcase LytD, an endo-acting GlcNAcase. In order to further investigate changes to PG metabolism a metabolic labeling strategy was established using the fluorescent amino acid, HADA, and monitored by fluorescent microscopy. There is evidence that when treated with HADA and masarimycin, there is less turnover of old material resulting in increased fluorescence and a thicker cell wall compared to wild-type cells. This was seen in both our fluorophore incorporation experiment and our PG-recycling experiment. Finally, the incorporation of HADA into PG appears to augment the expected morphology of the *B. subtilis* masarimycin induced morphology and creates ring-like structures.

This work was supported by the National Science Foundation

Analysis of *Bacillus subtilis* cell wall provides insight into masarimycin's mode-of-action.

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Bacillus subtilis is a Gram-positive rod-shaped bacterium that serves as a model organism for studying cell wall metabolism in rod-shaped bacteria. Peptidoglycan (PG) is the major structural polymer of the bacterial cell and is composed of alternating N-acetylglucosamine and N-acetylmuramic acid linked via a β-(1,4)-glycosidic linkage. Adjacent glycan strands are crosslinked via stem peptides attached to N-acetylmuramic acid. Autolysins are a broad class of bacterial enzymes that break down PG that are required for growth and division. Our lab has developed a chemical probe that inhibits the N-acetylglucosaminidase (GlcNAcase) LytG, the major active GlcNAcase of vegetative growth. To our knowledge, it is the only autolysin inhibitor that halts bacterial growth. The objective of this project is to assess changes to PG structure in autolysin deficient strains and masarimycin-treated cells using high-pressure liquid chromatography (HPLC) separation of muropeptides. We have previously shown that deletion of the autolysins LytC (amidase), LytF (endopeptidase), and LytD (N-acetylglucosaminidase) result in increased sensitivity to masarimycin. We hypothesize that changes in the PG structure will be unique in masarimycin-treated cells. Further, we hypothesize that the increased masarimycin sensitivity in the autolysin mutant strains is due to an inability to alter PG structure. Muropeptides were identified from the chromatograms using MALDI-mass spectrometry. The results show that in masarimycin-treated wild-type cells there is an increase in cross-linked muropeptides and N-deacetylation of N-acetylglucosamine. For the autolysin mutant strains, it is also seen that with masarimycin there is increased N-deacetylation in the ΔlytC strain while the ΔlytD strain showed a lack of muropeptides in the masarimycin treated sample. Analysis of cell surface proteins by SDS-PAGE showed distinct changes in surface protein profiles. Protein identification by mass spectrometry is currently underway.

This work was supported by the National Science Foundation

Stressed out: Investigating the Inhibitory Effects of Masarimycin on the Cell Wall

Monica Thoma, Jett DuVal, Jazmeen Hernandez & Christopher Reid

Biological & Biomedical Sciences, Bryant University, Smithfield, RI

Bacillus subtilis is a Gram-positive bacterium that serves as a model organism to study peptidoglycan metabolism in rod-shaped bacteria. Peptidoglycan (PG) is the major structural polymer made up of alternating chains of N-acetylglucosamine (GlcNAc) and N-acetylmuramic acid (MurNAc) residues connected by a β-(1,4)-glycosidic bond. Attached to the C-3 of MurNAc is a pentapeptide through which adjacent glycan strands are cross-linked to form a 3-dimensional mesh. PG offers shape and protection to bacterial cells and allows cell growth and division. Autolysins are degradative enzymes that regulate cell wall metabolism by cleaving specific bonds in PG to enable normal cell wall turnover. Autolysins contribute to cell growth, division, motility, PG maturation, protein secretion, and genetic competence. Our group has developed a chemical probe that inhibits the autolysin LytG from *B. subtilis*, called masarimycin. The purpose of this study is to determine if masarimycin induces cell wall stress and to confirm the mode of action of the probe. Previous experiments have shown that *B. subtilis* has increased masarimycin sensitivity in strains with deletions of autolysins LytC, LytD, and LytF. Thus, this study also investigates the cell wall stress response of masarimycin in an autolysin deficient background. Cell wall stress is monitored by measuring gene expression changes of the guanosine tetraphosphate (alarmone, ppGpp) synthetase relA, a key component in the stringent response. Results from qPCR showed an increase in relA expression in wild type *B. subtilis* treated with the β-lactam cefoxitin and masarimycin, indicating an activation of the stringent response and correlation with the observed masarimycin 'sausage link' phenotype. These results suggest that masarimycin has a similar mode of action to cefoxitin, a known cell-wall acting antibiotic. In contrast, when treated with masarimycin, the lytC mutant of *B. subtilis* showed a decrease in relA expression by over 1000 times, indicating decrease of the stress response and possibly contributing to the increased masarimycin sensitivity of this mutant background. Finally, in the lytD mutant, the increase in relA expression when treated with either masarimycin or cefoxitin indicates an increase in cell stress response. This is opposite to the effects in the lytC mutant, and we hypothesize that the differences in deleted enzymes in conjunction with masarimycin produce these effects on the cell wall.

This work was supported by the National Science Foundation

POSTER SESSION B

11:00 AM – 12:30 PM

Fascitelli Center for Advanced Engineering, 1st Floor

B-1 to B-23

Fascitelli Center for Advanced Engineering, Ground Floor

B-24 to B-51

Paramaz Avedisian ’54 Hall, College of Pharmacy

B-52 to B-76

Investigating the fitness costs of virus and host coevolution in RIM8 and *Synechococcus* spp. using quantitative PCR (qPCR)

Molly Matthews[1], Nathan Watlington[2] & Marcie Marston[1]

[1]Biology, Roger Williams University, Bristol, RI
[2]Marine Biology, Roger Williams Univeristy, Bristol, RI

In the age of the COVID-19 pandemic, understanding how viruses mutate and evolve has become more important than ever. New variants of the COVID-19 coronavirus are constantly being discovered and are continuing to infect humans despite attempts to build resistance through vaccines or direct exposure to the virus. Cyanophages are marine viruses that commonly infect the cyanobacteria host, *Synechococcus*. Both microorganisms are abundant in Narragansett Bay, and likely follow similar patterns of coevolution to the COVID-19 virus. The cyanophage isolate RIM8, a myovirus, and its host, *Synechococcus* strain WH7803, undergo antagonistic coevolution, an "evolutionary arms race" in which host cells evolve defenses against the virus, and the virus evolves to overcome host cell resistance. Host cells often develop resistance through changes in their cell surface receptors that can hinder viral attachment and infection. Subsequently, the virus can overcome this resistance through mutations in tail fiber proteins. In gaining the ability to infect resistant hosts, the coevolved virus may experience a fitness cost. For example, the coevolved virus may lose its ability to infect the ancestral host. In this experiment, we investigated the fitness of the coevolved cyanophage isolate RIM8 A-167-7 in comparison to the ancestral RIM8 viral strain. We hypothesized that as the virus evolves to overcome resistance and infect new coevolved hosts, its fitness will decrease on the ancestral host. We used a pairwise infectivity assay to collect filtered samples of viral lysate 0, 5, 24, and 48 hours after virus was initially introduced to the cells. Viral DNA replication was then quantified using qPCR. When infecting the ancestral host, there was a significant decrease in DNA replication of the coevolved virus compared to the ancestral viral strain. This suggests that there is a fitness trade off as the virus evolves to infect this resistant host. In future studies, we plan to explore viral fitness costs on different resistant hosts. Continuing to learn more about how marine viruses coevolve with their hosts can provide a better understanding of other coevolving viruses such as the COVID-19 coronavirus.

This work was supported by NSF-EPSCoR Track II & NSF BioOce

Changes in the distribution and genomic composition of cyanophages across environmental gradients

Veronica Ouellette[1], VEIL Lab[2], Jemma Dickson[1] & Marcie Marston[1]

[1]Biology & Marine Biology, Roger Williams University, Bristol, RI
[2]Plant & Soil Sciences, University of Delaware, Newark, DE

Synechococcus spp. are a type of unicellular cyanobacteria that are globally distributed and are responsible for up to 16% of net primary production in the ocean. Significant proportions of *Synechococcus* communities can be lysed daily by viruses called cyanophages. The distribution and genetic composition of *Synechococcus* and cyanophage communities are known to vary across environmental gradients. This study focuses on cyanophages in the Myoviridae family. These viruses have large dsDNA genomes (170 to 240 kb) and often carry host-derived genes in their genomes that can vary across environments. The purpose of this study was to characterize the distribution and genomic composition of *Synechococcus*-infecting cyanophages isolated from seawater samples obtained during a 10-day research cruise in August 2021. The samples were collected from 25 different sites with 4 depths per site (3 m, 15 m, chlorophyll max, and oxygen max) along a transect originating from Delaware Bay, through the Gulf Stream to a point approximately 140 nautical miles off the coast of Cape Hatteras, NC, and back. Viruses were isolated using enrichment plates where seawater samples were incubated with *Synechococcus* WH7803 cells. Individual viral isolates were then plaque purified, genotyped, and used in host range assays. Phylogenetic analyses based on DNA polymerase gene segments were done to compare the evolutionary relationships among cyanophage isolates obtained in this study and those in GenBank. Based on our enrichment plates, cyanophages were present in water samples from 13 different locations. However, no *Synechococcus*-infecting cyanophages were detected in water samples east of the Gulf Stream. In all, 120 viral isolates were characterized, and these fell into 22 different genetically distinct populations. Generally, cyanophage populations differed across sites and depths within a site. Next, full genome sequences were obtained for viral isolates representing different populations. The complete genome sequences of 14 viruses from this study are now being compared to the genomes of cyanophages isolated in Narragansett Bay, RI to examine the distribution of host-derived metabolic genes across large spatial scales. This is key to understanding how viruses may adapt to different environments and/or hosts and how gene exchange occurs across varying spatial scales.

This work was supported by NSF-EPSCoR Track II & NSF BioOce

Loss of photosynthetic genes in an evolved phage: investigating infection kinetics using quantitative PCR (qPCR)

Nathan Watlington, Molly Matthews & Marcie Marston

Biology & Marine Biology, Roger Williams University, Bristol, RI

Cyanobacteria are microorganisms that play an important role in regulating global biogeochemical cycles by contributing to oceanic primary production. Cyanophages are viruses that infect and kill cyanobacteria, resulting in high cyanobacteria mortality rates upward of 40% per day. Interactions among cyanobacteria and cyanophages result in selection pressures that lead to genetic variation within both populations. During repeated exposure to susceptible cyanobacteria, phages may evolve to better infect that particular host. Understanding the different types of mutations and how they influence infection dynamics could give insight into how viruses evolve to increase their host range and rate of infection. In this study, we investigated the infection kinetics of an RIM8 cyanophage isolate with a large deletion in the hypervariable gp16-gp17 region of its genome that is known to typically hold host-derived photosynthetic genes. The isolate was evolved in a 250 day evolution experiment on the *Synechococcus* WH8101 strain. The infection kinetics of the evolved virus were determined using quantitative polymerase chain reaction (qPCR), and were compared to the infection kinetics of the ancestral RIM8 virus on two different *Synechococcus* hosts. The replication of the two viral isolates was quantified on both *Synechococcus* strains WH8101 and WH7803. qPCR was performed on samples taken at 0, 5, 24, and 48 hours after introduction of the virus to the cyanobacteria. The samples were filtered through a 0.22 μm filter at the time of sampling to allow for quantification of free phage using primers for the viral DNA polymerase gene. Our hypothesis was that the genome deletion would allow for greater replication of the evolved virus as the shorter genome would allow for quicker production of virions. Results from the qPCR did not support this hypothesis as the two viral isolates showed similar replication rates when infecting the same cell type. Across cell type, the viral isolates replicated more on WH7803 than WH8101. These results suggest that under our experimental conditions, there was no consequence to the loss of genes in the evolved RIM8 isolate. In future experiments, we will be directly competing the evolved and ancestral viral isolates to determine if there are benefits provided by the large deletion in the evolved virus' genome.

This work was supported by NSF-EPSCoR Track II & NSF BioOce

Diversity of Bees in Trees on an Urban College Campus

Aidan Castricone & Rachael Bonoan

Biology, Providence College, Providence, RI

It is well known that insect pollinator populations are declining and a lack of nutrients (i.e flowering plants) may be a contributor to this decline. Insect pollinators forage for pollen and nectar, which provides proteins, fats, and carbohydrates, from flowering plants. Current research on pollinator foraging behavior focuses on pollinators that forage on the ground thus overlooking those that forage in trees. Trees such as Kousa dogwood (*Cornus kousa*) and crabapple (*Malus* sp.) bloom in early Spring, even before herbaceous perennials and forbs. Thus trees are likely the only places for early flying pollinators to find food. On Providence College campus, I studied bees foraging in trees using traps strung up via a pulley system. I chose 14 tree species that are likely to be pollinator-friendly based on past research and flowering times. Each tree had traps strung up both in the canopy and at the base. Each week contents of traps were collected and bees were pinned and identified to genus and when possible species. Pinned specimens will contribute to the Providence Pollinators reference collection and inform on-campus tree management for pollinator conservation.

This work was supported by a Providence College Walsh Fellowship

Assessment of population size, survival, and movement behavior of the rare frosted elfin butterfly

Breelyn Gilbert[1,2] & Rachael Bonoan[1]

[1]Biology, Providence College, Providence, RI
[2]Psychology, Providence College, Providence, RI

The frosted elfin butterfly (*Callophrys irus*) is a locally rare Lycaenaid with spotty distribution throughout the eastern United States. As host plant specialists, frosted elfins only lay eggs on two plant species. Yellow wild indigo (*Baptisia tinctoria*), one of the host plants for this species, is found in abundance at Gavins Pond (Foxboro MA). Since 2000, Massachusetts Butterfly Club members have been informally monitoring the frosted elfin population at this site; however, no systematic population surveys have been done. Using their data, I estimated annual population size from 2000 to 2022. In 2022, I also performed a mark-release-recapture study in 20 patches of indigo at Gavins Pond. I used my mark-recapture data to more accurately estimate population size, calculate daily survival of the species, and understand suspected territorial behavior. During the flight season, I walked the site as frequently as weather allowed. During these surveys, I marked captured butterflies and collected data on location, wing-wear, etc. I captured a total of 16 unique individuals with 17 recaptures, and I observed that few individuals seemed to express territoriality. These data show a strong need for conservation efforts to help restore the dwindling frosted elfin population at Gavins Pond.

This work was supported by the Southeastern New England Educational and Charitable Foundation

Floral nutritional value and plant-pollinator interactions in Providence, Rhode Island

Gracey Sorensen & Rachael Bonoan

Biology, Providence College, Providence, RI

Bees obtain nutrients from flowers. Pollen provides proteins and fats; nectar provides carbohydrates and some amino acids. With recent decline in pollinator populations, conservationists are interested in providing nutritious plants for pollinators. Counterintuitively, urban areas have the green space for pollinator friendly plantings. On Providence College's urban campus, I examined how floral abundance and pollinator interactions correlate to the nutritional value of plant species. Quadrat surveys were performed within 53 quadrats through the Spring and Summer for plant-pollinator interactions. Pollen was collected from all flowering species and analyzed for nitrogen and carbon content using an elemental analyzer. Nectar samples were collected and analyzed for amino acid concentration using a washing technique and colorimetric assay; nectar sugar analysis is ongoing. I predict that flowers higher in nutritional content will positively correlate with more visits by insect pollinators. The data collected will display which plants are of best nutritional quality and allow for floral management recommendations to benefit the pollinator population at Providence College and the surrounding city.

This work was supported by a Providence College Walsh Fellowship

Investigation of Langmuir and Freundlich Adsorption Isotherms of Essential Nutrient Anions (NO_2^-, NO_3^-, NH_4^+, PO_4^{3-}, and SO_4^{2-}) by Martian Regolith Simulants and Mount Hope Bay Sediments and Their Impact on Primary Producers' Growth

Isabel August, Teagan Bellitto, Carly Ferreira, Maggie Stefanowicz & Stephen O'Shea

Chemistry, Roger Williams University, Bristol, RI

For prolonged human missions to the Martian surface, available food sources will have to be cultivated on the planet to sustain the inhabitants. Food sources for both botanical and aquatic organisms require nutrient sources and growth substrates for rooting plants to thrive. This research investigated the potential of three various Martian regolith simulants from Exolith® in contrast to marine sediments from the Mount Hope Bay, R.I. to act as nutrient sources and growth substrates. The degree to which each of these substrates impact adsorption or desorption and their physio-chemical binding properties of nutrients, (NO_2^-, NO_3^-, NH_4^+, PO_4^{3-}, and SO_4^{2-}) will be a predictor of potential growth success. To address the equilibrium of nutrient binding to sediment, a series of Langmuir and Freundlich adsorption isotherms were created to establish qmax in mg nutrients/g adsorbate, adsorbate surface heterogeneity, and the strength of interaction. The impact of these substrates was further examined on the overlying water column for aquatic growth of primary producers, algae and diatoms, within BOD bottle microcosms by monitoring the extent of oxygen production and nutrient fluxes. The concentration of the various nutrients in solution was determined by spectro-colorimetric HACH® spot tests and the concentration of metal cations determined by ICP OES from filtered samples.

This work was supported by NASA Rhode Island Space Grant

Metal Ion Sequestration from Martian Simulant Regolith and Decarbonated Marine Sediment to Establish Their Suitability as Plant Substrate Growth Support

Teagan Bellitto[1], Isabel August[1], Maggie Stefanowicz[2], Carly Ferreira[1] & Stephen O'Shea[1]

[1]Chemistry, Roger Williams University, Bristol, RI
[2]Environmental Science, Roger Williams University, Bristol, RI

For Mars to become habitable for humans on extended missions it will ultimately require the establishment of a sustainable agricultural program. To this end, it will be necessary for the cultivation and facilitation of suitable rooted plants to have supportable substrates. This research investigated three various Martian regolith simulants from Exolith®, as potential substrates for plant growth and contrasted them with marine sediments collected from Mt. Hope Bay, RI. These sediment samples were further investigated to determine the impact of carbon and carbonate loss, the latter resembling mineral characteristics of the regolith, and their impact on pore water ion equilibria. Three regolith simulants, Mt. Hope Bay sediments, and their corresponding furnace-treated samples were analyzed for loosely available micronutrients and their impact on pH, ORP, granular size variation, and salinity as a model for ion dissolution into the overlying water column. To further model the extent of plants' potential to readily extract cations from substrates, deionized water was used as a baseline. Acetic acid and EDTA were used as plant siderophore mimics. HCl, as a total acid extractable metal, was used to identify maximum bioavailable trace metals. Pore water nutrient release was determined by spectropolarimetric HACH®spot tests and the concentration of trace metal cations was determined by ICP OES from filtered samples. The residue substrate extractant was washed with DI and further investigated by XRF to delineate the change in surface metal composition.

This work was supported by NASA and SURF

Investigation of Induced Single-Walled Carbon Nanotube Aggregation in Biologically Relevant Solvents

Ailill Smith, Matthew Card & Daniel Roxbury

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Carbon nanotubes require an amphiphilic functionalization in order to enhance dispersion in aqueous solutions. It is hypothesized that the stability and mobility of single-walled carbon nanotubes (SWCNTs) suspensions is related to the characteristics of the solvent in which the nanotubes are formulated. Solvents studied include: fetal bovine serum (FBS), which is a cell culture medium rich in proteins; and artificial seawater, which contains monovalent ions in the form of dissolved sodium chloride as well as divalent ions such as calcium and magnesium. The trends of aggregation in biological and marine environments may play a role in the consideration of SWCNTs as sensors in those aforementioned environments. Aggregation tends to affect the near-infrared (NIR) wavelength peak value(s) emitted by SWCNTs, most often by shifting them in the positive direction (a "red-shift"). This would discourage the use of solvents which highly aggregate SWCNT samples in sensing applications due to inaccurate wavelength data. SWCNTs were immobilized through a spin-coating technique which deposits nanotubes in solution on a hydrogel platform formulated with poly-(ethylene) glycol-diacrylate as the base polymer. NIR spectroscopy and photography was used in the capturing of images on this hydrogel and machine learning was used in the analysis of those pictures to quantify aggregate sizes.

This work was supported by RI NSF EPSCoR C-AIM

Cationic Functionalization of Single-Walled Carbon Nanotubes for Cytoplasmic Delivery into Synechococcus Cyanobacteria

Allison Schmaedeke, Aillil Smith, Matthew Card, Tania Thalyta Silva de Oliveira & Daniel Roxbury

Chemical Engineering, University of Rhode Island, Kingston, RI

Recently nanoparticles have gained a lot of interest and are significantly being used in many applications such as biosensing, bioimaging, and drug delivery. Nanoparticles are very versatile, modifiable, and robust research probes. Specifically, semiconducting single-walled carbon nanotubes (SWCNTs) have photostable near-infrared (NIR) fluorescence signals that respond to their local environment via modulations in wavelength and/or intensity. These qualities make it an easy and reliable tool for biosensing and bioimaging within living cells. However, investigations of NIR fluorescent SWCNTs interacting with bacteria remain largely unexplored. Therefore, this study aims to examine the interactions between SWCNTs and Cyanobacteria, i.e. a dominant photosynthetic, a bacteria responsible for harmful algal blooms. This was accomplished by inserting fluorescent SWCNTs functionalized with one of 3 different samples of amphiphilic polymers (e.g., GT15 DNA oligonucleotide, Lysozyme, and Chitosan, the latter two being cationic) into cyanobacteria. The methods for the study included using a Zetasizer instrument to measure the zeta potential (i.e. overall surface charge) of the three dispersed SWCNT samples. The interactions of SWCNTs and cyanobacteria were examined using NIR fluorescence microscopy to investigate the “spectral fingerprint” and localization of SWCNTs in the cyanobacteria at 3 different time points. We hypothesized that SWCNTs with a cationic functionalization will penetrate the multi-layer cell wall of the cyanobacteria, resulting in the SWCNTs entering the cytoplasm. Preliminary data suggest this hypothesis is true.

This work was supported by RI NSF EPSCoR C-AIM

The Microbial Ecology of Benthic Habitats in Rhode Island: A Ciliate Diversity Study at Point Judith Pond

Abigail Goodman

Cell & Molecular Biology, University of Rhode Island, Kingston, RI

Ciliates are single-celled eukaryotes (protists) which are able to thrive in a wide array of habitats globally, including anoxic habitats such as marine coastal sediments. Ciliates are integral members of microbial food webs and the anoxic ecosystem just below the surface of these sediments. While these organisms are extremely diverse both morphologically and ecologically, much of their diversity is still undescribed. The full diversity of anaerobic ciliates is especially unknown, largely due to difficulties in culturing. In this study we set out to investigate this underexplored diversity of ciliate and protist communities in the anoxic sediments of Point Judith Pond via high-throughput DNA sequencing. We collected 15 sediment samples from 5 different sites and sequenced the universal 18S rRNA marker genes from them in order to identify the members of these communities. We found a wide range of protist diversity among our samples, and this diversity was largely influenced by the relative geographic location of the sites. Gaining a better understanding of the members of these underexplored anoxic microbial communities is especially important as oceanic hypoxic zones expand globally due to climate change.

This work was supported by RI NSF EPSCoR C-AIM

Using Satellites to Analyze Temperature, Chlorophyll and Salinity in Narragansett Bay

Lorenzo Davidson & Daniel Wexler

Earth, Environmental, & Planetary Sciences, Brown University, Providence, RI

Narragansett Bay is the largest estuary in New England and is therefore of great environmental and economic importance. Not only does it support a biologically diverse ecosystem, but it also helps to filter the air we breathe and the water we drink. Since the start of the 20th century, Narragansett Bay has experienced above average increases in water temperature and significant pollution. Quantifying how different components of the Bay have changed over time in response to climate change, regionally specific anthropogenic actions, and natural processes, is critical to understanding the future of the Bay. There are a number of buoys in the region that collect data at a high temporal resolution, but this data is limited to only certain months of the year and to the specific locations of the buoys. In this study, we use remotely-sensed data from the Landsat 5, 7, and 8 satellites to expand the record of sea-surface temperature (SST) to the entirety of the Bay over a 38-year period (1984-2022), and we make progress towards being able to measure chlorophyll (Chl) and sea-surface salinity (SSS) from satellite imagery. For SST, we calibrate the thermal bands of the three satellites against the in-situ buoy measurements using two different methods of bias corrections, one that uses the entire dataset and another that splits the data at different temperature thresholds and calculates a different bias for each side of this split. For Chl, we use two different multi-band algorithms to extract chlorophyll readings from the blue and green bands of the satellites and compare their respective accuracies. For SSS, we test for correlations between individual Landsat bands and buoy measurements to see if salinity is connected to color. In the future, we will use our record of SST to quantify how different parts of the Bay have changed over time and measure the amount of warming that has taken place over the past 38 years. We also plan to use Extended Empirical Orthogonal Function (EEOF) analyses, a pattern recognition technique, to improve our calibrations for Chl and SSS. In addition to improving our understanding of the physical and biological processes that govern Narragansett Bay, these records of SST, Chl, and SSS could be useful for evaluating climate models of the region that predict future climatic trends.

This work was supported by RI NSF EPSCoR C-AIM

Describing neoplastic cells of hard clams, *Mercenaria mercenaria* and testing the survivability of the neoplastic cells under various environmental conditions.

Casey Dunbar, Abigail Scro & Roxanna Smolowitz

Aquatic Diagnostic Laboratory, Roger Williams University, Bristol, RI

Hemic neoplasia (HN) is a contagious, leukemic-like disease that affects several species of bivalves worldwide. HN also affects the commonly cultured hard-shell clam *Mercenaria mercenaria* on areas of Cape Cod, Massachusetts. The disease is transmitted among bivalves via filtration of water containing infectious cells. As the disease progresses, neoplastic cells proliferate in the hemolymph (blood) of the organism, filling the vascular system and resulting in significant mortality. Interestingly, neoplastic cells are only infectious for the species in which they originate. Preliminary work consisted of several staining methods to describe the morphology of neoplastic and normal hemocytes. It also examined the extent of PCNA staining of hemolymph smears to determine proliferation abundance of cancerous cells. This project focused on continuing to describe neoplastic hemocytes and testing the survivability of cells in the water column. First, an Oil Red O stain was used to evaluate lipid accumulation. The stain was expected to stain any lipid inclusions within the cytoplasm of the neoplastic cells (as have been seen in HN cells originating in *Mya arenaria*). However, no lipid inclusions were noted in the cytoplasm of Mercenaria mercenaria neoplastic cells. A PCNA analysis was performed to identify the location of highly proliferative cells in tissues. After staining, the neoplastic cells showed nuclear expression of PCNA and high amounts of PCNA-positivity; thus, indicating high proliferation rates. Additionally, a trypan blue stain will be used to analyze neoplastic cells throughout environmental water parameter (salinity, temperature, and pH) experiments to hopefully determine the preferred environments for diseased cells to survive. In future studies, the results from this project will aid in describing neoplastic cells and help understand the epidemiology concerning environmental conditions under which HN spreads from one animal to another.

This work was supported by the Mark Gould Endowed Scholarship and Research Award at Roger Williams University

Development and optimization of a qPCR assay to detect three common fish parasites, *Cryptocaryon irritans*, *Uronema marinum*, and *Neobenedenia* spp., in aquarium systems

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Cryptocaryon irritans, *Uronema marinum*, and *Neobenedenia* spp. are parasites responsible for common illnesses in aquarium fish. Marine "ich", or white spot disease, is caused by *C. irritans* and is the cause of large mortality events in many aquaria. The protozoan has a complex life cycle that allows for fast reproduction and infection. As a result, there would be a significant amount of *C. irritans* present in the water column before major infection takes place. The skin fluke *Neobenedenia* spp. has a similar free-swimming stage in its life cycle that would make it detectable in the water column for a period of time. The free-living parasite *U. marinum* does not need a living host to be able to survive, thus the parasite can remain in the water column for the entire duration of its life cycle, making its detection in water likely. At times of disease it is expected that increases in this parasite would occur in the water column. The ability to detect these parasites in the water column would allow aquarists to identify and quantify the abundance before major infection and subsequent mortality event occurs.

Archived 1L water samples were collected from the New England Aquarium and filtered through 0.22 μm filters. DNA was then extracted using the Qiagen PowerWater DNA Extraction kit and quantified. Primers previously designed for *C. irritans* targeting the 18S region by Taniguchi et al. 2011 were used. Primers were newly designed for *U. marinum* targeting the COX1 gene. Primers were confirmed via melt curve analysis in a SYBR Green qPCR assay, and linear plasmids were then developed. Probes were newly designed for both *C. irritans* (FAM) and *U. marinum* (Cy5). Singleplex TaqMan qPCR assays for *C. irritans* and *U. marinum* have been developed and verified. Primers have been newly designed for *Neobenedenia* spp., and development of the singleplex SYBR Green assay is ongoing. The goal of this research is to develop a multiplex TaqMan® qPCR assay with pathogen specific primers and probes to target *C. irritans*, *U. marinum*, and *Neobenedenia* spp. in the water column to monitor and predict parasite outbreaks. From these results, threshold levels can be developed and applied to aquarium systems worldwide. Future work includes filtering larger volumes (>100L) of water through Waterra eDNA capsules in order to obtain a more accurate sample size for tanks of larger volumes.

This work was supported by RI NSF EPSCoR C-AIM

Setting the Baseline: Determining Species Richness of Rhode Island Rhodomelaceae with Morphological and Molecular Data

Ben Carolan, Thomas Irvine & Brian Wysor

Biology, Marine Biology & Environmental Science, Roger Williams University, Bristol, RI

The Rhodomelaceae is a group of filamentous red algae with worldwide distribution. The family is well known to be one of the most species rich families of red algae, but it is also well known for the propensity of its species to be introduced beyond their native ranges. This propensity combined with challenges to morphological identification make DNA barcoding an essential tool for documenting the presence of Rhodomelaceae species, and detecting introduced species in local marine floras. In this study, species identities of local Rhode Island Rhodomelaceae were compared with morphological characters as well as sequences from DNA barcoding using two chloroplast encoded DNA markers, UPA and rbcL. Our preliminary data show general concordance between the UPA and rbcL markers, and in many but not all cases morphological interpretations are supported by molecular data. Using these data we evaluate species richness of Rhode Island Rhodomelaceae in the light of historical records. Eighteen distinct species are reported here which is consistent with historical records which report very similar diversity.

This work was supported by the Department of Biology, Roger Williams University

Thermal plasticity at different elevations in wild-caught *Drosophila pseudoobscura* from the Colorado Rocky Mountains

Jacqueline Jimenez, Celia Celona, Ingrid Kinder & Anna Schumacher

Biomedical sciences, Salve Regina University, Newport, RI

Due to climate change, temperatures are expected to increase and severe weather events to become more common. This is a matter of ongoing concern because temperature is a fundamental factor of an organism's niche, influencing viability at all levels, from cellular to organismal. The climate variability hypothesis (CVH) proposes that animals native to a thermally variable environment are more likely to have mechanisms that allow them to survive severe changes in temperatures than those originating from a thermally stable environment. Our goal was to obtain more data about how animals will adapt to climate change and whether there is a variation that permits them to do so. To investigate the CVH, we used wild-caught *Drosophila pseudoobscura* lines from various elevational transects in Colorado and measured their plasticity in response to heat stress. Elevation was used as a proxy for climate variability because temperatures tend to fluctuate more at higher elevations. To have a better picture of what happens when temperatures increase, we looked at the survivorship of each fly line (low, medium, and high elevation lines) at a comfortable and high temperature (20 vs. 25°C). We also performed an experiment testing critical thermal minimum (Ct min) to determine the temperature at which the flies lose control of their motor skills, as well as a heat knockdown experiment (HKD) to see how long it takes them to lose control. We predict that, in accordance with the CVH, *D. pseudoobscura* from higher elevations and thus more variable climates will exhibit a greater ability to survive and acclimate to hotter temperatures.

This work was supported by ThermoFly

Identification of Fly Species and Presence of *Wolbachia* Among Narragansett Bay Flies

Anna Schumacher, Theo Modla, Jackie Jimenez, Libby Baldwin, Emily Weed, Rebecca Bachtel, Ioulia Bespalova & Heather Axen

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Climate change models have shown that global temperatures are predicted to rise alongside the occurrence of extreme weather. Temperature changes may affect animals, especially ectotherms, health and growth. Some parasites and bacteria, including the bacteria *Wolbachia*, have been shown to have relationships with insects and may affect how insects respond to thermal stressors. Assessing the frequency of *Wolbachia* infection can help make predictions about endosymbionts and their effect on thermal tolerance. Also, different species may show different patterns of infection, suggesting that species richness is important to note. We focused on *Drosophila* (fruit flies) along Narragansett Bay to identify species and the presence of *Wolbachia*. We extracted DNA and amplified mitochondrial gene cytochrome one (COI) to conduct species identification. We then sequenced the gene of interest to identify the species of the flies through GenBank. To test for the presence of *Wolbachia* we amplified the frequency of *Wolbachia* surface protein gene (wsp). The preliminary results indicate that 0.23 of female *Drosophila* lines tested positive for *Wolbachia*. Five of the lines with *Wolbachia* were *Drosophila melanogaster* and six were unidentified. Further testing will be conducted to obtain a broader understanding of *Drosophila* populations along Narragansett Bay. Overall, the populations of *Drosophila* along Narragansett Bay provide an opportunity to understand how populations can respond to climate change.

This work was supported by ThermoFly

Metabolic Labeling of Zebrafish Embryonic Cell N-glycans Using Azido-Sugars

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Zebrafish embryonic cells are used in many facets of research such as maintenance of transient cell populations and control of chemical and mechanical cues received by cells, as well as their use as a model for environmental toxicology. A focus of our research is to use zebrafish as a model to investigate molecular changes that occur when vertebrates are exposed to pollutants (ie polyfluorinated alkylsulfonates (PFAS)). Protein glycosylation is a common post-translational modification involved in numerous biological functions such as cell to cell communication, protein trafficking, and cell adhesion. It is the process where sugars and carbohydrates (glycans) are covalently attached to proteins via an N- or O-glycosidic linkage. The objective of our research is to investigate the use of a bio-orthogonal metabolic labeling strategy using azido sugars to visualize changes in the cell N-glycosylation patterns in response to environmental stress. Our initial choice for synthesis of a metabolic label was N-acetylglucosamine as it is found in all N-glycans. Based on this we synthesized a 2-azidoacetamido-2-deoxy-D-glucopyranoside (GlcNAz) derivative. GlcNAz was synthesized from D-glucosamine hydrochloride via acetylation with chloroacetic anhydride. Next, the azide was introduced by addition of sodium azide in DMF at 50oC while stirring overnight. The resulting GlcNAz was peracetylated using acetic anhydride and pyridine to give 1,3,4,6-tetra-O-acetyl-2-azidoacetamido-2-deoxy-D-glucopyranoside (perOAcGlcNAz). The perOAcGlcNAz was purified by flash and reversed-phase chromatography. We tested the incorporation of GlcNAz into zebrafish cells in a titration experiment to investigate ideal sugar concentration for incorporation. A perOAcGlcNAz concentration range of 46µM to 138µM was screened. The cells were incubated for 3 days at 30°C until cell confluence was reached. Cells were harvested, lysed in phosphate buffered saline via sonication. Acetone precipitation of proteins was used to concentrate the samples. Glycans were then labeled with alkyne-(PEG4)-biotin in 1mM ascorbic acid and 1 mM copper (II) sulfate solution. The biotinylated glycans were detected in a dot blot with horseradish peroxidase labeled streptavidin. Preliminary results demonstrated weak labeling in cells incubated with 43 µM perOAcGlcNAz. Future experiments will involve flow cytometry and fluorescent microscopy to observe any changes within the cell.

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Inducing reproductive development and spore release in tubular *Ulva*

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The macroalgae *Ulva* are a ubiquitous, major contributor to biofouling colonies. Biofouling affects the durability and functionality of marine sensors amongst other marine technologies. *Ulva* has an isomorphic alternation of generations from a diploid sporophyte which produces quadriflagellate zoospores to haploid gametophytes that produce biflaggelate gametes that fuse to develop into the sporophyte. Quadriflagellate zoospores of *Ulva* are used as a model organism in many biofouling studies. The goal of this study was to develop a protocol to induce reproduction and spore release in *Ulva* to offset issues with current field collection methods. Common concerns include seasonal time constraints and inconsistent supply of reproductive *Ulva*. This prevents experiments from running consistently and timely. We quantified the effect of temperature shock, photoperiod, and nutrients on initiating reproduction of tubular Ulva. Experiments exposed the thalli to a temperature shock (4C) for 1 hour while submerged in 1 mL of sterile filtered seawater (- nutrients) or 1 mL of sterile filtered Von Stosch (+ nutrients). Half of the specimens were subsequently placed under a 12:12 light:dark photoperiod at 20C. The remaining half were placed under a 16:8 light:dark photoperiod at 20C. Thalli were monitored daily for signs of reproductive development and evidence of spore release. We found spore release generally peaked two days after the temperature shock. We also found most thalli in these experiments released biflagellate gametes and few released the desired quadriflagellate sporophytes. The specimens which released sporophytes are being replicated and maintained for further study.

This work was supported by RI NSF EPSCoR C-AIM

The RI C-AIM Narragansett Bay Observatory Project

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The RI C-AIM Narragansett Bay Observatory Project looks to achieve real-time monitoring and high frequency sampling of Rhode Island's Narragansett Bay. The effort towards a smart and interconnected bay is driven by the desire to better understand the ecosystem dynamics that contribute to the growth of harmful algal blooms (HABs) which disrupt the shellfish industry and coastal communities. HABs cause contamination of shellfish, and consumption can lead to illnesses such as paralytic shellfish poisoning, caused by saxitoxin, or amnesic shellfish poisoning caused by domoic acid. When a HAB event occurs, harmful species of algae dominate the phytoplankton community, and produce toxic organic compounds in large quantities. This results in build up of these toxic compounds in the tissues of higher trophic species like shellfish. The highest concern of shellfisheries in Narragansett Bay are the species Alexandrium and Pseudo-nitzschia, both of which produce neurotoxins that accumulate in shellfish tissue. The ability to predict these bloom events will allow for informed management decisions to mitigate their economic impact and risks to human health. To aid in the prediction of these events two coastal monitoring buoys were deployed near Jamestown and Greenwich Bay, in addition to a multi-depth pump station at Castle Hill Lighthouse in the East Passage. High-resolution data collected includes oceanographic parameters such as nutrients, chlorophyll a fluorescence, dissolved oxygen, turbidity, salinity, pH, and temperature. The data collected from these stations will be used to inform predictive algorithms that can forecast the likelihood of a HAB event based on real-time conditions within the bay. During the summer of 2022 improvements to the engineering of these autonomous buoy platforms were performed, including the design and fabrication of clamps to secure the oceanographic sensors. Created using CAD software and 3D printing, these clamps are a complete redesign of the current sensor retention mechanism, and eliminate the need for tools when divers service the sensors. Clamps were also designed to anchor data cables with unrestricted movement, which caused temporary data transmission blackouts when currents put strain on the connections. Lastly, modifications to the current Bay Observatory website were made, to create an interactive and educational component where the public can learn about the methods of this project and why the data collected is important.

This work was supported by RI NSF EPSCoR C-AIM

What are the Impacts of TLP on Salt Marsh Foraminifera?

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Salt marshes are an important environment for young fish and crustaceans and act as a buffer to protect our shorelines. Salt marshes are currently at risk due to the impacts of global climate change, such as sea level rise. If climate change continues as currently projected, Narragansett Bay salt marshes may be underwater within the next 100 years. As a way to combat this, the RI DEM (Rhode Island Department of Environmental Management) has been adding sediment to salt marshes through a method called thin layer placement (TLP) to assist in restoration and reduce the effects of degradation. The restoration has shown a positive effect on vegetation and reducing degradation in the marsh, but its effect in intertidal zones has not been studied. Within intertidal zones, single-celled organisms called foraminifera are abundant. Species within this phylum help mediate nitrogen and carbon cycling in salt marshes and can serve as important indicators of benthic health. By monitoring foraminifera populations in Narragansett Bay salt marshes we may be able to better understand how the TLP restoration has impacted the intertidal regions of salt marshes. In this study, three salt marsh sites with either no TLP restoration or TLP restoration performed 3 or 5 years ago were sampled to compare the abundance and diversity of foraminifera populations. Preliminary results suggest that restoration has not had a negative impact on foraminifera populations and may have increased foraminifera abundance and diversity within specific marshes.

This work was supported by RI NSF EPSCoR C-AIM

What's the story? Developing engaging narratives to communicate scientific research

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From developing ceramic water filters for accessible drinking water to creating computer models that predict future changes in Narragansett Bay, RI C-AIM research strives to answer important scientific questions regarding climate change in the Ocean State. There is a critical need for this research to reach diverse audiences outside the scientific community that hold a social, cultural and/or economic stake in the health of Narragansett Bay.

Storytelling is one of the most powerful ways to inspire and encourage a more engaged and informed public. Personal stories can demystify the processes and results of scientific research, which is often inaccessible to those without the expertise. Science communication can offer new insights into how science stories are as relevant to the social fabric as any other news story on politics or pop culture.

This project entails engaging with other SURF students to create journalistic pieces accompanied by artistic visuals that highlight their research, research significance, and experiences. I develop interview questions, conduct interviews, and craft narrative stories that effectively communicate the science of a given research topic, as well as people's personal pursuits and journeys.

The final project culminates in the form of a website, which contains all of the completed narrative stories and visuals ready for easy distribution.

This work was supported by RI NSF EPSCoR C-AIM

Characterizing the Cell-Associated Bacterial Community of two toxigenic *Pseudo-nitzschia* Species in Culture

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In 2016 and 2017, Narragansett Bay (NB), Rhode Island shellfishing harvests were shut down for the first time due to high levels of neurotoxin domoic acid (DA). Ingestion of DA by humans can cause short-term memory loss, brain damage, and, in severe cases, death. *Pseudo-nitzschia* is a genus of diatom capable of producing DA and is considered a harmful algal bloom (HAB) taxon. In NB, *Pseudo-nitzschia* has been recorded in Narragansett Bay since the 1960s but did not cause toxin-related closures until 2016-17. Species within the *Pseudo-nitzschia* genus vary in toxicity, making HAB forecasting difficult. Additionally, environmental factors, including the bacteria associated with *Pseudo-nitzschia* cells, affect DA production. Research has shown that increases in DA levels are correlated with a decrease in the biodiversity of the cell-associated bacteria community of *Pseudo-nitzschia* in field and culture samples. The correlation between DA levels and the biodiversity of bacteria leads researchers to believe that *Pseudo-nitzschia* produces DA as a deterrent to pathogenic bacteria. NB has a mix of toxic and non-toxic species, with two of the most common being the toxin producers, *P. pungens* and *P. multiseries*. In culture, isolates of the two species produce variable amounts of DA or do not produce the toxin. In this project, we assessed the growth rate of *Pseudo-nitzschia* cultures and genotyped the plankton (or cell)-associated bacteria using 16S amplicon sequencing.
Single-cell isolates of *P. pungens* and *P. multiseries* were obtained from two sites in NB: the NB Long-term Plankton Time Series (NBPTS) and Whale Rock. To identify the microbiomes of our DA-producing (DA+) and non-DA-producing (DA-) cultures, we targeted the 16S rRNA gene and assigned taxonomy using lab pipelines. In preparation for further work, we ran growth curves in triplicate on the cultures to identify the timing of late exponential growth for DA+ and DA- P. multiseries and *P. pungens* cultures. With this information, we will be able to characterize the microbiomes of these cultures and compare them with recent isolates and field samples from NB. Understanding the microbiome of toxic *Pseudo-nitzschia* species in NB is critical for public health protection as it is one of the many environmental factors that affect DA production.

This work was supported by RI NSF EPSCoR C-AIM

Platform Development for collection of Neural Activity in Hand Pre-shaping to Grasp using Noninvasive EEG

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Hand dexterity and grasp disability in patients with spinal cord injury has devastating impacts over their lifespan. Restoration of hand dexterity is the highest priority among this population. While some invasive brain-machine interfaces (BMI) are customized to assist such patients to perform reach-and-grasp tasks with a robotic system, they lack dexterity, generalizability, and are cost inefficient. This creates difficulties and prevents accessibility for employing these devices in larger patient populations. The purpose of this study is to develop a noninvasive BMI platform that has the ability to predict planned imaginary grip types and different pre-shaping actions based on electroencephalogram (EEG) recordings from a new, low-cost eight channel EEG headset (Unicorn Hybrid Black). Data collection for our pilot study included a preliminary protocol that consisted of collecting EEG recordings when the human subjects were instructed to consecutively power grip their left and right hands for a specified duration of time. Additionally, we are in the process of enlarging our dataset to collect planned grip types for dominant hands. As the initial step, we developed a machine learning pipeline using Python language. We analyzed the EEG data from the pilot study by considering the baseline and filtering into different bands. Based on the significance of slow oscillations recorded within the premotor cortex from invasive BMI studies, we selected the power of the δ (delta) band for feature extraction and classification. We focused on channels C3 and C4 to distinguish between left and right hand power grips. This yielded a percent accuracy of 55% (C3) and 80% (C4) using thresholding methods. Selected grip types for the new study are the transverse and lateral cylindrical grips. Developing a PC - controlled turntable for object presentation enables us to collect EEG data to investigate whether the brain engages in pre-shaping of grip movement prior to the action or motor imagery of the grip movement. Classification of this period prior to the grip/imagery task will aid the process of overcoming limitations that restrict dexterous interaction of neurorehabilitation devices with objects. Our vision is to use this low-cost platform to manipulate objects with a robotic arm among the disabled population.

This work was supported by RI-INBRE

Head Motion Detector as a Universal Joystick for Disabled Patients

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With 5.4 million individuals in the United States alone who suffer from paralysis, assistive devices are important to grant independence to these patients. Such devices may be invasive such as a recent implantable BCI designed for control of complex assistive devices or less invasive such as computer cursor control interfaces that have been developed using electroencephalography and head motion tracking with a camera. This study aims to develop a new and non-invasive head motion detector to serve as a universal joystick in controlling 2D and 3D assistive device applications. In our current work, the sensor and the 3D-printed, head-mounted case have been successfully integrated to comfortably control a computer cursor and virtual robotic arm. We used an Arduino MPU-6050 as our inertial measurement unit (IMU) and an Arduino Pro Micro as our microcontroller. With the ability to perform digital motion processing (DMP), our IMU can process head motion with six degrees of freedom. As a user performs head movements, the microcontroller receives data from the IMU and relays it to a Python script to be translated to cursor movement. In our Arduino script, a median filter with nine points in its filter window is applied to our raw data for processing, and the script has a sampling frequency of 100 Hz. In our pilot study development for a 2D cursor control task, the two degrees of freedom needed were pitch and yaw, and these motions were recorded using gravitational acceleration as a means to understand head orientation. We developed a custom graphical user interface (GUI) that serves to test our 2D cursor control and familiarize users with the required head motion. Modifying this platform, we transitioned to controlling a 3D virtual robotic arm. This eliminated our GUI and directly controlled the 3D virtual robotic arm with 2 degrees of head movement. Our future goal is to investigate the usability of our platform in controlling a complex physical robotic arm with grasping capabilities, while addressing the control trade-off between a human and an intelligent robot.

This work was supported by RI-INBRE

Prototype Development and Testing of 'Nearly No-Prep' Tripolar Concentric Ring Electrode for use in Electrical Brain Signal Detection

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Electroencephalogram, or EEG, is a widely used, non-invasive, and cost-effective procedure for investigating neural activity produced by the brain. It is used in both clinical and research settings for diagnostics, research, and treatment of many neurological conditions. EEG preparation contains several tedious steps to ensure good electrical conductivity between the scalp and electrode. A Hydrogel filled electrode based on the geometry of the tripolar concentric ring electrode (TCRE) was developed for use performing tripolar EEG (tEEG) with minimal, or 'Nearly No Prep', work. To develop this, computer-aided design (CAD) software was utilized to design the prototype and it was 3D printed using SLA UV-cure resin for the best resolution and access to materials available. Testing of the prototype compared its performance to that of a commercially available TCRE. Skin-to-electrode impedance, general usability, signal detection, EEG, and ECG tests were conducted. Modification of the design will continue based on the performance results of these tests. The 'Nearly No Prep' prototype in its current design shows potential that it will perform similarly to the reference TCRE while also reducing the number of preparation steps needed to utilize them for tEEG purposes. It has met the <5 kOhms impedance range and successfully detected a 10 Hz sine wave signal. After further research and development, this will allow researchers and students to more easily and efficiently utilize these electrodes for EEG research purposes.

This work was supported by RI-INBRE

Rehabilitation Bike for Motor-Impaired Patients

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Patients who have suffered from a stroke or neurodegenerative disorders such as Parkinson's Disease (PD) can suffer from a decrease in mobility and movement disabilities. They need to engage in regular exercise to maintain the connections between their neurons to slow the progression of the disease. Forced exercise improves motor function in patients with neurological disorders by promoting neuroplasticity.

The overarching aim of this project is to create a closed loop system that takes in a patient's heart rate and converts the measurement into a motor speed dependent on the patient's selected heart rate zone. Subsequently, a virtual reality environment will run and adjust after each iteration period.

Mainly, this summer I focused on ensuring bike functioning and doing a feasibility study with healthy participants before integrating VR with the Bike. This involved debugging the existing code and filtering the ECG output using a bandpass filter to calculate the heart rate. I also needed to add a newly designed PCB that would control the bike reliably during a voltage shift. The procedure of the feasibility study involves 2 surveys, a 20-minute ride on the bike, Time up and Go, 10 meter Walk Test, and 30 second Sit to Stand. We aim to get data from 5 healthy and 5 Parkinson-diagnosed participants. So far, we have collected data from healthy participants and soon we will start collecting data from Parkinson's patients

This work was supported by RI-INBRE and the URI Science & Engineering Fellows

Mental Health and the Environment for Unaccompanied Children in the United States: Survey Development and Preliminary Results

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Unaccompanied Immigrant Children (UC) are a vulnerable and understudied population of immigrants who have been arriving to the United States (US) in increasing numbers in recent years. There is a lack of knowledge about mental health outcomes for UC in the US, specifically how their environment impacts their resilience and acculturation. The aim of this project is to develop a survey that will enhance understanding of mental health outcomes for UC in the US. Participants are from a convenience sample of all UC in the US who are receiving post release services (PRS) from Heartland Alliance partner agencies. PRS are community-based case management services that help connect UC with education, health, and legal support. The survey includes demographic data and four different scales: the Strengths & Difficulties Questionnaire (SDQ), Child and Youth Resilience Measure (CYRM), Sense of Community Index (SCI) and the Acculturation, Habits, and Interests Multicultural Scale for Adolescents (AHIMSA). These instruments measure mental distress, resilience, environmental factors, and acculturation, respectively. Preliminary findings from the SDQ were collected during Spring 2022 by PRS case managers and these results informed our survey development. Results show differences in total SDQ score by gender and country of origin; however, these differences are not statistically significant. These preliminary results suggest that UC have unique mental health challenges, but also display strong resilience. The preliminary SDQ results enhance our baseline understanding of mental health outcomes for UC and will inform our interpretation of future research that considers factors such as resilience, community, and acculturation.

This work was supported by RI-INBRE

Neural Correlates of Visual Statistical Learning and Morphological Processing

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Implicit Statistical Learning (ISL) is the mechanism that enables humans to pick up on patterns in the environment. Is ISL being used to acquire word structure or morphology when reading? Do individual differences in ISL influence the ability to acquire word structure? Reading and writing are essential skills, especially in the world today. Still, learning disorders, such as dyslexia, make it difficult for individuals to acquire these much-needed skills. By understanding the role that ISL plays in the acquisition of word structure and the extent to which individual differences influence this process, we will have a better understanding of how to aid individuals, such as those with dyslexia, to acquire the skills of reading and writing. This study's overall aim is to measure reader's lexical representation quality, sensitivity to the internal structure of words, and the ability to notice statistical patterns in visual sequences. These behavioral measures will then be correlated with event-related potentials (ERPs) to identify neural signals of ISL and determine if similar neural signals are used during the decomposition of complex words. The data presented here are electrophysiological data and demographic data which suggest that there are two different types of readers that can be differentiated based on their ability to decompose word structure when reading.

This work was supported by RI-INBRE

Transitive Inference in Rats

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Transitive inference (TI) is a form of deductive reasoning where once stimulus pairs are learned (A>B; B>C), individuals can infer that A>C. Humans and some animals use TI to create mental hierarchies that can aid in decision-making tasks. Studies evaluating TI in rodents are limited, but the use of rodent models could shed light on the evolutionary development of TI as a cognitive process. Ten male Long-Evans rats were trained to discriminate four stimuli (odor) pairs for one list (A>B; C>D; C>E; list 1) by reinforcing the higher stimulus in each adjacent premise pair and F>G>H>I>. After reaching the criterion on the adjacent stimulus pairs, rats received non-differentially reinforced probe trials of non-adjacent stimulus pairs (e.g. B, D) to determine the extent to which rats constructed a list or mental hierarchy. Rats were then trained on a second list using the same procedure as described above with the exception that specific odors were consistently presented in unique spatial locations. Previous research has shown that when lists are spatial, acquisition increases, and other research also suggests inferred orders might share a common magnitude system with space. Preliminary results suggest that list acquisition did increase with spatially presented stimuli. Results on the first list and their probe pairs indicate that rats can learn a list through premise pair training, but it is not yet clear if the rats use TI to construct a list. With the use of rodent models, insight can be gained into the evolutionary advantages of being able to utilize TI.

This work was supported by RI-INBRE

Development towards a 3D Bioprinted Near-Infrared (NIR) Light Responsive Core-Shell Hydrogel Patch as a Topical Wound Infection Therapy

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In the United States, over 2.8 million antibiotic-resistant wound infections occurred in 2019 leading to over 35,000 deaths, posing a significant clinical challenge. Current treatments include hydrogels, which are highly hydrophilic polymers that can encapsulate cargo (e.g., antibiotics) and provide an aqueous environment to reduce swelling and inflammation at infection sites, making them suitable for wound treatments in healthcare settings. However, their inability to control the release of antibiotics gives rise to skin toxicity and more resistant strains of bacteria; therefore, more work is needed to prevent the exacerbation of open wound infections. In this study, a three-dimensional (3D) bioprinted photoresponsive hydrogel patch with a core-shell architecture is proposed, and methods of its printing and simulating its drug release are investigated. The hypothesized mechanism involves NIR irradiation of the hydrogels, which triggers the localized surface plasmon resonance (LSPR) activity of the NIR-tuned gold nanorods (Au NRs) encapsulated in the shell, locally increasing the heat and therefore the solubility of ciprofloxacin encapsulated within the core, thermally controlling its on-demand release. Hydrogel substrate and bioprinting parameter optimization led to the clearest delineation between core and shell using a hybrid shell (calcium-crosslinked 3% (w/v) alginate and 10% (v/v) poly (ethylene glycol) diacrylate (PEGDA)) and a core (10% (v/v) PEGDA) printed at 50 and 20 kPa respectively, with an extrusion feed rate of 1.3 mm/s and a minute-long exposure to UV light. Image analysis is conducted to quantify diameters to obtain spreading ratios for the shell (1.063 ± 0.052) and core (1.756 ± 0.110) on all straight edges of the hydrogels. During printer parameter optimization, a macroscopic, cylindrical version of the core-shell construct using 10% (v/v) PEGDA (core) and 3% (w/v) alginate (shell) is developed via separate template molding. The cumulative release behavior of ciprofloxacin is investigated at either 23°C or 40°C, mimicking a heat profile of encapsulated Au NRs upon NIR irradiation. Ciprofloxacin release is manipulated with an on/off heat cycle after an initial burst release from the core-shell system, exhibiting user control of ciprofloxacin release over time.

This work was supported by Brown University Undergraduate Teaching and Research Awards

Toward thiocyanate detection in saliva: Improvements in chemical synthesis of functionalized metalloporphyrins

Julie Gerstner, Antal Lee & Clifford Murphy

Chemistry, Roger Williams University, Bristol, RI

Thiocyanate concentration in saliva has been suggested as a chemical marker for a screening analysis for oral and gastric cancers. In this project, we seek to adapt an existing technology for the electrochemical detection of thiocyanate in seawater to an application as a screening tool for use as a part of a routine dental exam. The chemosensor electrode is comprised of a fluorine-doped tin oxide (FTO) coated glass substrate that has been subsequentially functionalized with: a doctor bladed and cured anatase TiO_2 layer; silanation with ethnyltrimethylsilane; covalently bound iron (II/III) tetratolylporphyrin via Sonogashira coupling. In this poster, we specifically look to improve yields of the iron porphyrin species by following a one-pot procedure (Sun, Molecules, 2011) which synthesized tetratolylporphyrin and subsequently iron (II) tetratolylporphyrin with an expected yield of 28.7%. Previously, this was a multi-step process (Adler, J.Org. Chem, 1967) in which tetratolylporphyrin was initially synthesized in one reaction and then moved to another pot to be metallated with iron (III) chloride in a second reaction. The two processes combined produced an overall yield of just 4.99%. Synthesized materials were characterized by thin layer chromatography, NMR, and UV/visible light. Our criteria for improvement in the synthetic methods are increased overall yield, time to arrive at pure product, and consistency of response between functionalized electrode materials.

This work was supported by RI-INBRE

Toward thiocyanate detection in saliva: Machining TiO_2 application to FTO substrates

Antal Lee, Julie Gerstner & Clifford Murphy

Chemistry, Roger Williams University, Bristol, RI

Thiocyanate concentration in saliva has been suggested as a chemical marker for a screening analysis for oral and gastric cancers. In this project, we seek to adapt an existing technology for the electrochemical detection of thiocyanate in seawater to application as a screening tool for use as part of a routine dental exam. The chemosensor electrode is comprised of a fluorine-doped tin oxide (FTO) coated glass substrate that has been subsequently functionalized with a doctor-bladed and cured anatase TiO_2 layer, silanation with ethynyltrimethylsilane, and covalently bound iron (II/III) tetratolylporphyrin via Sonogashira coupling. In this poster, we specifically look to optimize the blading of the TiO_2 layer by developing a machine process for applying the layer in a more uniform and repeatable manner. Typically, each substrate has been hand-crafted individually. Lacking the proper mechanical advantage, hand-blading yields sequentially inconsistent electrodes by producing uneven TiO_2 layers. Blading each substrate by hand limits the scalability of electrode production in the lab. We designed a blading machine to combat this by implementing slide racks with ball bearings to limit the motion of the blade to one axis, and a handle to ergonomically apply pressure onto the substrates while blading. This allows the coating of smooth, homogeneous layers across multiple substrates simultaneously. The criteria for improvement in the fabrication of the electrode layers are consistency of the layer thickness and appearance from one electrode to the next, contact angle measurements for the TiO_2 layer both before and after silanation, and consistency of response between functionalized electrode materials.

This work was supported by RI-INBRE

Synthesis and Reactivity of Iron Tricarbonyl Complexes Containing Redox-Active Ligands

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The objective of this project is to synthesize and characterize iron complexes that contain redox active ligands. Such complexes can potentially be used as catalysts for the reduction of carbon dioxide, to form carbon monoxide, formate, methanol, or methane, or the reduction of protons, to produce hydrogen gas. The development of new catalysts for these reactions is important because they could contribute to a decrease in CO_2 levels in the atmosphere. We have synthesized iron tricarbonyl complexes that contain either α-diimine (ArDABMe) or iminopyridine (ArImMePy) ligands, in order to assess the effect of varying the aryl substituents on the electronic structure of the complexes. The complexes $Fe(PhImMePy)(CO)_3$ (Ph = phenyl, Me = methyl) and $Fe(2,6iPr2PhImMePy)(CO)_3$ (2,6iPr2Ph = 2,6-diisopropylphenyl, Me = methyl) have been synthesized and characterized by IR, NMR, and single crystal X-ray diffraction, and their structural and redox properties were compared. We also explored reactions of these complexes with the reducing agent potassium graphite, KC8, and a variety of phosphine ligands, in order to increase the electron density in the complexes. By doing so, we seek to determine the effect on the electronic structure of the complexes and to make the complexes more susceptible to reaction with protons or carbon dioxide. Based on IR spectral analysis, $Fe(PhImMePy)(CO)_3$ and $Fe(2,3,4,5,6-F5PhDABMe)(CO)_3$ are reduced by KC8, as evidenced by the decrease in carbonyl stretching frequencies. In the presence of alkyl and aryl phosphines and trimethylamine N-oxide, a decarbonylating agent, $Fe(PhImMePy)(CO)_3$ and $Fe(3,5-CF3PhDABMe)(CO)_3$ undergo substitution of a carbonyl ligand by phosphine. These species have been detected by IR and 31P NMR spectroscopies. Attempts to characterize these complexes structurally by single crystal X-ray diffraction are ongoing.

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Synthesis of Beta-Carboline Atropisomers

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β-Carbolines are nitrogen-containing small molecules that have been shown to have significant biochemical activity, including anticancer, antibacterial, antifungal, and antipsychotic properties. If an aryl ring is appended to the 1-position, these molecules have the potential for restricted rotation around one of their C¬¬–C sigma bonds that can lock these molecules in two distinct, non-superimposable mirror image forms called atropisomers. Accessing one or the other of these forms efficiently has remained a challenge in the field of organic synthesis. The energetic barriers to rotation (ΔGrot, kcal/mol) for two 1-naphthyl-β-carboline derivatives have previously been determined computationally and experimentally, confirming their long-lasting configurational stability. However, our previous method used a racemic synthesis of the β-carboline scaffold. Current efforts have been directed towards developing an asymmetric synthetic route, producing one atropisomer only. Here we report an optimized, six-step synthesis of the 2-substituted-1-naphthyl-β-carboline scaffold. Emphasis was placed on optimization of the transition metal-catalyzed [2+2+2] cyclotrimerization reaction, including catalyst complexation and solvent effects. Further studies will entail the use of a chiral phosphine to promote asymmetric induction, which we will measure by chiral HPLC. We plan to determine the product's absolute configuration via electronic/vibrational circular dichroism and extend this synthesis to other 2-substituted-1-naphthyl substituents.

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Site-Directed Mutagenesis and Purification of R71L and R71K Mutants of StarD6 Protein

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Alzheimer's disease (AD) is a neurodegenerative disease affecting cognitive functions and memory and is the leading cause of dementia. The physical manifestation of AD is the formation of beta-amyloid plaques and tau tangles in the brain, but the biochemical mechanisms that give rise to these changes are relatively unknown. Recently, it was reported that a single nucleotide polymorphism (SNP) near the gene for the StAR related lipid transfer domain containing 6 (StarD6) protein could be correlated with AD development. StarD6 is expressed in the brain and testes, binds cholesterol and testosterone, and may be neuroprotective. Herein, we describe a method of site-directed mutagenesis and protein purification to synthesize and purify R71L and R71K mutants of StarD6. We hypothesize that the R71L mutation will decrease the protein's ability to form hydrogen bonds with its ligands, while the R71K mutation will maintain the protein's ability to form hydrogen bonds and will not affect its ligand-binding abilities. Further studies are necessary to determine the mutant proteins' affinity for cholesterol and testosterone and to determine how their binding constants compare to wild-type StarD6.

This work was supported by RI-INBRE

Protein Purification and Differential Scanning Fluorimetry of Wild Type StarD6

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Alzheimer's (AD) is a neurodegenerative brain disorder that is the leading cause in dementia. It has been proven that amyloid beta and tau proteins are involved in the later stages of AD, but there is very little knowledge on the early molecular mechanisms that occur. Recent genetic studies have suggested that a single nucleotide polymorphism (SNP) near the gene for the protein StAR related lipid transfer domain containing 6 (StarD6) is associated with risk for developing Alzheimer's disease. StarD6 is expressed in the testes, ovaries, and brain regions. StarD6 binds testosterone, cholesterol, and pregnenolone, it has also been proposed that this protein might bind cholesterol- derived neuroprotective steroids. We hypothesize that StarD6 plays a neuroprotective role, most likely by binding and delivering neurosteroids to their site of action. In order to test this hypothesis, we will site-direct mutagenesis along with differential scanning fluorimetry (DSF). We are currently optimizing the protein purification protocol established by Letourneau et al and the DSF assay developed by Bai et al for use with our protein of interest, StarD6.

This work was supported by RI-INBRE

Site-Directed Mutagenesis and Purification of StarD6 Mutants Y81H and Y81F

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Alzheimer's Disease, a neurodegenerative disease, is known to be caused by excess beta-amyloid plaques and neurofibrillary tangles of the tau protein. This disease damages the brain in areas including the hippocampus, which affects cognition, neuronal communication, behavior, and increases the likelihood of developing dementia. There is significantly less information on the initial molecular causes of this disease, but recent genetic studies focused on a single nucleotide polymorphism (SNP) near the gene encoding StAR Related Lipid Transfer Domain Containing 6 (StarD6) have shown a promising correlation with the development of Alzheimer's Disease. Considering its affinity in binding to testosterone and cholesterol, it is possible that StarD6 also binds to neurosteriods and delivers them to the degenerated areas of the brain as a form of neuroprotection. A 2016 study by Letourneau et al. focused on StarD6 and its ligand binding ability. While studying this protein's interactions with testosterone, it was suggested that certain amino acids are involved in ligand binding through the hydrogen bonds they form. One of the amino acids found to bind to the hydroxyl group of testosterone through polar interactions was the tyrosine at position 81. We hypothesize that site directed mutagenesis of key amino acids in StarD6 may alter the protein's ability to bind its ligands. In this study, we have focused on the tyrosine residue located at position 81 (Y81). Using site directed mutagenesis, we designed two mutations that would replace tyrosine, Y81H and Y81F. Substituting histidine for the tyrosine residue at position 81 (Y81H) is expected to maintain its hydrogen bonding capabilities, while the phenylalanine substitution for tyrosine (Y81F) is expected to decrease its hydrogen bonding capabilities. In turn, this may affect the protein's possible neuroprotective facilitation of neurosteroids to areas of the brain affected by Alzheimer's Disease. The current goal is to purify the proteins for these mutations and observe their ligand binding properties.

This work was supported by RI-INBRE

Comparison of Two Human Gut Microbiome-Derived Drug-Metabolizing Dehydrogenases

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Individuals have unique microbiomes, which can lead to various responses to drug therapies. Consequently, these varied responses can be dangerous and costly due to unpredicted adverse side effects. Personalized medicine aims to address this issue by focusing on an individual's genome and microbiome to provide tailored treatments. However, the molecular mechanisms by which gut microbes metabolize drugs have not received much attention. This problem is made more complicated due to the diversity of gut microbiomes. To address this problem, we have targeted several proteins expressed by human gut microbes *Eggerthella lenta*, *Clostridium scindens*, and *Bifidobacterium adolescentis*, that metabolize and alter drug structures. These enzymes include the 20-hydroxysteroid dehydrogenases (20-HSDHs) B0NC68 (a 20α-HSDH) and WP_008310233.1 (a 20β-HSDH), which change the structure of the host-produced steroid, cortisol. Therefore, we hypothesized that these enzymes affect drug metabolism on corticosteroids, therapeutics structurally similar to cortisol. We have successfully cloned, purified, and tested two 20-HSDHs of opposite stereospecificity. Both enzymes were found to modify cortisol and the anti-inflammatory drug prednisone, while the 20β-HSDH is more efficient and faster than the 20α-HSDH. This investigation serves as a springboard into the development of personalized medicine based on how a unique community of gut microbes modifies therapeutics, helping predict drug metabolism by knowing the makeup of an individual's microbiome.

This work was supported by RI-INBRE

Network Analysis of Gut Microbiome Azoreductases involved in Drug and Food Dye Metabolism

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Human gut bacteria produce enzymes that can metabolize drugs, which may lead to unintended side effects and differences in drug efficacy between individuals. Personalized medicine would alleviate this problem by making treatment dependent on an individual's genome and gut metagenome. However, this process is generally unpredictable, as the gut microbiome can vary between individuals and even over time. This process is also hindered as the hypothetical enzymes discovered in metagenomes are poorly annotated and their functions are undetermined. One such group of enzymes is the azoreductase (AzoR) family, flavin mononucleotide-dependent enzymes. Although no gut-derived azoreductase enzymes have been shown to reduce azo bonds in drugs, there is an AzoR produced by *Pseudomonas aeruginosa* with this drug-modifying activity. We have identified candidate genes in the gut microbiome using network analysis, which we predict to have similar drug-modifying activity. We have begun to express, purify, and test these diverse gut-derived AzoR proteins for their ability to metabolize different azo compounds. These substrates include azo-drugs, several FDA-approved food dyes, indicators, and other azo compounds. In examining the enzymatic function of azoreductases on different substrates found in human gut, we will expand the ability to predict an individual's metabolism based on their gut microbiome.

This work was supported by RI-INBRE

The Tracking Game: How Bluegill Move Their Fins Under Turbulence

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Neuromuscular disorders and injuries can cause impairment of local muscle control in humans, deteriorating quality of life. This study uses the dorsal fin erector muscles of bluegill (*Lepomis macrochirus*) as a proxy to study local muscular control and sensory perception to help humans in rehabilitation. The epaxial and spine erector muscles of the bluegill were implanted with double insulated electrodes quickly followed by the injection of one of three treatments: lidocaine (an analgesic drug), flaxedil (a muscle relaxant) or saline control. The fish were then exposed to turbulent or non-turbulent conditions, muscle activity was logged, and two high speed cameras recorded the top and side views of the fish in the tank. The dorsal view of the fish in the tank allows us to see the displacement of the fish through the depth of the tank which is not as visible in the lateral view. The video footage was analyzed to find the relative displacement and relative velocity of the spiny dorsal fin as well as the movement of the fish center of mass. The muscle activity was analyzed to find the burst duration and magnitude. It was found in control conditions turbulence caused higher fin displacement and relative velocity . This pattern was reversed under the influence of lidocaine and there was no difference under treatment with flaxedil. Based on the dorsal view, the relative displacement of the center of mass appears to increase under turbulent conditions as expected. Additionally, when treated with lidocaine or flaxedil, the fish appear to have less control over their stability when compared to the control. They are often found swimming on their side while turning rather than remaining upright. In turbulent conditions the erector muscles have longer burst durations under control conditions, similar durations under flaxedil and shorter durations under lidocaine. The magnitude of the erector muscles is higher in turbulent conditions except under treatment with lidocaine. Gaining a better understanding of local motor control and sensory perception may lead to creating human prosthetics which are easier to control and more stable.

This work was supported by RI-INBRE

Illuminating the Neural Pathways of the Spiny Dorsal Fin of Bluegill

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Perceiving sensory information accurately plays an important role in one's ability to react appropriately. The inability to modulate muscle contractions as a result of inaccurate sensory perception leads to loss of fine motor control and instability. The spiny dorsal fin of fish is thought to play an important role in stability and is particularly amenable to serve as a proxy for human prosthetics since it has a limited range of motion, moving mostly rostro-caudally to be erected or depressed. The aim of this study is to investigate the innervation of the spiny dorsal fin of bluegill (*Lepomis macrochirus*). We hypothesize that the spiny dorsal fin and the membrane surrounding the spines have free nerve endings and/or encapsulated mechanoreceptors that are used to detect flow perturbations, similarly to other fish fins. We postulated that afferent or sensory nerves would be smaller and more prevalent in the fin membrane than efferent or motor nerves. The spiny dorsal fins of four bluegill were isolated in spine pairs before following a fixative protocol. To visualize innervation, dorsal spines were stained with either anti-acetylated tubulin (AAT), calcitonin gene-related peptide (CGRP), or both, followed by a corresponding secondary antibody. AAT is known to stain both afferent and efferent nerves, while CGRP only stains afferent nerves. Samples were then imaged using a fluorescent microscope. Neural tissue was then identified, and nerve diameter was measured in four different regions (fin tip, mid-fin, fin base, and muscle) using NIS-Elements D and ImageJ. AAT-stained specimens were observed to have clusters of sensory cells and innervations within the fin membrane as well as neuromuscular junctions and afferent nerves within the muscle. CGRP-stained specimens were observed to have bundles of sensory cells and free nerve endings throughout the fin membrane, but primarily at the fin tip, connected to the fin base by CGRP-stained innervation. The average nerve diameter of sensory neurons was found to be smaller than that of motor neurons and these nerves were more abundant within the fin membrane than in the muscle. Our measurements support our hypothesis that sensory nerves are smaller than efferent nerves in diameter and that these are the only nerves present in the fin membrane. By elucidating how this simple joint is innervated, we can model sensorimotor integration which can be used in bioinspired solutions for more responsive human prostheses.

This work was supported by RI-INBRE

The Effects of Dehydration stress on W118 and Sod null *Drosophila* mutants through biochemical Assays and MDA readings

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Superoxide dismutase (SOD) is an enzyme found in all organisms and has many key uses, one of which is the breakdown of dangerous oxygen molecules within the cell. During this lab we used *Drosophila* with a modified SOD gene in order to test its resistance to dehydration stress compared to a control group of regular W118 flies. We hypothesized that flies infected with a mutated SOD gene would have lower survival and an increased rate of oxidative damage due to the lack of oxygen breakdown within their cells. To test this hypothesis, we compared the rates of survival between W118 and mutant sod null *Drosophila* under dehydrative stress. We then tested the amount of water stored within the two different genotypes by weighing the flies and dehydrating them for 48 hours in an oven. Finally we tested how the two genotypes responded to oxidative damage by running several assays in order to test Malondialdehyde (MDA) levels. Our results showed that W118 flies were able to survive over 24 hours under dehydration conditions, whereas SOD null flies survived less than six hours. We found that both groups' body weight was roughly 76% water. The biochemical assays revealed that the dehydrated flies had double, sometimes even triple the amount of MDA compared to flies that were left in an agar solution. These results explain why *Drosophila* with a mutated SOD gene have a shorter lifespan, as they are in constant need of water to avoid dehydration. Flies without water also have dangerously high levels of malondialdehyde (MDA) causing oxidative stress and severe health complications. Further research into the effects of MDA and oxidative damage will allow us to study the role of the SOD1 enzyme and why it is vital in all organisms.

This work was supported by RI-INBRE

Determining the Effects of H71Y in the Background of *Drosophila melanogaster* Autophagy Mutants on the Redox State of Mitochondria.

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Amyotrophic Lateral Sclerosis (ALS) is a progressive neurodegenerative disease that is characterized by the degeneration of motor neurons, the abnormal aggregation of proteins, and muscle wasting. ALS may begin in the first or second decade of life and is known to cause more than 1 in 500 deaths in adults. Cases of ALS caused by SOD1 or FUS mutations display abnormal SOD1 or FUS proteins respectively, and H71Y mutants exhibit recessive ALS- associated phenotypes such as a shortened life span and some dosage-sensitive toxic gain of function effects. Autophagy is a critical pathway for the elimination of damaged organelles and protein aggregates in cells, whose defects have been associated with mutations of ALS-linked genes including SOD1. To understand the interactions between SOD1 oxidative stress pathways and autophagy, we developed multiple lines of autophagy mutants with different numbers of H71Y alleles in the background and analyzed the redox states of their respective mitochondria. Pre-existing *Drosophila melanogaster* stocks were crossed to generate lines expressing autophagy mutant, mitorhoGFP, and H71Y SOD1 mutant phenotypes. Male and female autophagy mutants were isolated with one or two copies of the H71Y gene by dominant selectable markers and dissected in PBS. The ventral nerve cords from these samples were fixed, mounted, and imaged through confocal microscopy. Using ImageJ, we created ratios of the 405 and 488 wavelength expressions of mitochondrial damage for each sample to compare oxidative damage in the process of mitophagy. We found that the H71Y SOD1 mutation enhances the phenotype of autophagy mutants as indicated by higher signal ratios between the 405 and 488 wavelengths, which displays a greater level of mitochondrial damage. Future studies will be aimed at visualizing the enhancing effects of knocking down lamtor (62202), a critical gene involved in the autophagy pathway, in a humanized sod mutant background.

This work was supported by RI-INBRE

Isolation and Identification of Manganese-Reducing and Oxidizing Bacteria from Rhode Island Wetland Soils

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Manganese-reducing bacteria play an important role in the biogeochemical cycling of manganese. In this work, we used a functional assay for Mn reduction to enrich and screen for Mn-reducing bacteria in RI wetland soils. We performed this work using indicator of reduction in soils (IRIS) films that we have previously used to quantify Mn-reducing activity in the soil. These films are coated with birnessite paint which is comprised of oxidized manganese species. The reduction of this brown paint solubilizes the birnessite and reveals the white PVC film. We isolated individual bacteria capable of removing birnessite paint from manganese IRIS films, and we identified these isolates using 16S ribosomal RNA gene sequencing. We identified bacteria from seven genera including *Bacillus*, *Pseudomonas*, *Acinetobacter*, *Lysinibacillus*, *Burkholderia*, *Vogesella*, and *Defltia*. Though some of these isolates have been previously implicated as manganese reducers, we were surprised to find that approximately half of our isolates have been identified as manganese oxidizers. Though each of these isolates is capable of removing birnessite paint from IRIS films, we have found that paint removal is greatly accelerated by the presence of both manganese reducers and oxidizers. Because the birnessite paint contains both Mn^{3+} and Mn^{4+} species in a crystalline lattice, our working model to explain this synergy is that manganese oxidizers and reducers work together to disrupt the structure of the birnessite lattice. This could explain our observation that removal of birnessite paint from films occurs more rapidly in wetland soil samples than in laboratory monocultures.

This work was supported by RI-INBRE

Lipid Metabolism Dysregulation in FTD Affected *Drosophila*

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Metabolic reprogramming is a common hallmark of many diseases. In recent years, the focus on metabolic change in cancerous tissues has increased. However, fewer studies have investigated the metabolic shifts in neurodegenerative diseases. Metabolic reprogramming in neurodegenerative diseases has been well documented and glucose uptake is even used as a key diagnostic indicator for some of these diseases. We are utilizing an established *Drosophila* model of the neurodegenerative disease, Frontotemporal Dementia (FTD), to investigate dysregulation in lipid metabolism. To do so we will be using immunofluorescence to detect lipid accumulation, metabolite assays to detect changes in triglycerides, and quantitative PCR to measure the expression of metabolic enzymes. The goal of this project is to determine if lipid metabolism could be a potential target for the treatment of FTD.

This work was supported by RI-INBRE

Developing a protocol for microinjection of rotifer *Brachionus plicatilis* eggs

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Rotifers have many characteristics that make them suitable for research in the biology of aging, including parthenogenic reproduction, a natural life span of only two weeks, and a somatic cell mitosis that is complete before the adult emerges from the egg. Of the >2,000 species of rotifers documented, *Brachionus plicatilis* is commercially useful as a nutritional food source for larval fish. The *B. plicatilis* annotated genome was published in 2019, opening the doors for experimental targeted genetic mutations. Among a small handful of publications, there appears to be a significant degree of variability in the efficiency of genetic modification and in egg viability following the introduction of genetic material in the rotifer.

Microinjection is a form of mechanical delivery for transferring biological materials into living cells and has been practiced for over a century. Here, I work to develop a protocol for the microinjection of amictic rotifer eggs. Our protocol consists of first harvesting the amictic eggs from the rotifers, holding the rotifer egg in place with a holding pipet, and penetrating the egg chorion using a quartz microinjection needle. We determine whether an injection was successful by injecting a fluorescent FITC-dextran compound and assessing egg survivability at different injection pressures and times to optimize the delivery of material to rotifer egg cells.

Our preliminary data will guide our experiments using a CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) nuclease targeted to the rotifer insulin-like growth factor receptor (IGFR1) gene. We will microinject lipophilic vesicles containing CRISPR-Cas12a complexed with a gRNA targeting the *B. plicatilis* IGFR1. I hypothesize that non-homologous end joining following a double-strand DNA break at the IGFR1 locus will result in a functional gene deletion that will extend the rotifer lifespan, similar to what is seen in deleting the IGFR1 ortholog in *C. elegans*. We will also test whether a repair template containing GFP (green fluorescent protein) under the control of a rotifer constitutive promoter will allow for the integration of the repair template into the mutation site via homology-directed repair. This genetic modification has many potential applications, such as enhancing diagnostic and research techniques regarding both larval fish, as well as CRISPR and genetic editing techniques.

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Analysis of Gnao1 in 3T3 Cells

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Caenorhabditis elegans (roundworm) and *Saccharomyces cerevisiae* (yeast) shared a common ancestor approximately 1.3 billion years ago, making these two model organisms ideal for the study of evolutionary conservation. These two model organisms have also been at the forefront of discovery of key cellular pathways that modulate the rate of aging. If an aging gene identified in yeast is shown to have a conserved ortholog in *C. elegans*, we can hypothesize that this gene or pathway may be conserved in higher eukaryotic organisms such as mammals.

Genetic screens in yeast identified the G protein GPA1 as a gene involved in aging (deletion of GPA1 causes an extension in yeast life span). G-proteins are involved in transmitting extracellular signals and then sending them through pathways inside the cell. Our lab has found that RNA interference against a GPA1 ortholog in *C. elegans* (gpa-7) also extends the roundworm median life span by 57%. These finding suggest that mammalian orthologs of G proteins might be involved in aging as well. The mouse Gnao1 protein is 56% identical and 72% similar to that of the gpa-7, representing a high degree of amino acid sequence conservation. This research aim explores whether knockdown of Gnao1 and a mouse 3T3 cell line exhibits anti-aging characteristics.

To do this we use short interfering RNAs (siRNA) packaged in lipophilic vesicles (Lipofectamine) to silence the expression of Gnao1 in 3T3 cells. siRNA binds to the RNA induces silencing complex which surveys messenger RNA (mRNA) with complementary sequence. RISC degrades these target mRNA leading to silencing of the expression. In this study we have seen efficient knockdown of Gnao1 in the 3T3 cells using siRNA. Future work involves inducing a stress response where we will test oxidative stress (with paraquat testing) and heat shock.

This work was supported by an externally funded grant

Long-Lived Yeast Strains Show an Increased Tolerance to Stress

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Yeast is an excellent model organism for the study of aging due to its short natural life span, relative ease of genetic manipulation, and amenability to high throughput screening. Many gene mutations identified in yeast that mediate longevity are conserved in higher eukaryotes, such as roundworms, fruit flies, and mice. In addition, most long-lived mutant strains also show an extension in the period of 'healthy living,' meaning that behaviors and phenotypes associated with youth are observed at higher frequencies in old age in long-lived mutant strains. However, it remains unclear whether any single discrete genetic change associated with an extension in life span impinges upon one or more clusters of pro-longevity cellular pathways. Here, we test the hypothesis that long-lived yeast strains are more adept at mitigating normal cellular damage that naturally accumulates with time.

Several yeast mutations are associated with exceptional chronological longevity, including deletion of the VPS51 gene (Vacuolar Protein Sorting gene 51). VPS51 is a component of the GARP (Golgi-associated retrograde protein) complex that is involved in the recycling of proteins from endosomes to the late Golgi. In this research aim I test the hypothesis that long-lived yeast mutants are tolerant to different forms of stress, including heat and other environmental exposures. Our preliminary data indicate that the long-lived VPS51 deletion strain tolerates heat and acid stress better than the wild-type strain.

Several cellular mechanisms may explain this differential. One mechanism may involve the stress-induced translocation of transcription factors MSN2 and MSN4 (multicopy suppressor of SNF1 mutation 2 and 4) from the cytosol to the nucleus. These proteins help regulate a global stress response of S. cerevisiae that includes the upregulation of autophagy and an increase in the expression of proteins that reduce reactive oxygen species. Future plans include testing whether the VPS51 mutant shows an increase in nuclear translocation of GFP-tagged MSN2 and MSN4 proteins. Alternatively, given the nature of VPS proteins in protein trafficking, we hypothesize that the unfolded protein response (UPR) may be highly active in the VPS51 delete strain. We plan to test this hypothesis by looking at splicing intermediates of the Hac1 mRNA, which is known to undergo splicing while the UPR is activated.

This work was supported by an externally funded grant

Life Span Machine Helps Identify Longevity Promoting Pharmaceuticals in Rotifers

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Aquatic rotifers are a relatively new model organism to the biology of aging. Rotifers possess many characteristics that make them suitable for aging studies, such as a short natural life span and parthenogenic reproduction. *Brachionus plicatilis* is a free-swimming rotifer that feeds upon single-celled algae in brackish water. We have adapted *B. plicatilis* to laboratory husbandry and measured their life span under various feeding conditions. We also find that the *B. plicatilis* life span can be extended by rapamycin at concentrations as low as 5µM, which is approximately 20-fold less than the required dose to extend the life span of the roundworm *C. elegans* (100µM).

In collaboration with the Nathan Shock Healthy Aging and Longevity Institute (University of Washington) we have developed a high-throughput robotic life span machine that can assay the life span of rotifers. The 'Rotibot' consists of an optical table capable of accommodating twelve, 12-well plates (for a total of 144 wells) and a movable lighting and camera system that images each well along X- and Y-dimensions. Images are captured twice for each well at 5 second intervals, and OpenCV computer vison and machine learning software scores movement of swimming rotifers by comparing images (pixels) across the 5 second interval. Additional computer algorithms then generate Kaplan-Meier life span curves by tracking the loss of active rotifers in each well. Since our previous data on rapamycin suggests that pharmaceuticals are bioavailable at low culture concentrations, we propose using the Rotibot in conjunction with a screen of a FDA-approved drug library at both high and low concentrations. We propose that the rotifer model will allow for the identification of longevity-promoting pharmaceuticals that might have been missed in other model organisms.

This work was supported by RI-INBRE

Longevity phenotype of gpa-7 knockdown in *C. elegans* is mediated through the reduction of second messenger cAMP

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Loss-of-function screens have been used in studies of aging, as deletion of a gene that promotes cellular aging could be identified by a resultant increase of organismal lifespan. Bacterial collections of RNA interference (RNAi) for *C. elegans* are commercially available and provide an accessible way of performing comparative lifespan analysis. Our previous work with RNAi has revealed that knockdown of a g-protein, specifically named gpa-7 in C. elegans and gpa-1 in *S. cerevisiae*, resulted in a significant lifespan extension in both model organisms.

G-proteins are trimeric GTP-binding signaling molecules that are composed of α-, β-, and γ- subunits. When a signaling molecule binds to the G-protein-coupled receptor, it exchanges GDP for GTP on the α-subunit. Activated G-α-subunits activate the enzyme adenylyl cyclase, which converts ATP to cyclic AMP (cAMP), while activated βγ-subunits activate phospholipase. cAMP binds to regulatory subunits of Protein Kinase A (PKA). We hypothesize that reducing the expression of gpa-7 causes a change in the G-protein mechanism that results in lifespan extension. The first possibility is that reducing the expression of gpa-7 effectively inhibits the production of cAMP, which would then prevent the activation of PKA. The other possibility is that inhibition of the α-subunit results in increased activity of the βγ-subunits. To test these theories and provide further evidence that loss of G-α-subunit activity extends lifespan, we plan to test the effects of gene knockdown on other genes that are important in the production of cAMP, such as acy-1 and acy-2, which affect adenylyl cyclase. We will also examine the differences between empty vector (EV) and R10H10.5 (gpa-7 RNAi) treated CF1330 worms via immunofluorescent microscopy. These worms were modified by the Caenorhabditis Genetics Center (CGC) to bind green fluorescent protein to the DAF-16/FOXO transcription factor and will be stained with DAPI to look for colocalization of DAF-16 and the nucleus. In addition, we plan on exposing N2 worms treated with EV and R10H10.5 to various stressors, such as heat shock and paraquat exposure to see if there is increased tolerance to these stressors in the R10H10.5 treated worms. We are partnering with researchers at the University of New Hampshire to use RNAseq on genetic material from both EV and R10H10.5 treated worms to map downstream effects of discrete genetic changes and gain more insight into how this mechanism works.

This work was supported by an externally funded grant

Quantification of an antibacterial agent found in *Liriodendron tulipifera* twigs and comparison of the metabolites produced in related species of *Liriodendron*

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Infections caused by *Staphylococcus aureus*, a common human bacterium found on people's skin, pose a significant health burden, especially in regard to hospital-acquired infections. Previous research has shown that excessive use of antibiotics can result in the development of drug resistant strains of this gram-positive bacterium. In the case of drug-resistant pathogens, there is a need for newly developed antibiotics to combat these microbes. Medicinal plants are valuable resources for developing new drugs in light of their cultural and historical use as traditional medicines. Currently, at the University of Rhode Island, an extract library from medicinal plants - the Principal Rhode Island Secondary Metabolite Library (PRISM), is used to screen plant extracts against biological endpoints (e.g., antibacterial screening). One such extract from the tulip tree (*Liriodendron tulipifera*) showed the ability to inhibit the growth of methicillin-susceptible *S. aureus* (MSSA). Further studies isolated and identified the antibacterial component as the sesquiterpene lactone laurenobiolide. Intriguingly, this tree was used historically by indigenous Americans in order to combat malaria, bacterial infections, fevers, and many other maladies. Our initial studies showed that the antibacterial metabolite produced by the tulip tree was most abundant in the twigs. Our current research quantified the amounts of the antibacterial agent in the different parts of the plant and confirmed that the twig was the most abundant source of laurenobiolide. Additionally, we determined the spatial distribution of laurenobiolide in the twigs. The greatest quantity of the active compound was found in twig growth from recent years, which means that the active compound was mainly in the newly growth twigs versus the older growth. Lastly, we compared the metabolites produced by closely related species of the tulip tree, *L. tulipifera*, *L. chinense*, and *L. tulipifera x chinense* Hybrid. Results also showed that L. chinense does not contain the active compound lauernobiolide and its extract was not active against MSSA. The hybrid contained a small amount of the compound. As the extract library expands, future efforts will go towards understanding if laurenobiolide can be developed as an antibiotic. Regarding this development, research will need to determine the potency and selectivity of laurenobiolide against many strains of pathogenic bacteria.

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Determining the triggers of domoic acid production using cultivated strains of *Pseudo-nitzschia* and an untargeted metabolomics of phytoplankton and Narragansett Bay mussels.

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Multiple species of *Pseudo-nitzschia* that grow in Narragansett Bay produce domoic acid, a neurotoxin that can cause amnesiac shellfish poisoning in humans and other marine life. This poisoning can cause cognitive problems, permanent short-term memory loss, and even death. Toxic algal blooms have resulted in multiple shellfish closures in 2016 and 2017 in Narragansett Bay, Rhode Island. There is uncertainty as to the environmental trigger or triggers that cause domoic acid production in *Pseudo-nitzschia*. We are evaluating how parameter manipulation in culture affects domoic acid production. We were sent cultures with various limited nutrients from the University of Southern California. We performed aqueous methanol extractions (90% water, 10% methanol) and quantified domoic acid concentrations using sensitive and selective LC-MS/MS methods. We observed that high and low concentrations of CO_2 in cultures elicited increases in domoic acid production, although these increases were not statistically significant. However, additional studies will probe this relationship further. Following the parameter manipulation experiments, we conducted untargeted metabolomics analysis on cultured *Pseudo-nitzschia* strains isolated from Narragansett Bay and mussels collected during high *Pseudo-nitzschia* cells counts. While we did not detect domoic acid in the mussels, we did annotate several pharmaceutical, cosmetic, and agricultural products, which provokes intriguing questions with respect to the human health relevance of these molecules in consumed seafood.

This work was supported by RI-INBRE

Acidic pH Conditions Improve ABH3 Oxidation of Epigenetic Marker, 5-Methylcytosine and Its Derivatives

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5-Methylcytosine (5mC) is an epigenetic modification of DNA that has been implicated in regulation of transcription through different mechanisms. In mammals, methylation at the 5-position of cytosine occurs mainly within the promoter region of a gene on CpG islands, or long stretches of repeating, cytosine-guanine dinucleotides. One of the key functions of 5mC in this context is to ensure a silenced gene remains inactive. Previously, the AlkB-family enzymes, such as ABH3, have been shown to oxidize 5mC and its derivatives in DNA in vitro. Furthermore, AlkB-family enzyme oxidation of DNA modifications can be modulated by numerous environmental factors including pH conditions. One of the hallmarks of cancer pathology is the abnormal pH conditions observed within the tumor microenvironment. This phenomenon is especially relevant to ABH3, as it is overexpressed in certain forms of cancer. If abnormal pH conditions ultimately lead to a change in promoter methylation status for a proto-oncogene or tumor-suppressor gene via ABH3, malignant transformation of a healthy cell into cancerous one will occur. This study investigated the in vitro effects of different pH conditions on the ability of ABH3 to oxidize 5-methylcytosine in DNA. The impact of pH conditions on ABH3-mediated oxidation of methylated DNA was quantified by reverse-phase HPLC. Our findings show that pH significantly improves the ability of these enzymes to oxidize epigenetic marks, especially under somewhat acidic conditions. This pH effect indicates that under cancer conditions the epigenetic pattern will be different than normal cells due to ABH3-mediated oxidation of 5mC.

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Inhibitory effects and binding affinities of epigallocatechin gallate with elastase

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Epigallocatechin gallate (EGCG) is one of the most abundant polyphenols from green tea (*Camellia sinensis*). EGCG is also known for its antioxidant and anti-inflammatory properties in a series of diseases. However, it's still unclear if EGCG exerts beneficial skin effects. Elastase is one of the major proteases responsible for the degradation of tissue elastin, which is an essential protein for maintaining skin mechanical structures. Inhibitors of elastase, especially from herbal extracts, have been reported to show ameliorative effects against elastase-associated wrinkle formation. In this study, we evaluated EGCG's anti-elastase activities by using in vitro and in silico assays. Elastase inhibition assay was performed to determine the anti-elastase effects of EGCG as well as its inhibitory rate. In addition, computational docking was conducted to investigate the binding mode and binding affinity between EGCG and elastase. Our results showed that EGCG, at a concentration of 100 µM, displayed 40% inhibitory percentage against elastase. In addition, computational docking demonstrated that EGCG had an estimated free binding energy of -5.72 Kcal/mol with an estimated inhibition constant (Ki) of 64.08 µM. In conclusion, EGCG is an elastase inhibitor, which may contribute to its overall skin beneficial effects and support its potential for cosmeceutical applications.

This work was supported by RI-INBRE

Identification and Initial Characterization of Novel Synaptic Genes in *Drosophila*

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The synapse is a crucial part of the nervous system. It's the site of communication between neurons or neurons and muscles, providing neural control of physical movement. The goal of my project is to identify new genes involved in synapse development and or function. Uncharacterized genes that share a developmental transcriptional profile with known synaptic genes may also have roles in synapses formation or function. To test this, we knock out candidate synaptic genes using CRISPR. We then assessed synaptic growth at the neuromuscular junctions using immunohistochemistry and confocal imaging. To determine if synaptic function was disrupted, we used larvae crawling and dissection assay to evaluate locomotion. This investigation may reveal new genes that regulate synapse formation and/or function.

This work was supported by RI-INBRE

Investigating behavioral sequences in obsessive-compulsive and hoarding disorders

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Classified as an executive function, sequence processing allows people to perform a series of tasks to accomplish overarching goals. Obsessive-compulsive disorder (OCD) is a condition that is characterized by obsessive thoughts accompanied by repeated behaviors, known as compulsions. Hoarding disorder (HD) is classified by the difficulty to discard possessions regardless of their real value. People with both disorders have been shown to have deficits in executive function and may have difficulty completing tasks towards a sequential goal. Though these disorders are similar (HD has been considered a subtype of OCD), there are key distinctions between the two. Previous studies reported more deficits in HD in non-sequential executive functions than in OCD. Sequence processing has not been tested in these populations. Therefore, we tested the hypothesis that those with HD will experience greater sequential deficits compared to participants with OCD. Participants performed a task where they completed a sequence of remembered judgements performed on simple stimuli (e.g. color, color, shape, shape). We found that participants with HD had greater deficits in the time to initiate sequences (i.e., longer reaction times) when their symptoms were more severe, measured by a clinical measure of incompleteness. In contrast, participants with OCD did not show this correlation and were significantly different from patients with HD. These results suggest that symptom severity plays a significant role in sequential deficits in HD and not in OCD. In the future investigating symptomatology may help predict sequence processing deficits in HD and other psychiatric disorders.

This work was supported by RI-INBRE

Distinct dopamine microcircuits underlying alcohol-induced locomotor activity and alcohol associated memories in *Drosophila melanogaster*

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Drugs of abuse such as alcohol disrupt dopaminergic reward pathways, leading to maladaptive goal-directed behaviors. In both mammals and *Drosophila*, evidence suggests that dopamine also mediates ethanol-induced locomotor activity, however, it is unclear if dopamine neurons identified as important for reward are also important for alcohol-induced locomotor activity. Using flyGrAM, an automated group activity monitor, we investigated whether distinct populations of DANS are differentially recruited to support ethanol-induced activity in a dose-dependent manner. We show that both the PAM and PPL1 subsets of dopamine neurons innervating the Mushroom Body, a memory encoding central brain structure, play dynamic roles in modulating alcohol-induced locomotor activity. Inactivation of dopamine neurons had the most profound effect during the later stages of alcohol intoxication and at higher doses, suggesting that more dopamine neurons are recruited at higher doses to counter the sedating effects of alcohol. Interestingly, we found subsets of dopamine neurons important for alcohol reward memory retrieval did not modulate low dose alcohol-induced locomotor activity suggesting that testing at higher doses of alcohol is necessary to rule out their role in modulating activity at lower doses. We then conducted high content behavioral analysis of individual flies using FlyTracker, a post processing computer vision software which suggests that distinct dopamine neurons modulate activity metrics such as velocity, angular velocity, and distance to walls of the arena. Future work will employ Ctrax to examine both individual and social behavioral features to better characterize the effects of dopamine neurons on alcohol induced activity.

This work was supported by RI-INBRE

Neural Mechanisms Underlying Maladaptive Reward Memories in *Drosophila*

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Alcohol use disorder (AUD), is often associated with long lasting preference for cues associated with alcohol which persist in the face of aversive consequences. Knowledge of the circuitry mechanisms that underlie the encoding and expression of alcohol-associated memories is critical to understanding why these memories are resistant to change. *Drosophila* is an impressive model organism to explore circuitry mechanisms associated with alcohol-associated memories because of 1) its lower complexity 2) the similarity that exists in reward circuitry function 3) the availability of neurogenetic tools that permit dissection of memory circuits with impressive spatial resolution. Like mammals, *Drosophila* develops a long-lasting preference for alcohol-associated cues. Preferences persist despite aversive consequences such as bitter taste or shock, suggesting that *Drosophila* alcohol-associated memories are similarly inflexible. Previous work identified the circuit required for the acquisition and retrieval of alcohol reward memories. Specifically, acquisition requires population-level dopamine activity and activity of this population increased in response to intoxication. However, an understanding of how dopamine neural dynamics change across multiple intoxicating experiences is still lacking. We will utilize the genetic tractability of *Drosophila* and a newly developed tool that permits visualization of extracellular dopamine concentration, and two-photon microscopy to investigate how dopamine dynamics are altered by alcohol. Recording from awake and behaving flies during the first, second, and third cue alcohol association session will provide an opportunity to capture the precise changes in dopamine neural dynamics while flies learn to associate odor cues with alcohol intoxication. We hypothesize that alcohol-associated memories are unusually enduring because alcohol engages significantly more dopamine neurons across sessions. Thus we expect to see increased extracellular dopamine concentrations as measured by increased fluorescence in the MB lobes. These changes in dopamine neural dynamics likely contribute to the stability of alcohol reward memory and drive subsequent reward acquisition responses.

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Mapping neural circuits important for alcohol reward memories in *Drosophila*

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Alcohol addiction constitutes one of the most serious public health problems worldwide. Despite its devastating impact, there are few effective treatments. Circuit based approaches to treat addiction provide a powerful opportunity to develop more specific and effective treatments. However, circuit complexity has made it difficult to achieve a comprehensive understanding of the mechanisms by which memory circuits are altered to create enduring preferences for alcohol associated cues. *Drosophila melanogaster* is an opportune model organism to address these challenges because of the availability of neurogenetic tools that permit dissection of heterogenous circuits with exact temporal and spatial resolution. Although only 100,000 neurons comprise the central nervous system of *Drosophila* melanogaster, the neural circuitry underlying reward is remarkably complex and similar to mammals (Scaplen and Kaun, 2016). Like mammals, *Drosophila* show robust preference for cues associated with alcohol intoxication. Recently using a set of highly specific GAL4 and split-GAL4 driver lines in combination with thermogenetics, in-vivo calcium imaging, and genetic anatomical tracing tools, we identified circuits important for alcohol reward acquisition and retrieval. Acquisition of alcohol reward associations require population level dopaminergic modulation of the mushroom body, a learning and memory brain region. The expression of these memories, however, requires two anatomically distinct microcircuits which converge in downstream structures. The current project seeks to understand how activity across these circuits change across time to drive alcohol associated preference. Using a binary expression system to target neurons important for alcohol reward and a newly developed transcriptional reporter of intracellular calcium (TRIC), we measured neuronal activity following multiple intoxicating alcohol-associated sessions. We hypothesize that neurons important for alcohol associated memories will dramatically increase their activity across time as compared to odor controls. These experiments will inform functional experiments aimed at investigating the functional connectivity of MB circuits important for alcohol reward. Ultimately, understanding general circuitry principles described first in *Drosophila* will provide insight to how alcohol co-opts mammalian circuits to create enduring preference for alcohol and drive maladaptive choice.

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Cognitive impairments in a mouse model of bipolar disorder

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Cognitive deficits are a core symptomatic category of neuropsychiatric illness. In fact, they are predictive of disease outcome and quality of life. Despite the importance of these cognitive deficits, they are understudied and undertreated compared to other symptomatic categories. This is especially true in bipolar disorder (BD), partly due to a scarcity of valid rodent models. One cognitive symptom that is impaired in BD patients is cognitive flexibility, a type of executive functioning that is found to be impaired across all mood states and ages in BD. Also disrupted is recognition memory, which is the ability to identify familiar events, objects, or people. In this study, we make use of a transgenic mouse line that models BD. These ClockΔ19 mice have a genetic mutation that disrupts circadian rhythms and produces a behavioral phenotype similar to BD. We will test cognition using rodent tasks that model human cognitive functions, such as cognitive flexibility and recognition memory. The novel object recognition (NOR) task measures recognition memory by comparing time spent exploring a novel versus a familiar object. The attentional set-shifting task (AST) assesses cognitive flexibility using a digging game where the rules change continually. In the last few weeks, we have successfully optimized these cognitive tasks with wildtype mice and are currently comparing mutant mice to the wildtype mice. We predict that ClockΔ19 transgenic mice will be impaired in both tasks when compared to the wildtype mice. Our results will provide clarification for the sex-specific role of clock genes in cognition and serve to further validate the ClockΔ19 transgenic mouse model for use in translational studies of cognitive behaviors in BD.

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Aging and Neuronal Excitability in iPSC-derived Neurons

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Amyotrophic lateral sclerosis (ALS) is a complex neurodegenerative disorder that affects motor neurons in the brain and spinal cord, leading to progressive functional weakness and cell death. Although the precise cause of neuronal degeneration is still unknown, multiple cellular mechanisms have been connected to ALS, including organelle transport deficits and irregular neuronal activity. However, it remains unclear how age and genetic mutations associated with ALS, such as mutations in the C9ORF72 locus, affect these phenotypes. In this project, we used induced-pluripotent stem cells carrying the GGGGCC hexanucleotide repeat expansion (HRE) in the C9ORF72 locus (i.e., 4a cells), and its isogenic control line (i.e., 2A) in which the mutation was corrected via gene editing technology. After differentiating the stem cells to cortical neurons, we used transient transfection to track mitochondrial trafficking and neuronal activity over time through live-cell microscopy. Preliminary results suggest that mitochondrial trafficking and the rate of neuronal firing are not affected in the mutant neurons compared to controls. However, we found that three and four week old cells have increased neuronal firing compared to younger cells. It is clear that the neurons become more active as they age and mature, but the effect of the mutation on this cellular behavior is still unclear. Future work will test the rate of mitochondrial trafficking and neuronal firing in older neurons to better understand how aging compounds the negative effects of ALS-linked mutations leading to neuronal dysfunction and death.

This work was supported by RI-INBRE

Muscle Fiber Distribution and Motor Unit Contractile Properties in a Rabbit Model of Cerebral Palsy

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Cerebral palsy (CP) is a movement disorder caused by disruption of blood flow to the developing fetus. The developing motor system is affected by CP, but the links between muscle fiber types, motor unit contractile properties, and cerebral palsy are unstudied. Thus, it is unknown how prenatal hypoxic-ischemic (HI) injury disrupts the developing neuromuscular system. In the present study, we investigated the physiological differences of myofibers in neonatal rabbits modeling CP versus control rabbits. Using immunohistochemistry, we labeled the different muscle fiber types - type I (slow-oxidative), IIa (fast-oxidative), IIx (fast-glycolytic), and IIb (fast-glycolytic) - to evaluate fiber type distributions in the lateral gastrocnemius (LG). We found that the muscle fiber type composition of the LG was altered in HI kits compared to control. Electrophysiological recordings of LG motor units (MUs) were taken to investigate contractile properties, compared in sham and HI groups. Recordings of MUs from HI kits exhibited high levels of spontaneous, tonic MU activity compared to control/sham groups. Tonic MU contractions were observed to last more than 10-20 minutes. The persistent MU tonic activity exhibited could account for joint stiffness, a symptom of CP that is demonstrated in the CP rabbit model. Overall, changes to skeletal muscle contractile properties may contribute to motor dysfunction in CP.

This work was supported by RI-INBRE

Assessment of Silver Carboxylate's Ability to Decrease Bacterial Viability and Penetrate the Bacterial Outer Membrane

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As the world approaches a post-antibiotic era, alternative methods of prevention and treatment of infections are required. Current antibiotic eluting implants used in orthopedic surgeries are proving less effective due to resistance. This lab utilizes an antibiotic-independent antimicrobial coating consisting of 95% titanium-dioxide:5% polydimethylsiloxane matrix with a controlled release of silver carboxylate. This study will assess the efficacy of silver carboxylate as an antimicrobial agent as compared to nanoparticle and colloidal silver. Plates containing differing concentrations of silver carboxylate, nanoparticle silver, and colloidal silver were incubated with $1x10^6$ cells per ml of one of the two bacteria, *Staphylococcus aureus* or *Serratia marcescens*. Dose response data was generated to compare the cytotoxicity of silver carboxylate with the other conditions plated. Silver carboxylate, ranging from concentrations of 1µM to 300µM, outperformed all included nanoparticle and colloidal silver formulations as expressed in a lower optical density corresponding to less bacterial growth. The permeability of silver conditions into the bacterial outer membrane was evaluated with the 1-N-phenylnaphthylamine assay. The assay showed little difference between control and conditions meaning the treatment time should be increased to allow for penetrance. This lab's novel antimicrobial agent has the potential to establish a new standard for implants and prosthetics.

This work was supported by RI-INBRE

Developing an assay to compare translation by *Francisella tularensis* ribosomes with different bS21 homologs

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Francisella tularensis is a Gram-negative pathogenic bacterium which causes tularemia, an infectious disease that can cause serious illness and death. *F. tularensis* is considered a potential bioweapon due to its ease of aerosolization and extraordinary infectivity; we work with the live vaccine strain (LVS), which does not cause disease in humans. Notably, *F. tularensis* encodes three distinct homologs of the small ribosome protein bS21. Cells with and without bS21-2 have changes in protein abundance consistent with bS21 regulating translation. In particular, type VI secretion system proteins, which are essential for virulence, are less abundant in cells lacking bS21-2. PdpA is one such protein, and its 5´ UTR is sufficient to lead to differential translation of reporter genes in a *F. tularensis* cell-based assay. It is unknown what elements of the pdpA 5’ UTR are sufficient to confer regulation by bS21-2 nor what type of interaction there is between bS21-2 and the 5´ UTR. Our goal is to develop an in vitro method to measure the levels of translation of certain mRNAs between ribosomes containing the three homologs of bS21. The assay allows more flexibility and efficiency in testing elements of the pdpA 5´ UTR than previous methods and may help us determine if the bS21-2-mRNA interaction is direct or indirect. Our results so far confirm the assay works; we have successfully measured reporter output, which is bioluminescence. This assay will help us better understand the role of bS21 in translation and its connection to virulence of *F. tularensis*.

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Investigating the Antimicrobial Activity of Sesquiterpene Lactone Laurenobiolide

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With constantly evolving bacteria threatening the efficacy of antibiotics, the search for novel antimicrobials is imperative. Natural products historically have been used medicinally and have provided lead compounds for drug development. Laurenobiolide is a sesquiterpene lactone isolated from the North American tulip tree *Liriodendron tulipifera*. It has antimicrobial activity on methicillin-resistant *Staphylococcus aureus* (MRSA). We validated the antimicrobial activity of laurenobiolide on a methicillin-sensitive strain of *S. aureus*, found that it is active against *Francisella tularensis*, and observed no antimicrobial effects on *Escherichia coli* at the tested concentration. We isolated colonies that may be laurenobiolide-resistant *S. aureus* mutants. We also investigated the antimicrobial activity of several chemical extracts: one from *L. tulipifera*, two from *Liriodendron chinense* trees, which do not contain laurenobiolide, and an extract from a *L. tulipifera* - *L. chinense* hybrid tree which also does not contain laurenobiolide. While the extract from *L. tulipifera* showed antimicrobial activity against *S. aureus*, the other extracts did not, consistent with laurenobiolide functioning as the antimicrobial compound. Conversely, *F. tularensis* exhibited sensitivity to all extracts, suggesting the presence of one or more additional active compounds in the tree extracts. In this project, we validated that laurenobiolide shows antimicrobial activity on *S. aureus* and explored its potential on other bacterial species. The results we identified suggest the potential for a compound based on laurenobiolide to be developed as an antimicrobial.

This work was supported by RI-INBRE

Determining the genetic requirements for *Francisella tularensis* survival in freshwater

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Francisella tularensis is considered a potential bioweapon because it is the causative agent of the potentially deadly disease tularemia and has an extremely low infectious dose. In freshwater aquatic environments, *F. tularensis* can survive for long periods of time, creating a reservoir of bacteria that can potentially infect and cause disease in animals and humans. Temperatures near (but above) freezing allow for longer survival of *F. tularensis* in freshwater. Last semester, I found that the attenuated strain of *F. tularensis*, the live vaccine strain (LVS), can survive at 4°C in freshwater for 35 days. We hypothesize that there are specific gene(s) that are involved in the survival of F. tularensis LVS at 4°C in freshwater. This summer I validated that *F. tularensis* remain viable for over a month at 4°C. In the repeated viability experiment, bacteria were able to survive for 21 days longer, up to 56 days at 4°C. Given this variability, I am currently repeating this experiment for a third time. Additionally, I created a transposon-containing plasmid, pKR141, that will be used to create mutant bacteria libraries for a transposon insertion sequencing (Tn-Seq) experiment. This experiment identifies those genes specifically required for survival of *F. tularensis* LVS in freshwater. Currently, I am testing transformation efficiency, and the Tn-Seq experiment will be conducted in the fall. This experiment will allow us to determine which gene(s) are essential for *F. tularensis* LVS survival in freshwater and shed light on a poorly-understood aspect of *F. tularensis* environmental survival.

This work was supported by RI-INBRE

Synthesis of 2-Alkyl-4 Quinolones for Further Investigation into their Biological Activities

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Understanding the functionality of bacterial signaling molecules will lead to important bacterial interventions, including anti-quorum sensing mediation and increased effectiveness of antibiotics. The marine bacterium belonging to the genus *Pseudoalteromonas* utilizes the 4-quinolone molecule to perform many processes, such as iron acquisition, quorum sensing, regulation of outer membrane vesicles, and immune function. As this molecule was recently isolated from the marine bacterium, the full extent of its communicable functions as well as the mechanism of its activities is unknown. The synthesis of the 4-quinolone molecule and a library of quinolone derivatives will increase our understanding of its functions and possible manipulations. We are currently working towards synthesizing a quinolone hypothesized to contain a 14-carbon unsaturated chain. The double bond is suspected to be in conjugation with the carbonyl double bond in the quinolone ring structure. We have worked towards optimizing the reaction conditions for the cyclization reaction that produces the quinolone ring structure, using both conventional and microwave heating. Upon synthesizing these quinolones, we increase our overall understanding of bacterial communication and optimize the resulting interventions that may stem from this knowledge.

This work was supported by RI-INBRE

Peptide Synthesis to Aid in the Targeting of Quiescent *E. coli* for Prevention of Recurrent Urinary Tract Infections

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Out of the 11 million urinary tract infections (UTI) reported in the US per year, 27% of infected patients suffer a resurgence within a year, many despite successful antibiotic treatment. Up to 80% of all UTIs are caused by uropathogenic *E. coli* (UPEC) and evidence suggests that these UPEC can withstand antibiotic treatment by entering a quiescent state. Peptidoglycan (PG) stem tetra- and pentapeptides are known to bring UPEC out of their quiescent state, but the pharmacophore is still unspecified. By synthesizing derivatives of the PG stem peptide fragments and comparing the effectiveness of these slightly altered amino acids, we will be able to distinguish the pharmacophore. Synthesized peptides are produced by manual solid-phase peptide synthesis used in our lab. This research will later lead to the development of a drug able to combat UPEC from entering the quiescent state and therefore limit or cease recurring infections.

This work was supported by RI-INBRE

HPLC Purification of Synthesized Peptides for *E. Coli* Quiescent Studies

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Of the 11 million urinary tract infections (UTIs) reported each year, twenty-seven percent of the patients that were already treated with antibiotics will experience a recurrence within twelve months. The leading cause of all initial and recurring UTIs is Uropathogenic *Escherichia coli* (UPEC), which has been found to enter a quiescent state within bladder epithelial cells that allows it to survive antibiotic treatment. Additionally, it is known that differing tetra- and pentapeptide structures function as environmental cues that enable quiescent UPEC cells to regain antibiotic-sensitivity. This research will identify the peptide structures that best prevent uropathogenic bacteria from entering an antibiotic-tolerant, quiescent state, that may lead to a new treatment for recurrent UTI infections with multidrug-resistant strains. My role in this research project is to determine the best method to purify the newly synthesized peptidoglycan (PG) stem tetra- and pentapeptides using high performance liquid chromatography (HPLC) to contribute to future studies regarding the reversal of quiescent cells.

This work was supported by RI-INBRE

Organic Synthesis of Peptides to Combat Recurrent Urinary Tract Infections Caused by Quiescent Bacteria

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Urinary tract infections (UTIs) are becoming an increasingly alarming problem due to antibiotic and multidrug resistance, making it more difficult than ever to find a safe, cost-effective way of treating and preventing UTIs. The bacteria associated with over 80% of UTIs is uropathogenic *Escherichia coli* (UPEC), commonly known as *E. coli*, which plays a crucial role in pathogenesis due to its extreme diversity. Due to *E. coli*'s ability to enter a dormant, non-proliferative state, called quiescence, these bacteria can remain in the body long after antibiotic treatment has ceased, where they can begin to proliferate and reinfect the body months later, where it has been reported that 27% of women suffer from another UTI within 12 months of the initial infection.1 The bacteria's ability to enter into a quiescent state is what researchers believe is attributed to recurrent UTIs, which can potentially progress into exceedingly fatal infections, such as urosepsis. To combat this rising issue, various peptides have been synthesized through structure activity relationship studies (SARS) in hopes of synthesizing small, organic lead compounds with the ability to drive *E. coli* out of quiescence, therefore inducing proliferation and allowing for successful eradication of the bacteria from the body when treated in conjunction with antibiotics. These studies will enhance the understanding of the quiescent state in UPEC, the ability of certain peptides to reverse this quiescent state, and potentially lead to an approach for treatment of recurrent UTIs.

This work was supported by RI-INBRE

Methylation Patterns of Phase Locked Flagellar Mutants of *Salmonella enterica* and Their Impact on the Plant Immune Response

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Salmonella enterica is a common causative agent of food-related illness. Wild-type *S. enterica* serovar Typhimurium expresses flagellar filaments comprised of either FliC or FljB proteins. Phase-locked mutants express a single flagellin protein allowing these to be studied in isolation. Flagella contribute to the virulence of many bacteria and are recognized by TLR-5 and FLS2 immune receptors in animals and plants, respectively. This research aims to determine whether selective expression of one flagellin type influences the recognition of flagella by the plant's immune system. It is expected that *S. enterica* expressing FljB will colonize plants more efficiently and elicit a weaker immune response and reduced flagellin recognition by FLS2, due to increased methylation and surface hydrophobicity. High methylation levels are also expected to influence adhesion to a bacterial plant cell wall model and biofilm formation, which could also contribute successful plant colonization. Work is ongoing to develop protocols to measure callose detection, stomata closure, and reactive oxygen species production in *Arabidopsis thaliana* using flg22 (a flagellin peptide) to activate FLS2-mediated immune responses. Adherence to a cellulose-based plant cell wall model was increased for FljB-expressing bacteria, which is inconsistent with our expectation that methylation would inhibit hydrophilic interactions. Adherence to a cell wall containing cellulose and pectin is currently underway to evaluate adherence to a hydrophobic surface. Biofilm formation is enhanced in FljB-expressing *S.* Typhimurium, which correlates with enhanced methylation leading to enhanced biofilm formation. Swimming and swarming motility are unaffected by flagellin expression, which is consistent with published data.

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The Impact of Flagellin Methylation on Plant Immune Responses against Human Pathogens

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Salmonella enterica is a Gram-negative bacterium that causes gastroenteritis. Its motility is mediated by thin appendages called flagella. The flagellar filaments in *S*. Typhimurium are comprised of either FliC or FljB flagellin proteins. Flagellins in *S. enterica* are post-translationally methylated by the FliB methylase. Previously, flagellin methylation was found to increase adhesion to hydrophobic phospholipids and promote colonization of epithelial cells. When *S. enterica* enters a plant cell, its flagella interact with the flagellin-sensing 2 receptors (FLS2). If flagellin is detected, the plant's immune response is triggered. We hypothesize that FliC and FljB methylation helps *S. enterica* bypass immunogenic attacks by interfering with flagellin recognition by FLS2 receptors. For this study, we observed *Arabidopsis thaliana* immune responses in reaction to infiltration with wild-type *S. enterica* and fliB deletion mutants (these mutants have low or no flagellin methylation). Preliminary data suggests that fliB mutants of *S.* Typhimurium and *S.* Senftenberg are more motile than their respective wild-type strains. Additionally, a fliB mutant of *S*. Senftenberg exhibits increased biofilm formation compared to the wild-type, which is unexpected due to its decreased hydrophobicity. The fliB mutant of *S.* Typhimurium displays increased adherence to an artificial plant cell wall comprised of cellulose but does not adhere differently than the wild-type to a cell wall containing cellulose and pectin. This suggests that deletion of the fliB gene and subsequent reduction in surface hydrophobicity improves adherence to hydrophilic surfaces. Overall, these data suggest that deletion of fliB in S. enterica increases biofilm formation, motility, and adherence to a hydrophilic plant cell wall model.

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The Influence of the O-antigen Capsule of *Salmonella enterica* serovar Typhimurium on Flagellin Methylation and Cell-to-Cell Interactions

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Salmonella enterica can be found on a variety of produce and meat products, making it one of the most common culprits of foodborne illness in the US. The flagella that surround this bacterium enable motility and influence interactions between this bacterium and its environment. Most *S. enterica* serovars can express two types of flagellin proteins, FliC and FljB. The reasons for changes in the relative expression of FliC and FljB are poorly understood, but deletion of the yihO gene, which abrogates O-antigen capsule expression, leads to the exclusive expression of the FliC flagellin, whereas the wild-type strain preferentially expresses the FljB flagellin. Bacterial flagella are recognized by the flagellin-sensing 2 (FLS2) receptor in plant cells, which triggers an immune response against the bacterial colonizer. The objective of this research is to determine whether the loss of O-antigen capsule in a *S.* Typhimurium yihO mutant impacts flagellar methylation and FLS2 recognition in *Arabidopsis thaliana*. Mass spectrometry analysis of purified flagellin proteins revealed that flagellins in the yihO mutant have higher methylation levels than those of the wild-type strain. This suggests *S. enterica* may alter flagellin expression patterns and methylation levels to modulate overall surface properties, in this case to compensate for loss of the capsule. Deletion of the yihO gene does not affect biofilm formations but does increase swimming motility relative to the wild-type. This mutant is also less adherent to a bacterial plant cell wall model containing cellulose with or without pectin than the wild type. At this time, it is not clear whether these phenotypic changes in the yihO mutant are due to the loss of the O-antigen capsule itself, or the resulting changes in flagellin expression and methylation.

This work was supported by RI-INBRE

Optimization of an ELISA Using Dengue Protein NS1

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Non-structural protein 1 (NS1) is a protein that plays a major role in the replication of Dengue Virus, and is an important biomarker when testing a patient for its presence. But, the mechanisms that allow NS1 to be utilized as this vital biomarker are still very unclear. The overall objective of this project is to grasp a better understanding of the elusive glycoprotein NS1 and how to measure its presence in a more reliable, efficient way. Through methods that utilize NS1 antigens from serotypes 1-4 at 0.5μg/mL and antibodies DV1-DV4, and Pan serotype at 1x and 2x concentrations, optimal reactivity of the NS1 protein were determined.

Prior testing in our laboratory included running the same tests but using different incubation times and temperatures, along with using different coating, washing, and diluent buffers in order to generate the most optimal protocol when only considering these variables. Ultimately, the findings of further experimentation with an increase in antibody concentration determined that there was only a slight positive impact on the reactivity of NS1. In conclusion, this data demonstrates that antibody concentration has a negligible effect when determining optimal conditions for acquiring better reactivity when compared to other variables such as incubation conditions and buffer selection.

This work was supported by the Institute for Immunology and Informatics

TMBI-4 is a conserved transmembrane protein and potential regulator of cell death in *C. elegans*

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TMBI-4 is a conserved transmembrane protein and potential regulator of cell death in *C. elegans*
Apoptosis is a conserved mechanism of cell death essential for the development and maintenance of healthy tissues in mammals and other multicellular organisms. While the regulation of apoptosis in mammals has been historically associated with the activity of the BCL-2 family of proteins in mitochondria, a novel group of cell death regulators, the Transmembrane BAX Inhibitor-1 Motif-containing (TMBIM) protein family members have emerged. Recent findings have shown that this group of cell death regulators contribute to diverse diseases such as cancer, diabetes, and neurodegeneration. Therefore, elucidating the cellular functions of TMBIM proteins may reveal potential therapeutic targets to treat human disease. There are at least six highly conserved TMBIM proteins expressed in mammals, with homologs in yeast, insects, fish, plants, and viruses. TMBIM family members are integral membrane proteins that share sequence homology within the six hydrophobic modules of the UPF005 domain that mediates localization in organelle membranes. To determine where TMBI-4 protein is localized in C. elegans tissues, we constructed a translational reporter TMBI-4::GFP. We found that TMBI-4::GFP is expressed in several tissues including the hypodermis (epidermis), pharyngeal muscles, body wall muscles, and the intestine. To determine the subcellular localization of TMBI-4, we used fluorescent reporters that label either the Golgi or endoplasmic reticulum (ER) in the intestine. Contrary to our hypothesis, we did not observe co-localization of TMBI-4::GFP in the Golgi or ER of intestinal cells. Future studies will determine the subcellular localization of TMBI-4 in other tissues.

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