Winthrop University

Summer Undergraduate Research Experience 2024

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Exploring the Host Range of Bacteriophages

Atkinson, Alexis (2026) Wilczak, Karissa (2026) **Mentor(s): Victoria Frost**

Bacteriophages are viruses that specifically infect bacteria, often resulting in the destruction of the cell following phage replication. Practical applications of this behavior has influenced their use in the medical field, agriculture, and the food industry. Phages that are able to infect more than one host could be especially useful when targeting a range of different bacterial strains. In the summer of 2024, we explored the host-range of Winthrop's bacteriophage collection. Host range is defined as the number of hosts a phage has the capacity to infect. There were two main types of host range that we were focusing on in this project: narrow and broad. Narrow range describes when a phage can only complete its life cycle in one host. To date, this type of phage has been predominantly studied due to biased isolation techniques. Broad range is used to describe a phage that can complete its life cycle in more than one host (two or more). The majority of Winthrop's phage collection were originally isolated on bacterial host *Mycobacterium smegmatis*. In this study, *Mycobacterium aurum* and *Mycobacterium nonchromogenicum* were selected as alternative hosts. We are particularly interested in using virulent phages in host-range experiments since these phage ultimately cause cell death. Using the alternative bacterial hosts, we tested 18 of Winthrop's sequenced phages that are predicted to use the lytic cycle only. Serial dilutions of high titer phage were plated onto bacterial lawns. Then, using efficiency of plating calculations, we found that only B cluster phages were able to infect our alternative host *Mycobacterium nonchromogenicum*. In the future, we plan to investigate additional bacterial hosts, and phage from a variety of clusters, to increase our knowledge of phage that have the ability to infect multiple bacterial strains.

Effects of TiO_2 Nanoparticles on Goldfish (*Carassius Auratus*) Upper Thermotolerance

Leliana Bohanan (May 2025) **Dr. Sal Blair**

Titanium dioxide (TiO_2) nanoparticles can be found in a wide variety of products such as sunscreen, soap, and even foods such as M&Ms. Due to their widespread use, these nanoparticles often find their way to aquatic environments making them a potential threat to aquatic life, which has stimulated a surge in research efforts toward understanding the environmental implications of nanotechnology. Our initial objective was to see if TiO_2 inhibited fish's ability to cope with increasing aquatic temperatures as this could present a dual threat when considering the effects of climate change on water temperature. To perform the experiment, we subjected four groups of fish (n=8) separated into non-injected, saline-injected, polyacrylic acid capsule injected, and TiO_2 injected treatments to a critical thermal maximum (CT_{Max}) test and sampled them after loss of equilibrium (LOE). The LOE temperatures for each fish were recorded and an ANOVA test comparing each treatment group to the control group revealed that the TiO2-injected fish demonstrated a significantly reduced thermal maximum compared to controls ($p=0.0262$). Currently, we are investigating potential mechanisms through which the fish's thermotolerance was lowered, including using immunohistochemistry to find if certain heat shock proteins were under expressed in TiO_2-injected fish.

I'd like to thank the Winthrop TRIO McNair program, Winthrop Research Council, and SC INBRE branch for providing funding for this project

Illustrating Carolina Native Fish Species at Southern 8ths Farm: A Fish Guidebook

Abby Bowers (2026) **Mentor(s): Dr. Salvatore Blair**

Knowledge of native fish is vital to the conservation and success of resident populations. The Carolinas contain roughly 250 resident freshwater fish species, from the diverse Cyprinidae family to the many important game fish species sought by anglers. Southern 8ths Farm is an ecological research area on the South Carolina/North Carolina border where the piedmont and coastal plain meet. This property is dedicated to the creation of a "corridor of green" and dedicated to ecological preservation/documentation. One goal of the foundation is to catalog the rich biodiversity of the area, and our lab has been involved in surveying the fish populations of Thompson Creek, a tributary of the Great Pee Dee River. In association with Carolina Wildlands Foundation, a series of digitally rendered images were created displaying the variety of species that have been recorded in Thomson Creek, paired with photographs to create a fish field guidebook for the property. Illustrations were made in great detail, down to scale counts and fin rays, both characteristics useful in identifying individuals. Currently, 9 images have been created of native fish, and 8 more are anticipated before publication of the fish field guide using these images along with biological descriptions of each species.

Funding: Carolina Wildlands Foundation

Effects of G4-Stabilizing Ligands on RYBP mRNA and Protein Expression in Glioblastoma Cell Lines

Lannea Brown (May 2025), Melanie Williams (May 2025) **Mentor(s): Dr. Dan Stovall**

Glioblastoma multiforme (GBM) is the most common central nervous system cancer, with a median survival of less than 15 months. Gene dysregulation plays a significant role in driving GBM progression. RYBP (RING1- and YY1-binding protein) is part of the Polycomb transcription factor family and is a chromatin-modifying protein that stimulates tumor cell death in various cancer types, acting as a tumor suppressor gene. Nearly 50% of GBM tumors display decreased RYBP expression in comparison to normal brain tissue, but the mechanism of this downregulation is unknown. We hypothesized that GBM cells may decrease RYBP by aberrantly resolving G-quadruplexes (G4s), a type of secondary structure in DNA formed by guanine-rich sequences. We treated T-98 and U-87 glioblastoma cell lines with the G4-stabilizing ligands TMPYP4, PHENDC3, and pyridostatin or a vehicle control. After 48 hours, we isolated and quantified RNA using the Nanodrop 2000 and subjected samples to RT-qPCR using the Luna Universal One-Step RT-qPCR Kit to detect differences in RYBP levels. Subsequently, we examined RYBP expression at the protein level by performing protein isolation and quantification, along with SDS page and Western Blot. In both T-98 and U-87 cells, G4 stabilization with PHENDC3 and pyridostatin significantly increased RYBP mRNA levels compared to vehicle-treated control cells. However, PHENDC3 and pyridostatin surprisingly decreased RYBP protein levels compared to DMSO-treated control cells. No differences in RYBP mRNA or protein levels were observed upon treatment with TMPYP4. It is possible that G4 stabilization may increase RYBP mRNA expression, but that post-transcriptional mechanisms may compensate to suppress RYBP protein synthesis.

Optimization of Troponin C Purification

Sarah Buchanan (2027) **Mentor(s): Dr. Nicholas Grossoehme**

The detrimental effects of cadmium on the human body are well-documented; however, the biochemical mechanism behind cadmium toxicity in cardiac tissue is not well understood. It is theorized that cadmium binds to Troponin C with a higher (or equal) affinity than calcium, thus disturbing the function of the troponin complex. To investigate further, we must have a reliable method to yield and purify troponin C for experimentation. Two methods were evaluated using qualitative data from fast protein liquid chromatography (FPLC) and gel electrophoresis, and quantitative data from isothermal titration calorimetry (ITC). The first method involves cold shocking the cells with an overnight induction, which triggers the upregulation of molecular chaperone proteins which promote the protein's expression and ensure proper folding. The second method uses a leader sequence (pel B) in the plasmid, which signals the cell to secrete the protein into the periplasmic space, which should simplify the protein purification process. Results from gel electrophoresis indicate a markedly significant increase in troponin C levels using the first method and a poor yield using the second method. Troponin C purification was optimized using a combination anion exchange and gel filtration chromatography, which yielded protein estimated to be > 95% pure. Proper folding of the protein was confirmed using ITC to investigate the calcium binding affinities.

Support was provided by an SC-INBRE grant from the National Institute for General Medical Sciences (P20GM103499) and an NSF RUI Award (2203467)

What are the Preventative Measures made by Juvenile Justice Practitioners to keep Juveniles from Incarceration?

Ansley Coleman (Dec 2024) **Mentor: Dr. Bradley Tripp**

This research seeks to examine how Juvenile Justice Officers view their jobs and the young people they process. This research specifically seeks to better understand how and if American Juvenile Justice Officers approach their work with orientations towards preventative rather than punitive processes. Existing research asserts that a preventative orientation is underutilized. Our research will find if this is true in the everyday efforts of the officers interviewed. We conducted online interviews with eight officers from different departments in the state of South Carolina. We plan to apply criminological theory to the way officers conduct their procedures and treat children.

Explorations of Machine Learning Methodologies to Enhance the Design of RNA-based Dopamine Biosensors

Students: James Craven (2025)
Matthew Lindsey (2025)

Mentors: Dr. Kristen Abernathy, Dr. Zach Abernathy, Dr. Timea Fernandez

In this project, we seek to understand how we can utilize machine learning (ML) to inform the design of biosensors to emit a strong fluorescence in the presence of dopamine. Dopamine is a neurotransmitter and plays a role in a variety of functions such as memory, learning, and reward systems. Detecting dopamine levels could help with diagnosing addiction, mental illness, and neurodegenerative disorders. We develop a framework for one-hot encoding nucleotide sequences for training ML models, create a data preprocessing pipeline to pad sequences and normalize output values, and construct neural net model architecture for training on both sequence and numerical data. Using a published toehold switch dataset along with a ribosensor dataset provided by Dr. Timea Fernandez's lab, we explore the accuracy of several different regression models in predicting biosensor effectiveness by training on both nucleotide sequence data and calculated thermodynamic parameter data. We find that a neural network model, specifically a multilayer perceptron, typically outperforms other regression models such as linear regression, random forests, and support vector machines in both datasets, and that training on sequence data appears to be more predictive than training on thermodynamic parameter data. We also suggest potential directions to pursue transfer learning between the two datasets.

Support for this research was provided by ADAPT in SC, an EPSCoR program supported by NSF Award #OIA-2242812.

Exploring the Impact of Cd^{2+} on the Interaction Between Troponin-C and Troponin-I

Morgan Dukes (2026) **Mentor(s): Dr. Nicholas Grossoehme**
Cierra Ari Randolph (2026)

Cd^{2+} is a toxic heavy metal known to negatively impact cardiovascular muscle function via a complicated mechanism that involves several proteins working synergistically to carry out and regulate the process. The troponin complex, composed of troponin-C, troponin-I, and troponin-T, serves as the link between brain signaling and muscle function. Upon nerve impulse, Ca2+ binds to troponin and signals muscle contraction. Troponin-C is the calcium binding site of the complex and interacts with troponin-I when Ca^{2+} binds, specifically troponin-I's switch peptide and 1-73 region, which directly interact with troponin-C upon Ca^{2+} binding. Recent evidence shows Cd^{2+} can bind to troponin in place of Ca^{2+}. Our hypothesis is that the mechanism of Cd^{2+} toxicity is related to the interaction between troponin and metal. The aim of our research is to understand how Cd^{2+} impacts the interaction between troponin-C and the troponin-I switch and 1-73 peptides. To date, our research has established reliable purification strategies for the maltose binding protein (MBP)-tagged troponin-I constructs and confirmed a low affinity calcium-dependent interaction between troponin-C and troponin-I. Current efforts are focused on optimizing quantitative strategies to investigate this interaction.

Support was provided by an SC-INBRE grant from the National Institute for General Medical Sciences (P20GM103499), the U.S. Department of Education McNair Grant (P217A180094), and an NSF RUI Award (2203467).

RitR is a Redox-Sensitive Iron Sensor in Streptococcus pneumoniae

Nick Fatigante (2027) **Mentor(s): Dr. Nicholas Grossoehme**

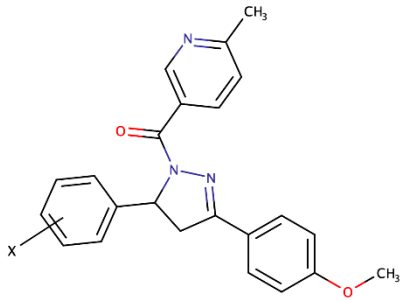
Streptococcus pneumoniae is a gram-positive bacterium that contributes to diseases like meningitis and pneumonia. These diseases contribute to over 1 million deaths each year. *S. pneumoniae* contains a novel iron uptake regulation system with no known intracellular iron detection mechanisms. RitR (Repressor of Iron Transport) is a protein known to repress expression of the *piu* operon, a gene cluster responsible for creation of iron uptake machinery. This research seeks to investigate the relationship between oxidation and DNA-binding potential of the regulatory protein, RitR. Further, this project explores the potential of RitR to serve as an intracellular iron detector within *S. pneumonaie's* iron uptake regulation system. RitR was grown in *Escherichia coli* and purified through cation exchange chromatography. RitR dimer was theorized to be formed under oxidative stress caused by conditions of excess iron. Formation of the RitR dimer was performed by adding hydrogen peroxide through overnight dialysis in a buffer that contains high 100 mM malate. Dimer formation was confirmed through gel electrophoresis. A segment of the Piu operon was marked with a Fluorescein tag and prepared to form double stranded DNA. Formation of double-stranded DNA was confirmed through fluorescence anisotropy with a serial dilution with varying DNA concentrations. Surprisingly, our results establish that 100 mM malate is able to stabilize iron (III) at concentrations of at least 10 mM. This malate-stabilized iron (III) was added to a solution containing the labelled DNA and analyzed with fluorescence anisotropy in multiple experiments that varied concentrations of Dimer-DNA complex or iron (III). Our results indicate that the RitR-dimer forms a tight complex with the piu DNA above a threshold concentration of 1 mM iron (III) stabilized in 100 mM malate. Using the known equilibrium constant for iron (III) binding to malate, we can approximate that RitR-DNA complex formation is driven by sub-picomolar concentrations of free iron (III). Further, these results suggest that RitR is highly tuned to repressing iron uptake under oxidative conditions (both dimerization and iron (III) formation are favored under such conditions), and that iron (III) directly contributes to binding of RitR dimer to the piu operon, suppressing its expression and potentially acting as an iron detection mechanism in *S. pneumoniae.*

Support was provided by an SC-INBRE grant from the National Institute for General Medical Sciences (P20GM103499)

Synthesis of BRAF Inhibitors—Derivatives of 4,5-dihydropyrazoles

Jessica Gengler (2024) **Mentor: T. Christian Grattan**

This research is dedicated to developing inhibitors of the BRAF cancer protein to help shut down its expression as a way of treating cancer. Cancer develops from the abnormal growth of cells within the cell division process. Abnormal cells, instead of healthy cells, go through cell division to grow and multiply. One cell signal mutation that is prevalent in cancer cells is in the BRAF protein. This protein is responsible for functions such as cell growth, differentiation, and proliferation. With the substitution of a glutamic acid to a valine at the 600th position, the protein becomes about 500 times more active than the non-mutated BRAF protein. This mutation (BRAFV600E) is found in several types of cancers, most frequently observed in 50-70% of melanoma tumors, and found in papillary thyroid cancer, colorectal cancer, and nonsmall cell lung cancer. The mutation of the position 600 glutamic acid to valine in BRAF allows for the cancer cell's survival by continuing ERK activity, driving proliferation and survival, and contributing to neoangiogenesis to provide necessary tumor growth and maintenance functions. Inhibitors designed to stop the expression of BRAFV600E are currently used as anti-cancer drugs, specifically niacinamide and 4,5-dihydropyrazole derivatives. With this information, this research aims to synthesize similar 4,5-dihydropyrazole derivatives incorporating substitution geometry and different halogens (bromine, chlorine, and fluorine), to have the most compatibility with the active site of BRAFV600E. The success of these inhibitors will be determined using bioassays.



*X-Br, Cl

Support was provided by an SC-INBRE grant from the National Institute for General Medical Sciences (P20GM103499)

Molecular marker analysis of Helianthus schweinitzii Torr. & A. Gray

Zosia Gordon (2025), Gwendolyn Tomlin (2027) **Mentor: Kunsiri Grubbs**

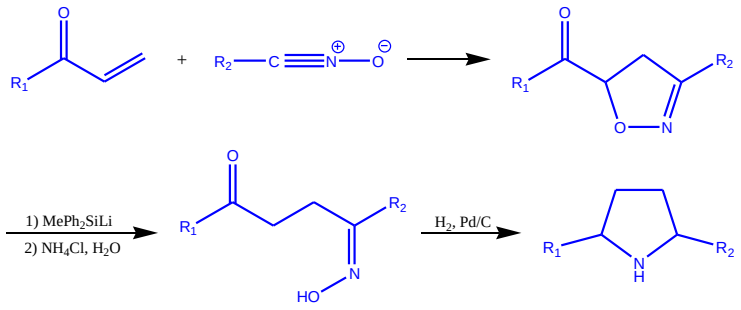
Helianthus schweinitzii is an endangered sunflower endemic to the Carolina Piedmont, and is a tetraploid hybrid of H. microcephalus and H. giganteus. One unique feature of this species is its ability to asexually reproduce through cloning. Though this feature has aided in the survival of many populations after drastic reductions in size from urbanization, clonality may lead to a lack of genetic diversity. Therefore, it is important for the conservation of the species to understand the genetic makeup of each individual population. This research determines the genetic diversity both within and among populations of H. schweinitzii. We used samples of H. schweinitzii from the Gold Hill (South Carolina) population as a model for determining genetic diversity within a population, and samples from various locations in North Carolina as a model for determining genetic diversity among populations. We performed DNA extractions, did PCR testing of potential 19 molecular markers, and used gel electrophoresis to ensure viability of the DNA and PCR products. We found appropriate methods extracting and purifying DNA samples. Additionally, we tested and found optimal conditions for the PCR protocol that we plan to use for our future research. Our next step is to investigate the genetic analysis by including the use of chosen molecular markers attached with M13 which will bind with the fluorophores. This fragment length analysis of these molecular markers will provide essential data for determining imperative population information, such as clonality, genetic diversity, population structures, etc. among the populations of H. schweinitzii that can aid in the conservation of this endangered species

Synthesis of 2,5-Dialkylpyrrolidines via Reduction of γ-Ketooximes

Kayla Hall (2026) **Mentor: Aaron M. Hartel**

γ-Ketooximes are versatile synthetic intermediates that serve as direct precursors to several important compound classes. We are interested in transforming these valuable intermediates into 2,5-dialkylpyrrolidines, some of which occur naturally in the venoms of various species of *Solenopsis* ants. Related piperidine alkaloids from other *Solenopsis* species have shown significant antiangiogenesis activity and have been investigated as a potential treatment for cancer. The synthetic strategy involves the 1,3-dipolar cycloaddition of a nitrile oxide with an α,β-unsaturated ketone to give an acylisoxazoline, followed by a ring-opening Brook rearrangement. Selective reduction of the resulting oxime would then initiate an intramolecular reductive amination to give the target pyrrolidine.

The project's current focus is determining conditions appropriate for the final reductive cyclization. Because the γ-ketooximes required for the proposed syntheses can only be made in small quantities, the reductive cyclization was modeled using the inexpensive, commercially available reagents cyclohexanone and cyclohexanone oxime.



This project was funded by the Winthrop Research Council (SC23004 & SC23007)

Exploring Potential Relationships Between Caffeine Intake, Mental Wellbeing, and Gastrointestinal Symptoms in College Students

Kelsie Johnston (Spring 2025) **Mentor(s): Jessie Hoffman, PhD, RD**

Caffeine consumption is common among college students, with research suggesting the high prevalence of consumption to manage day-to-day life and fatigue, may also contribute to gastrointestinal symptoms. Two factors that can influence GI symptoms are diet and stress, both of which can be affected by caffeine intake. Because college represents a time of dietary, lifestyle, and stress changes, the objective of this study was to assess caffeine consumption and beliefs about caffeine in college students and the potential impact these may have on digestive health.

Methods:

This cross-sectional survey was conducted at Winthrop University in Spring 2024. 103 students completed a survey assessing caffeine intake, stress levels, sleep habits, and gastrointestinal health. Data was collected using Qualtrics and analyzed using SPSS. Statistical significance was set at $p<0.05$.

Results:

The median age of participants was 21 years old, 76% of participants were female, and 52% lived on campus. 44% of participants reported being more likely to use caffeine to avoid eating more than they should, and 39% reported using caffeine to skip meals entirely. 58% of participants reported feeling jittery after caffeine consumption and 47% reported an irregular heartbeat and upset stomach after caffeine consumption. Interestingly, we observed a significant positive correlation between reports of feeling stressed after caffeine and being bothered by loose stools in the past week ($p=0.003$).

Conclusions:

This study provides insights into how caffeine contributes to gastrointestinal and stress-related symptoms within this population. The findings suggest that while college students feel that caffeine may enhance their alertness and motivation, it may also lead to disordered eating patterns and physical discomfort. Future research should examine the relationship between caffeine intake and disordered eating habits in college students.

Topping time: vertebrate herbivory on shrubby oaks

Sabrina Rocha (Fall 2024) **Mentor: Dr. Jennifer Schafer**

Amber Mercer (Spring 2027)

Vertebrate herbivores can significantly impact plant populations and primary productivity due to their high metabolic rates and broad dietary preferences. To defend themselves against herbivores, plants contain a variety of chemical and physical defenses. Our research investigated factors influencing vertebrate herbivory on multi-shoot resprouting shrubs in recently burned Florida scrubby flatwoods. Specifically, we studied three shrub species - *Quercus inopina*, *Quercus chapmanii*, and *Quercus geminata* - that dominate scrubby flatwoods and exhibit different levels of physical and chemical defenses. We compared the number of individuals with topped (herbivorized) shoots and the percent of shoots topped among species and betweeninterior and edge habitats. We assessed a total of 435 shoots from 90 individuals (30 of each species). Our findings showed that *Quercus geminata* had the highest number of individuals with topped shoots and the highest percent of shoots topped. No difference was observed in the extent of shoot damage between interior and edge locations. Our results indicate that vertebrate herbivory varies among shrub species but not between edge and interior areas.

Research reported in this publication was supported by the National Institute of General Medical Sciences of the National Institutes of Health under Award Number P20GM103499. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

Understanding the Importance of Phage Gene Roles, and the Significance of Their Loss

Jessica E. Morgan (2025) **Mentor(s): Dr. Victoria J. Frost**

Winthrop University is home to two populations of bacteriophage ExplosioNervosa; a mutant and wild-type. Sequencing and annotation of the wild-type genome revealed 96 putative genes. The mutant population contains a ~4000bp (11 genes) deletion that the wild-type retains. Thus far, both the mutant and the wild-type population are still capable of infecting their original isolation host, *Mycobacterium smegmatis*. At the genetic level, to understand whether loss of genes in this region is significant, it is important to elucidate their potential role first. This was done by amplifying and cloning each individual phage gene present in the wild-type phage, over-expressing them in their host, and observing the phenotypic effects on host growth. Two out of the eleven deleted genes have been shown to interact with the host in this way; gp80 and gp81. These genes were chosen to be further investigated using a Bacterial 2 Hybrid assay. A B2H assay employs the use of two additional cloning plasmids; one containing random fragments of the host proteome (pCI), the other (p2Hα) contains the phage gene insert. We can induce the expression of these plasmids, and if they interact, they will form a connection that allows for the expression of reporter genes. Both gp80 and gp81 have successfully been cloned into p2Hα plasmids. So far, 30 interactions have been identified for gp81, and once they are confirmed to be true "hits", they will be sequenced to characterize the identity of the host fragment. This information provides clues as to possible phage gene function, and could suggest consequences of gene loss.

Support was provided by an SC-INBRE grant from the National Institute for General Medical Sciences (P20GM103499), Winthrop Biology Department, and HHMI.

Machine learning enhanced design of RNA-based fluorescent biosensors for the detection of the neurotransmitter dopamine

Student(s): Abbie Nation (2026), Emma Westmoreland (2025) **Mentor(s): Dr. Timea G. Fernandez**

Dopamine (DA) is a neurotransmitter that plays a role in the regulation of physical and emotional well-being. Irregularities in DA production have been linked to several addictive behaviors such as smoking, alcoholism, and obesity, as well as neurodegenerative disorders like Parkinson's disease. Early detection of DA abnormalities is paramount for effective diagnosis and treatment, while real-time imaging of DA could assist in the comprehension of their underlying mechanisms. As such, our project aims to design a DA-sensing RNA-based fluorescent (RBF) biosensor for initial in vitro experimentation and characterization. Using existing platforms, we can fabricate RBF biosensors that combine a ligand-sensing RNA aptamer with a fluorescent RNA aptamer to indicate the presence of biologically relevant molecules. Previous studies have used electrochemical and protein-based biosensors in the detection of neurotransmitters; yet, to our knowledge, no studies have developed a viable RBF biosensor for the detection of DA in vitro or in vivo.

Previously, our collaborators in the Abernathy group developed an A.I. algorithm to predict which sensor is expected to be effective at detecting its ligand. The algorithm was based on analyzing the thermodynamic parameters of ~100 existing sensors coupled with their ability in detecting their ligand. Thermodynamic parameters included: entropy, ΔG, number of hydrogen bonds, length, melting temperature of the transducer, as well as the melting temperature of the sensor.

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A New Generalization of the continuous Bernoulli Distribution

Garrett Nix (Spring 2027),
Lucas Robenolt (Spring 2025)

Mentor(s):
Dr. Gihanee Senadheera,
Dr. Duha Hamed

The aim of this research was to create a new generalization of a recently introduced distribution known as the continuous Bernoulli (CB) distribution. Many known distributions are useful in modeling data but often have limited shapes, such as being right skewed, left skewed, or symmetric. These limitations make it difficult to use these distributions in broader applications, where data may not follow the shape of these models. By generalizing distributions, adding more parameters increases the flexibility of these distributions in most cases. This allows the new generalizations to have a wider range of applications than the original distribution. With this research, we propose a new generalization of the continuous Bernoulli distribution using the T-R{Y} framework, where we define random variables T, R, and Y that follow specified distributions. With this framework we introduce the T-CB{Cauchy} as well as the T-CB{logistic} families of distributions and investigate the properties of these families. We also investigate the properties of members within the families of these distributions, introducing the normal-CB{Cauchy} and the normal-CB{logistic} distributions.

“We First Eat with our Eyes.” Why not Eat *for* your Eyes?: A collection of Evidence-Based Recipes that Promote Eye Health and Function

Jessa Ordile (Dec 2024) **Mentor(s): Hope Lima**

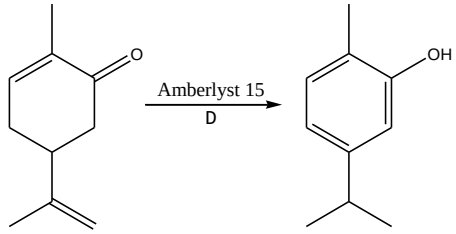
This project is a culmination of ocular and nutrient-related research reports paired with an evidenced-based recipe book targeted toward eye health and function. The eyes are complicated organs that serve as one of our most fundamental functions: sight. “See Food” is a collection of recipes that focus on not just flavor and presentation but primarily on the nutritional contents in the food that benefit the eyes. There are several nutrients addressed in this report and in the recipes that have a role in eye maintenance and function. The nutrient analysis consists of vitamins C, D, and E, zinc, omega-3 fatty acids, and carotenoids lutein and zeaxanthin. The report explains the role that these nutrients have in preventing macular degeneration, dry eye syndrome, and cataracts. The recipes themselves were collected based on a variety of cultures, flavors, and pairings. Each recipe has been physically created and tested to ensure a delicious and accurate recreation for those reading the recipes.

Acid-catalyzed Rearrangements of Terpenes: Formation of Carvacrol from Carvone and Rearrangement of Limonene

Leonor Paisana (2025) **Mentor: Aaron M. Hartel**

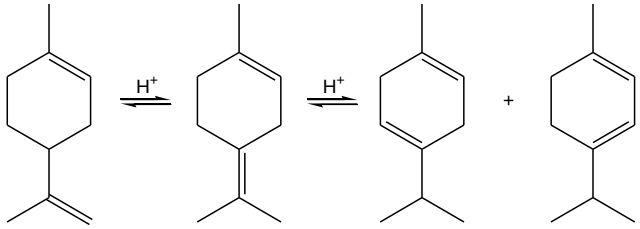
Terpenes and terpenoids are important classes of naturally occurring compounds consisting of repeating "isoprene" units. We have developed two experiments for an organic chemistry lab course in which a familiar-scented terpene/terpenoid undergoes acid-catalyzed rearrangement. In the first experiment, carvone (spearmint oil) is rearranged to form the terpenoid carvacrol (oregano). In the second experiment, limonene (lemon oil) is rearranged into several isomers and product ratios are compared to those predicted by molecular modeling.

Experiment 1:



carvone (spearmint) carvacrol (oregano)

Experiment 2:



A limonene B C D

Aspects of Bootstrap Percolation on the Random Geometric Graph

Isaac Pelletier (2024), Ethel Sakyi (2026), Gabriel Tristano (2027) **Mentors: Arran Hamm and Jessie Hamm**

Bootstrap percolation (BP) is a process on a graph which can be used to model the spread of an infection throughout a graph. First, an initially active set of vertices is given. Then, loosely, vertices are activated if they're in contact with enough active vertices. More precisely, suppose G is a graph, k is the bootstrap parameter, and a set of active vertices at time 0 is given. In the next time step, any inactive vertex with at least k edges to active vertices becomes active; the process continues until no new active vertices are created. To see how this can be used to model the spread of disease, take people as vertices, interactions between people as edges, and make "active" mean "infected".

The random geometric graph (RGG) is formed choosing points from the unit square uniformly at random and joining points that are *close enough* with an edge. We note that the random graph tends to be particularly relevant when modeling a community practicing social distancing.

Bootstrap percolation on RGGs has been studied and largely resolved for fixed bootstrap parameters. Our research focused on a handful of variations to the standard BP on the RGG problem. First, we incorporated a classical SIR-type model. The SIR model is used when modeling the spread of disease with differential equations; the S, I , and R stand for susceptible population, infected population, and recovered population, respectively. We then incorporated the idea of vaccinations into the BP on the RGG problem which ends up resembling a variation on the classical firefighter problem on graphs. We next considered allowing the bootstrap parameter to be time-dependent as was done in the recent article titled "Minimal percolating sets for mutating infectious diseases"; doing so models the idea that a disease may be more or less infectious over time. The last stand-alone variation we considered was to make the disease spreading process randomized which is in direct contrast with standard bootstrap percolation. Finally, we considered various pairings of these variations. In each case, partial results were obtained.

This project was supported by an Institutional Development Award from the National Institute of General Medical Sciences (2 P20 GM103499 20) of the National Institutes of Health.

Phenotypic Analysis of Phage-Host Interactions

Lidia Peralta (2026) **Mentor(s): Dr. Victoria J. Frost**

Bacteriophages compete among themselves to infect and gain control of their bacterial host. Once inside the cell it is evolutionarily advantageous for an established phage to protect the host for its own use. In our lab, we have been attempting to reveal which phage genes may play a role in this defensive process by using a phenotypic defense assay. Individual genes from mycobacteriophage ExplosioNervosa are transformed into the bacterial host *Mycobacterium smegmatis*. Lawns of the transformed host are created, ExplosioNervosa's gene of interest is induced, and dilutions of external phage lysates are spotted onto the lawn. Several phage lysates from both the same, and different, subclusters were applied in this way. ExplosioNervosa itself (wild-type and mutant variants) was investigated for its ability to infect the host as a specific ExplosioNervosa gene was expressed by the cell (homotypic defense). Other phage lysates tested include Ashballer (A1), Bombitas (J), and Jinnie (unclustered). We have studied nine of ExplosioNervosa's genes so far, including gp1, 19, 23, 32, 41, 55, 57, 73, and 75. Of this group, gp75 has shown potential defense of the host against external phage infection, which is particularly interesting as this gene product is predicted to be an immunity repressor. Immunity repressors are known to maintain lysogeny, as well as play a role in host defense. Understanding these phage-phage interactions can add to our knowledge of phage biology and behavior.

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Nucleic Acid Aptamer Au Nanoparticle Conjugates as Trojan-Horse drug Delivery Vehicles in the Fight Against Bacterial Infections

Jadyn Willams (December 2024) **Mentor(s): Dr. Timea Fernandez**
Julianne Phu (May 2026)

Illnesses caused by bacteria are a major public health concern since microorganisms have become increasingly resistant to available antibiotics. At the same time, big pharma has gradually shifted its focus from developing drugs that cure diseases to those that treat chronic conditions. Thus, rediscovering old drugs and using them for new purposes is becoming more important. The long-term goal of this project is to use nucleic acid aptamer-nanoparticle conjugates as vehicles for targeted delivery of antibiotics to bacteria that are resistant to them.

Currently, we are investigating the therapeutic potency of nucleic acid-gold nanoparticle conjugates as carriers of tetracycline and ampicillin to treat infections caused by Gram-Negative model organisms. We hypothesize that by attaching nucleic acids that binds to these antibiotics, to gold nanoparticles, the resulting conjugates will work as a "Trojan-horse" antibiotic-delivery vehicle that smuggles the antibiotic into the cell without being detected by cellular defense systems.

To test the viability of this idea we used tetracycline and ampicillin binding DNA aptamers that were developed for detection of these antibiotics. We optimized conditions to attach these DNA aptamers to gold nanoparticles. We are currently testing the antimicrobial effect of these aptamer nanoparticle conjugates using the Gram-negative model organism *E. coli* (ATTC strain 29522). MTS assays were used to verify that the aptamer-nanoparticle conjugates do not harm mammalian cells. Recently we began using DNA aptamers that were developed for pathogen detection to target these antibiotic-nanoparticle conjugates to specific pathogens.

Our studies indicated that Au nanoparticle antibiotic conjugates are more effective at reducing the growth of Gram-negative model organisms than the respective antibiotics alone without harming mammalian cells.

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Deference to Prestige and Climate Policy

Ty'teona Shannon (December 2024) **Mentor(s): Dr. Jeffrey Sinn**

Values and ideologies can shape attitudes toward effective climate change policies. For instance, I support significantly expanding infrastructure for public transportation. The ideology of Social Dominance Orientation (SDO) is associated with both economic conservatism and environmentalism. This study will investigate an additional orientation: Deference to Prestige (DTP) and its potential impact on climate-related attitudes. DTP argues individuals tend to defer to the wealthy or those in high-status positions. We hypothesize that DTP may serve as an indicator of resistance to climate policies, as individuals may fear losing access to resources if restrictions are placed on the wealthy. Hierarchical regression was used to test predictive power. We used SDO and exploratory measures of DTP to predict support for climate change policies. We confirmed SDO to be a power and the strongest predictor. As SDO increased, support for climate change policies decreased. Our additional findings were unexpected. Both Social Conservatism and a DTP measure (Heroic Economic Individualism) were positive predictors of support for climate change policies. We are of the opinion that social conservatism encourages conformity to support the proposed climate action and that Heroic Individualism fosters a readiness to support bold policy actions.

Investigating the effects of Semaphorin 3A, an axonal guidance molecule, on chick RGC and DRG neurons

Mallika Kaya Singh (graduating May 2026) **Mentor: Dr. Eric Birgbauer**

During the development of the visual system, axons from the retinal ganglion cells (RGCs) in the eye must form connections with their targets in the brain in order to relay information. Molecules that act as axonal guidance cues aid in this development. The axon has motile, finger-like projections at its end known as the "growth cone". The growth cone is responsible for the exploration of the axon's environment. The growth cone induces a response to specific guidance molecules in its path. This response can be attractive or repulsive. The repulsion of the growth cone can be seen in the retraction of the projections which serves as the basis for the growth cone collapse assay. A guidance cue previously documented is Semaphorin 3A (Sema 3A), which has been shown to have induced *in vitro* collapse in the growth cones of dorsal root ganglion (DRG) cells in chicken. Previous literature found that while DRGs respond to Sema 3A, RGCs of the central nervous system did not. However, students in the Birgbauer Lab found that chicken RGCs did respond to Sema 3A and growth cones showed collapse. To study this discrepancy, we set up a series of blind treatments using varying doses of Sema-3A for retinal explants taken from chick embryos at E6. As a parallel control experiment, we also set up Sema 3A blind treatments for DRG explants from E7 chick embryos. We found that while DRGs were confirmed to have a dramatic response to Sema-3A, axons from RGCs did not have a dose-dependent response to the molecule. This would indicate that Sema 3A may not play a role in the development of the visual system in chicken. To confirm this hypothesis, future experiments can use time-lapse microscopy to record the responses of growth cones to Sema 3A treatment.

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Embryonic Chicken Eye Explants Provide a Novel Model for Studying Corneal Wound Healing

Timothy Swartz (Spring 2025) **Mentor: Jena Chojnowski, Ph.D.**

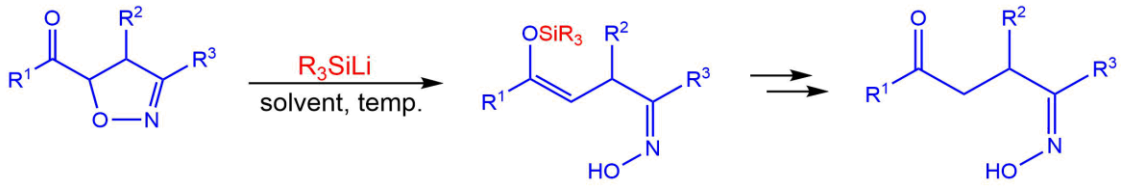
Aniridia is a congenital disease that presents as a lack of an iris in a patient's eye. This is condition is caused by a mutation in the PAX6 gene, a highly conserved transcription factor that is essential for development and regulating cellular repair in the eye. The objective of this research was to develop techniques that could be used to study the relationship between PAX6 and cellular repair. The cornea was used as the model tissue as PAX6 expression has been shown to influence cellular repair in the cornea, but a clear mechanism is unknown. PAX6 is well conserved across the animal kingdom, and chicken eyes are commonly used as analogs for human eyes. The objective of this research was to develop techniques that could be used to study the relationship between PAX6 and cellular repair. Embryonic chicken corneal explants were used as a model system. Whole eye explants were dissected from E-10 chicken embryos and cultured for 24 and 48 hours in different mediums to test the efficacy of culturing methods for use in further experiments. The corneas were dissected from the cultured eyes for histological analysis. The results showed that the culture medium used was effective in maintaining cellular viability.

Support was provided by an SC-INBRE grant.

Utilizing the Brook Rearrangement to Form γ-Ketooximes and Their Silyl Enol Ethers from Acylisoxazolines

Jaylin Sypolt (2025) **Mentor: Aaron M. Hartel**

γ-Ketooximes are versatile synthetic intermediates. Additionally, the corresponding silyl enol ethers of these useful structures have potential for the preparation of more highly-substituted variants. We have developed a method for the preparation of γ-ketooximes and their silyl enol ethers from acylisoxazolines using silyllithium reagents.



The reaction utilizes a Brook rearrangement: the migration of a silyl group from a carbon to an oxygen. By adjusting reaction parameters such as solvent, silyllithium reagent, and temperature, the reaction can be tuned to favor either the γ-ketooxime or the silyl enol ether product. Optimization experiments have shown the solvent used and the amount of silyllithium added to be the most impactful variables. Upon completion of the reaction, the γ-ketooxime or silyl enol ether can be isolated using column chromatography.

This project was funded by the Winthrop Research Council (SC23004 & SC23007)

Modifications of Sphingosine-Kinase Inhibitors to Improve Bioavailability

Ravyn Torres (2024) **Mentor: Dr. Grattan**

Sphingosine Kinase 1 (SPK-1) is a naturally occurring enzyme in the body derived from ceramides. It aids in forming sphingosine-1-phosphate (S1P) which can lead to the proliferation and survival of cells. In a cancerous system, S1P is overexpressed. This leads to the mass proliferation of cancerous cells causing tumors. To prevent this, an inhibitor needs to be developed to prevent cancer from being able to express S1P. Using a known template molecule of a sphingosine kinase inhibitor (SKI), eight inhibitors were synthesized in hopes of improving the oral bioavailability of the drug. Modifications were made on the first naphthaldehyde ring of the template molecule using various ring structures. The inhibitors made from these modifications will be tested to determine their efficiency at inhibiting SPK-1 against the template inhibitor.

This research was supported by the National Institute of General Medical Sciences of the National Institutes of Health under Award Number P20GM103499 (SC-INBRE).

Physiological Effects of Dual Environmental Stressors on Redear Sunfish

Sam Williamson (Fall 2025) **Mentor: Dr. Salvatore Blair**

Andrea Vega (Spring 2027)

Due to changing environmental conditions resulting from anthropogenic activities, it is necessary to understand how fish respond to changes in their environment. Freshwater fish can reversibly remodel their gill tissue in response to external stimuli. A compromise exists between the uptake of O2 and maintenance of osmotic balance, whereby the surface area of the gill tissue epithelium can be altered. Prior research has shown that the interlamellar cell mass (ILCM) grows or is shed relative to environmental conditions. However, prior research does not make clear how gills respond to simultaneous competing stressors such as temperature and salinity. Therefore, the focus of this study was to use redear sunfish (*Lepomis microlophus)* as a model organism to compare ILCM levels between fresh and elevated salinity water (17 ppt) and hot (26°C) and cold water (10°C), as well as to determine a potential cellular and molecular mechanism responsible for the observed gill remodeling. To test this, we created a matrix of varying combinations of the temperature and salinity conditions as different experimental 48-hour exposure treatments. The results thus far indicate that there is a significant difference in the lamellae ratio from the cold temperature to 26°C, but not from fresh to elevated saline, and the salinity treatment showed heightened plasma osmolality compared to freshwater in the 20°C condition. These preliminary results indicate that temperature is likely a stronger environmental stressor on the redear sunfish capable of stimulating changes at the gill.

Support was provided by an SC-INBRE grant

RYBP Does Not Sensitize GBM Cells to G4 Ligands' Cytotoxic Effects

Clara Whitehead (May 2025) **Mentor: Daniel B. Stovall, Ph.D.**

RYBP (Ring1 and YY1 Binding Protein) is a Polycomb group (PcG) protein known for its tumor suppressive functions, such as promoting cell death, inhibiting cell proliferation, and increasing chemosensitivity. In glioblastoma (GBM), an aggressive cancer of the central nervous system, PcG proteins, including RYBP, exhibit atypical expression patterns, often with downregulation in tumor cells. RYBP's role as a tumor suppressor suggests that its reactivation might sensitize GBM cells to cytotoxic therapies such as G-quadruplex (G4)-stabilizing ligands have anti-tumor effects by inducing DNA damage and cell cycle arrest. To test whether RYBP expression enhances the sensitivity of GBM cells to G4 ligands' cytotoxic effects, T98G GBM cells were transduced with lentivirus to ectopically express RYBP. Cell viability was assessed using a WST-1 assay after treatment with vehicle (DMSO), 10 μM PHENDC3, or 10 μM Pyridostatin for 72 hours. A two-way ANOVA revealed that RYBP expression did not significantly influence the cytotoxic effects of G4 ligands. While G4 ligands reduced cell viability, there was no significant interaction between RYBP expression and G4 ligand-induced cytotoxicity. Surprisingly, RYBP-expressing cells had higher viability at time 0 than their control counterparts, but this is likely due to potential plating inconsistencies, that may have confounded results. Consequently, RYBP does not appear to sensitize GBM cells to the cytotoxic effects of G4 ligands in this model. Future experiments will involve silencing RYBP expression using RNA interference to investigate whether the loss of RYBP impairs the cytotoxic effects of G4 ligands, providing further insights into RYBP's potential role in modulating G4 ligand sensitivity in GBM.

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