2016

Women in Science 2016

Clark Science Center's Summer Research Fellows Program

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2016 Women in Science

Clark Science Center’s Summer Research Fellows Program
INTRODUCTION

“The sciences at Smith will provide transformative opportunities for all students to engage with real problems while empowering them to generate innovative solutions that benefit our world.”

Our vision for the future is grounded in a shared aim to “cultivate the scientist in the next generation of women leaders.” Our strategic plan for the sciences (Vision for the Future, 2015) builds on Smith College’s strength as a national leader in science research and education among liberal arts colleges. An increasing number of Smith students study sciences. In 2016, forty percent of our current students declared a science major, a rate at least double the national average for women. In disciplines in which women are most under-represented (e.g., computer science), our students major at rates up to three times the national average. At Smith, our approach to education in the sciences also represents a response to societal matters – lower female participation in STEM higher education and later in work, as well as academic, economic, and political leaders’ advocacy of full representation of women in all STEM fields as a matter of equity and good policy based on the benefits that flow from diversity.

Ensuring access for all. As its first principle of excellence, the Association of American Colleges and Universities’ Liberal Education and America’s Promise (LEAP; 2011) initiative tells us to “aim high—and make excellence inclusive” (p6). For the sciences at Smith, we are guided by understanding that persistence and the best scientific thinking emerge from healthy climates that promote and value a diversity of perspectives. Smith’s strategic direction for access is to address disparities in gender, racial, and socioeconomic representation in the sciences by pairing rigorous learning expectations with robust support and community-building for our students.

Engaging with the world. Another essential principle of excellence in undergraduate education is to provide opportunities for students to engage with big questions and tackle real-world problems that connect their knowledge to solutions and action (AAC&U, 2011). At Smith, we are guided by the belief that interactions with bona fide scientific problems connecting our students to the larger world facilitate the best learning. Smith’s strategic direction for engaging the world is to connect our students with complex, real-world problems, ranging from local to global, that are often best understood through the multiple disciplinary lenses of the liberal arts.

Developing knowledge and skills. Research is a core practice of scientific education at Smith College. We are guided by a shared understanding that best-practices pedagogies and faculty-student research collaborations will result in optimal learning and future success for our students. As we move forward, we build on evidence that through rigorous coursework and undergraduate research opportunities that connect the work of students with cutting-edge faculty scholarship. This way we develop student mastery of the key concepts and competencies of our disciplines.

Fortifying agency and identity. Persistence and success in STEM rest not only on access, opportunity, and knowledge, but also on the actions taken by individual women in particular environments using specific social understandings. Smith faculty adopt a guiding principle that students’ mind sets, metacognition, and identity development are essential to learning as well as professional and personal fulfillment. We understand that through our cultivation of students’ agency, confidence, and resourcefulness in learning, we will foster their sense of identity as scientists.

Celebrate “Women in Science” with us!
Undergraduate research is a high-impact educational practice in which the sciences at Smith have considerable expertise. Research experiences and collaborative projects that occur in research labs, the Science Center's five multidisciplinary research centers, and our field research sites demand the applied and integrative learning that deepens student engagement and learning (AAC&U, 2011). The sciences at Smith have a strong history of providing meaningful research opportunities to students, with a thriving honors program, active faculty research labs in which students participate as collaborators, and almost 50 years of a vibrant Summer Research Fellowship (SURF) Program. Our students present their research in many venues, including at the annual campus-wide Celebrating Collaborations exhibition and Smith in the World Conference, in public honors thesis presentations, and at regional and national professional meetings in their disciplines. With at least one undergraduate student co-author on a third of science faculty members' peer-reviewed scholarship and SURF participation doubling the likelihood of our students pursuing a graduate degree (data provided, Smith College Institutional Research, 2016), we believe that challenging our students to work at the cutting edge of knowledge helps prepare them for their lives beyond Smith.

Women in Science 2016 summarizes research done by Smith College’s Summer Research Fellowship (SURF) Program participants. Ever since its 1967 start, SURF has been a cornerstone of Smith's science education. In 2016, 150 students participated in SURF (144 hosted on campus and nearby field sites), supervised by 56 faculty mentor-advisors drawn from the Clark Science Center and connected to its eighteen science, mathematics, and engineering departments and programs and associated centers and units. At summer's end, SURF participants were asked to summarize their research experiences for this publication.

We have many reasons to be proud of our 2016 SURF researchers.

- SURF researchers worked on some of the biggest research challenges of our times, including eradicating human disease, reexamining human life and the earth around us at the nano-scale, documenting climate change and its impact on the living world, testing and improving sustainable energy technologies, and developing materials and testing methods not just for earth but also for use in space.

- SURF research took place not just on the Smith campus, but in a wide variety of research settings in the wider world: including, locally (study of local Hemlock forests, wildlife diversity, and water quality at the Ada and Archibald MacLeish Field Station in West Whately, MA and sediment management in Paradise Pond on campus), nationally students worked on projects with NOAA scientists, such as the National Estuarine Research Reserve and the National Institute of Standards and Technology. Internationally students study coral reefs and deliver community conservation education to school children and their families in Belize.

- Technical know-how, quantitative literacy, and presentation skills grew as students used state-of-the-art instrumentation, analyzed data with specialized software, and presented their results in lab meetings, posters, and conference presentations. In lunchtime workshops, students developed personal narratives and made presentations to peers and visitors.

- SURF students learned how to work with mentors and peers and, for some, across the boundaries of academic disciplines with other research teams. More experienced undergraduate researchers learned how to mentor others and take on research leadership roles.

We are excited about what SURF participants say they learned from SURF:

- ACCESS FOR ALL
  
  - I had the outstanding privilege of attending The Biophysical Society's Motor Conference in Vancouver, where I met so many accomplished scientists and learned more about their research. The individuals I met there were truly inspiring, but even more than them, I was mostly inspired by my fellow Smithies who so gracefully presented their projects at the conference. Although they were the youngest presenters (and scientists) at this meeting, they received so much attention because of how interesting and significant their results are! I learned that greatness can truly be achieved at any level of schooling and that hard work, dedication and passion are the major keys!
Beyond question, the Summer Undergraduate Research Fellowship has opened doors for me to the rewarding career of research that I have always aspired to enter.

Before I began my work this summer I was particularly worried that this area of my research would hold me back due to my learning disability, which makes it hard to focus on and contextualize scientific writing. By working diligently and being patient with myself, I was able to develop strategies that allowed me to read and quickly gain information from scientific papers with confidence. This skill has proved extremely useful for my continuing research and I believe it will serve me well as I finish my time at Smith and move forward with my personal and career goals.

**ENGAGING THE WORLD**

While working on my SURF project this summer, I learned a lot about working in tropical marine research environments, including research techniques. I also learned a lot about the Mesoamerican Barrier Reef, and the social and environmental factors affecting conservation law in Belize. In addition to the research skills and field related knowledge I gained, I also learned a lot about engaging with cultures outside of my own, and working in a team setting.

This project allows me to apply what I learned in classes back at Smith to a real world problem.

I learned new protocols and adapted them to how they worked best for me. It was necessary for me to take this time to not only learn the procedures but to understand why my research is important and how it has a bigger picture.

**KNOWLEDGE AND SKILLS**

I learned much more about surveying and measuring streamflow than I anticipated. I feel comfortable working with surveying equipment and inputting data into GIS software, and I didn't know that the project would involve the use of drones. The project also included lots of working outside in public places, which led to more interaction with the general public during the course of the summer, which helped me learn to tailor my description of my experience to the different people that came to me with questions. The visibility of this project in particular was both a blessing and a curse, in that the students involved were interviewed for several college publications in order to get out the word of why the pond is important and how we are working to maintain it. In any science career, but research in particular, it's important to recognize that you will face opposition from people who disagree with you or don't understand your methods. That's a lesson I had the fortune to learn very early on, because my project had such a visible impact on one of the key selling points of the campus. Working with CEEDS, the Greene Court Gate, and College Relations to spread awareness of the end goals of the project and its benefits long term was a great learning experience for me as a budding scientist, and it was also something I hadn't even thought to consider before.

This summer taught me a lot about what goes into land and research management, from understanding what is happening on site for scientific studies to community use of and concerns over land. I also gained skills in ArcGIS software, which allowed me to understand the software for future use and generally learn how to better maneuver my way around computer programs. The independence my experience gave me in working on projects, learning software and working my way into understanding the politics of land management have really opened my eyes to a whole other portion of the environmental science and policy world, which is what I had really hoped to learn. I now feel that I am much more aware of some of my options for a career in this field, as well as understanding my own abilities in working with community members, scientific equipment and programs to further my engagement in the science world.

**FORTIFYING AGENCY AND IDENTITY**

While my new understanding of proteins alone fulfills what I had hoped to gain from SURF 2016, I learned so much more in the process. This summer I learned a wide variety of techniques, read a lot of primary literature, and most importantly, I learned how to be a scientist.
- Perhaps most importantly, I learned how to act independently and think critically about the problems I faced; this ultimately allowed me to find a new, unique approach to reaching my project’s overall goal.

- It gave me confidence in my skills as a researcher and helped me decide that I do want to further my education and that I love water quality work. The work was equal parts rewarding, challenging and encouraging. It helped me reinforce the practical skills I learned in class labs during my undergraduate experience and helped me understand how other fields (biology, statistics, chemistry, etc) tie in with my engineering expertise.

- This experience showed me what it would be like to continue doing research in Social Psychology and how much of it is trial and error.

- This summer’s SURF experience taught me more than I could have ever imagined. At the start I thought that it would be a gradual integration into the lab assisting on projects but it turned out to be immersive from the first day to the last day. I was given the freedom to create and carry out experiments and learned through trial and error with assistance as needed. It taught me not only invaluable skills such as in situ protocols and alcian blue protocols, but it also instilled in me a profound confidence in myself as a scientist. It inspired me to continue my SURF project into the school year and gave me a more in depth understanding of both neuroscience and developmental biology. It has opened my eyes further to the career path I hope to pursue and the steps necessary to attain my future goals. I had hoped to learn more about where my research interests lie within the STEM field which was a definite success. This was one of the greatest experiences of my academic career thus far, I feel it was an invaluable part of my education and I can’t wait to continue my project in the coming school year.

- **PERSISTENCE OF STUDENTS**

  - With all failures comes success, which was truly learned this summer. I learned more about research and working in a lab even though I have been involved in research before. I learned to keep in mind how certain things won’t go as scheduled at times due to unexpected events. I had hoped to learn new lab techniques and sharpen the skills that I already had, and I am happy that I was able to do that since I had to work full-time in the lab unlike the times I worked during the school year. It was valuable to have learned more about my research project which will be developed into my honors thesis.

- **DIVERSITY OF STUDY**

  - For my internship at the Chesapeake Bay National Estuarine Research Reserve in Virginia (CBNERRVA) stewardship office, I hoped to learn about the day-to-day life of working in a long-term habitat monitoring program. I was interested in what kind of monitoring efforts were conducted, how the work in the field was conducted, and what kind of individual creativity and influence employees have in the program. During my ten weeks, that was exactly my experience. I was able to go in the field 3-4 days a week, which was so much fun. I loved being outdoors and having all the hands-on experiences. I learned many practical field skills and was exposed to all the different types of research which occur at CBNERRVA. I also learned some new lab techniques and processed some of the samples I collected. I developed a protocol for establishing a nest protection program of diamondback terrapins. The diamondback terrapin is a species with “very high conservation need” in Virginia. This project gave me the opportunity to think creatively, dive into the literature and produce a useful product which will be used to begin this program at CBNERRVA in the summer of 2017.

  - I learned how to navigate a project on my own and teach myself a programming language to facilitate data processing. This was an excellent confidence builder and will allow me take on similar endeavors in the future.

  - This summer’s SURF experience enabled me to build upon the skills that I developed over my last few years at Smith College. I continued to do field work and develop my skills as an ecologist, as well I learned about the process of writing, formatting, and submitting scientific papers for publication.
• UNEXPECTED BENEFITS

- I was really pleased with the hands-on experience I got in the lab. I learned more about using instruments and doing complicated calculations and I feel as though my confidence grew in important ways.

The comments are drawn from two surveys SURF participants completed in August 2016. First was a Smith survey (unpublished) in which students were asked what they had learned and how they expected to build on their SURF experience. Second was the SURF III national survey administered by Grinnell College (Smith student comments extracted from an anonymous summary provided by the survey administrators). See the following reports of the SURF survey data across a large number of colleges and universities: Lopatto, D. (2004). Survey of Undergraduate Research Experiences (SURF): First Findings. Cell Biology Education, 3, 270-277 and Lopatto, D. (2007). Undergraduate research experiences support science career decisions and active learning. CBE - Life Sciences Education, 6, 297-306.

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Smith College offices and units:

Center for the Environment, Ecological Design & Sustainability (CEEDS)
Committee on Faculty Compensation and Development (CFCD)
Provost's Office
Facilities Management

Some undergraduates were able to participate in summer research internships away from campus with support from the PRAXIS Internship Program.

We wish to recognize and express gratitude to the faculty members and staff who provided supervision, guidance, encouragement and support to SURF participants in the lab, doing field research, on-campus, and away from campus. SURF would not be possible without your devoted and generous contributions.

Thank you, Smith College students, faculty, staff, friends, and benefactors. It truly takes a diverse and dedicated community to sustain a program like SURF.

Margaret Lamb, Ph.D.,
Administrative Director,
Clark Science Center
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Development of Immunoprecipitation Protocol for Heat Shock Protein 25 Interactome Mapping

Daisy Crego/2016

Establishing an immunoprecipitation (IP) protocol is critical to mapping the cellular interactions of heat shock protein 25 (Hsp25), a ~25 kDa protein found in skeletal muscle that has been directly linked to heat stress. Hsp25 plays a role in cellular homeostasis and in times of stress influences actin polymerization. Mapping the Hsp25 interactome in skeletal muscle may inform the regulation of the Hsp25-mediated pathways and its linkage to related protein networks, for example estrogen-mediated pathways.

The use of protein A/G-bound magnetic beads for the IP was inspired by an investigation into the interaction of Hsp25 with F-actin in neuroendocrine PC12 cells. Several modifications to this protocol enhanced the Hsp25 IP, primarily the use of TBS-T rather than RIPA or myosin extraction buffers.

Also this summer, there was a transition in the Scordilis lab from methanol to methanol-free transfer buffer. This both increased the number of times a buffer can be used before disposal and thereby decreased the production of hazardous waste. Transfers in methanol-free buffer confirmed that blots of acceptable and reproducible quality could be obtained. Furthermore, the methanol-free transfer buffer can be used more than 10 times before it needs to be replaced (compared to a maximum of 4 uses of methanol transfer buffer).

Immunoblots were performed to confirm that Hsp25 extracted using TBS-T was present in high enough levels for detection by the primary antibody used in the IP. Once it was confirmed that the desired primary antibody could produce reliable blot results, initial IPs were performed. The four elution buffers used for these initial IPs contained 20 mM Tris-HCl and salt concentrations that ranged from 50 to 500 mM NaCl. Blot analysis of the washes suggest that Hsp25 was eluted primarily in the (Figure 1 lanes 6 and 7). However, repetition produced inconsistent results. Further work will determine the necessary buffer concentrations to elute Hsp25’s unknown binding partners. This work will continue in the fall.

![Figure 1: Immunoblot of immunoprecipitation washes (4 elution buffers contain 20 mM Tris HCl and between 50 and 500 mM NaCl). Lanes 2-5 show muscle extract supernatant elutions (50, 100, 150, 500 mM NaCl respectively). Similarly, lanes 6-9 show pellet elutions. Arrow points to Hsp25 band. Blot developed in rabbit anti-Hsp25 primary antibody (SPA-801, Enzo) and goat anti-rabbit HRP-coupled secondary antibody.](image)

(Supported by the Blakeslee Fund in the Biological Sciences)

Advisor: Stylianos Scordilis, Biochemistry

References:


The goal of this project is to establish a proteome profile for the rat plantaris muscle as a baseline for subsequent studies on aging and diet. The proteome is a “snapshot” of proteins present at a specific time point since the abundance of proteins may vary due to many factors, for example stress, diet or aging. Two-dimensional gel electrophoresis is used to map the proteins. Landmark proteins within the control gel can then be used to analyze protein abundance changes within experimental groups.

Rattus norvegicus plantaris was extracted in a buffer that solubilizes non-sarcomeric proteins (total protein estimation by the Lowry assay). Separation by isoelectric point and then by molecular weight in two dimensional gels was followed by spot excision and In-Gel trypic digestion of the spots of interest. These samples were identified by nano-scale liquid chromatography-mass spectrometry (LC/MS) and subsequent data mining using the rat genome.

Analysis of the extract supernate identified various glycolytic proteins present in high abundance (Figure 1), as well as creatine kinase, ATP synthase, and a fertility protein. Proteins found in all muscle samples are used as “landmarks” that allow selection of proteins that change two-fold in abundance in any experimental groups.

This is the initial step for the major project. A “master” gel is composed by overlaying multiple two dimensional gels from control samples to create a theoretical gel with all matching spots. The master gel is compared to experimental groups to identify spots that have changed in intensity by two-fold with a p<0.05. After determining the statistically significant spots in the experimental groups, spots will be excised and identified using LC/MS.

(Supported by the Howard Hughes Medical Institute)

Advisor: Stylianos Scordilis, Biochemistry

Fig. 1
Immunoblotting (or Western Blotting) is a common and powerful tool in biochemistry. Developed by Towbin et al. (1979), it allows for the quantitative analysis of specific proteins in biological samples. The procedure requires samples separated by apparent molecular weight with SDS polyacrylamide gel electrophoresis to be transferred from the gel to a hydrophobic membrane (PVDF). This membrane is then treated with a blocking solution and then with a protein-specific antibody to determine the relative or absolute amounts of the protein in the original sample. The transfer step from the gel to the PVDF is crucial to the procedure, as the amount of transferred protein is what determines the intensity of protein band in the final blot. It is performed by placing the gel on the PVDF in a transfer apparatus and running a current through the buffer solution to transfer the negatively charged proteins from the gel to the PVDF. The more effective the transfer, the more precise the quantitative analysis of the specific protein is.

The Scordilis laboratory makes frequent use of a modified Towbin procedure which calls for a transfer buffer containing methanol; this buffer can be used up to four times before losing efficacy. However, the literature provides no concrete explanation for the presence of methanol in the buffer. Presumably, it was used in the original experiment to offset the effects of using SDS in the buffer, which caused proteins to move too quickly from the gel to the membrane. With the necessity of methanol in question, we tested the effectiveness of methanol-free transfer buffer (using an equal volume of water to replace the methanol) against the standard buffer, by performing simultaneous transfers using gels that had been divided in half after separating standard skeletal muscle proteins in them.

After multiple simultaneous experiments using the two buffer systems, we determined that the methanol-free buffer was significantly more effective than the standard methanol buffer that has been used classically (Figures 1 and 2). We intend to test the effectiveness of this buffer on proteins of lower and higher molecular weights than those in the figures, but the results of these experiments provide significant motivation to change our protocol. An unintended consequence of the methanol elimination is that the waste transfer buffer is no longer a hazardous waste.
Figure 1: Stained SDS gel with different loadings of skeletal muscle extract (supernate and pellet) after transfer. Lanes 1-4 and 5-8 were loaded identically and electrophoresed, then transferred for the same time: 1-4 in standard buffer and 5-8 in the methanol-free buffer. The bands show the amount of protein still left in the gel (i.e., how much did not transfer to the PVDF).

Figure 2: Immunoblots using an actin antibody after transfer from the gels in Figure 1. The intensity of the detected antigen bands is proportional to the amount of successfully transferred protein. Far more protein transferred in the non-methanol buffer.

(Supported by the Schultz Foundation)

Advisor: Stylianos Scordilis, Biochemistry

References:

Identification of Proteins in Plantaris Muscle in Rats

Marguerite Pacheco/2019

Skeletal muscle tissue is composed of a myriad of proteins. Proteins facilitate most reactions in the body and by seeing what proteins are present in a muscle one can see the body’s activities in that moment. Studying protein composition of muscle provides the basis for comparisons between muscles, interactome research, etc. This project studied the protein composition of the plantaris muscle in a rat test subject.

The main way to establish protein abundance and identity is through a 2-D gel, but there are several steps to get to this point. The proteins were first extracted from a skeletal muscle sample of the plantaris, a muscle in the lower leg. An extraction buffer was used to suspend the homogenized tissue sample. The samples were centrifuged to separate the soluble, sarcomeric, proteins from the remaining insoluble proteins. Then, an equal amount of both pellet and supernate were used to rehydrate a gel strip each. These strips were loaded into an IEF cell, separating proteins by the collective charge of their respective amino acids. The strips were inserted into a gel that separates the proteins based on molecular weight. After a staining procedure one sees spots of proteins at a specific molecular weight and pI (charge). The spots are identified via LCMS.

The above gel shows the thirty-four protein spots that were most defined. Some spots had more than one ID match in which case the match with highest coverage percentage was chosen. The LCMS data lists the pI and molecular weight of the predicted protein, allowing one to cross reference it to the location on the gel and establish more certainty.

That we were able to produce the above gel speaks volumes; the proteins can be solubized uniformly, the proteins “focus” well, and the proteins identified in the gel match the approximate physical area in which they were found. This proves that for the muscle and specimen chosen, the protocol we would like to use gives us reproducible and verifiable data from start, solubizing, to finish, identification. The identifications proved helpful in showing that within this muscle we found proteins such as CK-M, a metabolic protein used in the anaerobic production of energy for the body, enolase, a protein used in glycolysis, and ATP synthase, a protein that produces massive quantities of energy for the human body. The presence of these proteins lets us know how successful this protocol is and that it can be used for future research with our current gel as a base model.

(Supported by the Howard Hughes Medical Institute)

Advisor: Stylianos Scordilis, Biochemistry
Dynein is a molecular motor protein responsible for intracellular transport of membrane-bound materials, facilitating chromosomal segregation, cell division and nuclear positioning in eukaryotic organisms. In achieving intracellular transport, dynein walk along cytoskeletal microtubules while simultaneously carrying and delivering cellular cargo. This molecular motor's ability to convert chemical energy in the form of ATP to mechanical energy enables its travel along minus-ended microtubules.

The peculiar structure of this motor protein also plays a vital role in its ability to walk along microtubules. Dynein is a large homodimer comprised of polypeptide subunits that constitute two identical heavy chains, each of which contain enzymes necessary for ATP hydrolysis, and are attached to individual globular motor domains. It also has a coil “stalk” that binds to the microtubule domain and a stem, responsible for attaching itself to neighboring microtubules. ATP hydrolysis generates energy that allows dynein to step in increments of 8-32 nm by alternating its two motor domains in either a hand over hand or inchworm stepping fashion.

In vitro experiments revealed that yeast and mammalian dynein entertain significantly different motile properties, despite their structural similarities. Mammalian dynein move faster than yeast dynein while yeast dynein travel longer distances along microtubules.

The focus of my project is to investigate the similarities and differences between yeast and mammalian dynein. In vitro experiments will reveal whether or not the binding domain is the suspect region responsible for the differences in motile behavior between the two kinds of dynein. This research requires recombinant protein engineering to develop hybrid dynein. In developing these hybrid dynein, the yeast microtubule-binding domains are replaced with mammalian ones. This summer I spent time comparing the distances traveled and average velocities of the hybrid proteins with those of the unmodified ones using a TIRF microscope to better understand their behaviors with respect to one another and to assess the significance of a small variation in the amino acid sequence of the microtubule-binding domain. I was also able to work with segmented rigid and flexible chassis, synthetic cargos made from DNA origami, which allow researchers in the Derr lab to study the motile properties of several motor proteins walking together collectively. I was able to conduct experiments in which I used 7-motor-rigid chassis to test whether or not the hybrid dynein would exhibit an increase in velocity or run length when working together in an ensemble. As I continue this research during the school year, I hope to learn more about what factors contribute to the differences in processivity between yeast and mammalian dynein.

(Supported by the Howard Hughes Medical Institute)

Advisor: Nathan Derr, Biological Sciences

Image 1: A kymograph of the control, a modified yeast motor containing a yeast microtubule-binding domain, used in in vitro assays. A total of 109 runs were analyzed to obtain an average velocity of 118 nm/s for the control motor in vitro.
Symbiotic Relationships in *Chilodonella uncinata* and *Spirostomum* sp.

Jacqueline Banuelos/2017 and Angela Lool/2017

Ciliates are highly evolved single celled microorganisms that are understudied. This summer the Katzlab worked on a project titled Symbiosis in Ciliate Species. The research studied *Spirostomum* species and *Chilodonella uncinata* in order to understand the relationships between the ciliate itself and its environment in regards to bacterial interactions. Endo and ecto-symbionts were found to be a possible phenomenon in these ciliate species. Endosymbiosis refers to bacteria that lives inside the ciliate either as a mutualistic, parasitic, or obligatory relationship, as opposed to ectosymbiosis where the organism lives on the outside of the ciliates 1,2. Based on prior literature results, it was hypothesized that *Spirostomum* species and *Chilodonella uncinata* have a bacterial endosymbiotic relationship that serves mutually in order to increase the survival of both organisms.

This experiment required single celled PCR, which entailed of picking both species with a mouth pipette. To ensure samples were non-contaminated the organisms were washed multiple times using filtered pond water. PCR was used to amplify the DNA using bacterial and eukaryotic primers (27YMF and 1492R). We ran a Gel electrophoresis in order to analyze whether bacterial DNA was present among the sample. When successful, cloning via gel isolation was used to sequence each individual sample. Once cloned and harvested, we selected for 12 individual colonies to which we prepared by using the method of Pick and Swish using LBK broth as the nutrient rich media to grow the cultured bacteria. Miniprep samples were prepared to a 96 well plate using the Direct Prep 96 Miniprep Kit in order to be sequenced.

Sanger sequencing allowed the samples to be analyzed using the BLAST database to find the most similar matches to bacteria in the sample. Preliminary results provided polynucleobacter with a >98% match to the samples. Polynucleobacter was found to be planktonic and free-living heterotrophic bacteria. It also has an obligate symbiotic relationship with *Euplotes*, another type of ciliate3. Next step for this project is to be able to reproduce the experiment for validating results. Not only do we want to replicate this with the *Spirostomum* species, but also repeat the experiment with *Chilodonella uncinata*.

This work will be continued into 2016-17 with special studies hoping to obtain more concrete results for both *Spirostomum* sp. and *Chilodonella uncinata* using Sanger sequencing and Fluorescence in situ hybridization. We hope to present our findings at Smith Collaborations in the spring semester.

(Supported by the National Science Foundation)

Advisor: Laura Katz, Biological Sciences

References:


Figure 1. Gel image of *Spirostomum* sp. Single Celled using PCR bacterial products (lanes 1-12) and positive bacterial primer (lane 13).
The red-backed salamander (*Plethodon cinereus*) is considered a forest floor keystone species and an important indicator of ecosystem health in the Northeastern United States due to its wide distribution, high-density occurrence, and established presence in the literature. *P. cinereus* has demonstrated a preference for eastern hemlock (*Tsuga canadensis*) conditions over those of mixed deciduous stands. The recent infestation of populations of *T. canadensis* by the invasive hemlock woolly adelgid (*Adelges tsugae*) is predicted to severely impact the structure of New England Hemlock stands, and could dramatically shift the dynamics of forest floor animal communities that depend on Hemlock understory, leaf litter, and soil conditions. This study aimed to investigate the ecological niche of *P. cinereus*, relative to bottom-up (e.g. *T. canadensis* associated) and top-down (e.g. predation pressure) forces using two study sites: the Smith College Ada and Archibald MacLeish Field Station in West Whately, MA, and a private residence in Chesterfield, MA. At each location, artificial cover objects (ACOs) were placed in adjacent plots of *T. canadensis* and Black Birch (*Betula lenta*) and monitored from June of 2014 to early May of 2016 for *P. cinereus* during summer field seasons. In addition to assessing *P. cinereus* substrate preference, we also explored influence of predation pressure by the Northern Short-Tailed Shrew (*Blarina brevicauda*) on *P. cinereus* distribution, as a means of determining the relative importance of bottom-up and top-down drivers on distribution and density of *P. cinereus*. On average, *P. cinereus* was 2.2x more frequent under ACOs in *T. canadensis* plots than *B. lenta* plots, and were 2.4x and 1.9x more likely to be found under ACOs without small mammal tunnels in Chesterfield and at the MacLeish Field Station, respectively. *P. cinereus* was also statistically more likely to share an ACO with its competitor, the Carabid ground beetle (Fig. 1). This may indicate the presence of overlapping zones of enemy free space in the *P. cinereus* and Carabid landscapes of fear. As such, the results highlight similarly strong effects of bottom-up versus top-down forces in determining the ecological niche of *P. cinereus*, and suggest a strong role for predation risk and enemy free space in affecting the distribution of *P. cinereus*. This study characterizes the interactions between *P. cinereus* and the most immediate members of its food chain, and puts this system into the broader context of *T. canadensis* decline and *B. lenta* succession.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Advisor: Jesse Bellemare, Biological Sciences

Figure 1. Spatial association between *P. cinereus* and Carabid ground beetles across 8 surveys.
Three Amigos: Interactions Between Three Herbivorous Gastropods in Rocky Intertidal Habitats

Alysha Putnam/Grad, Dara Brena/2017, Anastasia Konefal/2017

*Littorina littorea*, *L. obtusata*, and *L. saxatilis* are the only littorinid herbivorous gastropods found along New England coastlines. All three species occur in rocky intertidal habitats and show distinctive patterns of distribution: *L. saxatilis* occurs in the high intertidal zone; *L. littorea* shows widespread distribution throughout the intertidal regions; and *L. obtusata* is primarily found on fucoid algae in the high to mid intertidal zone. We explored interactions between these three closely related herbivores with laboratory and field experiments.

Because of their overlapping distributions, we conducted field experiments to investigate interspecific interactions between *Littorina littorea* and *L. obtusata* in Rhode Island and Maine. Treatment conditions included: control area (no density manipulation), and removal or addition of the congener in areas of high food availability. In both Maine and Rhode Island, removal of *L. littorea* from high and mid intertidal areas resulted in a significant increase in *L. obtusata* densities, while neither addition nor removal of *L. obtusata* had any effect on *L. littorea* abundance (Figs. 1 and 2). Thus, *L. littorea* exerts interspecific competition, likely via interference, on *L. obtusata*. We also conducted transplant experiments to assess growth rates of field populations of *L. littorea* at two sites varying in food resources. Control snails remained at their home site, while experimental snails were transplanted. *L. littorea* showed significantly lower growth at the site with low macroalgal abundances, indicating food limitation, and potentially strong intraspecific competition.

In the lab, we conducted paired-choice experiments to determine the dietary preferences of the littorinid species for *Fucus vesiculosus* vs. *Ulva lactuca*, both commonly occurring macroalgal species. *Ulva lactuca* was the preferred food of *Littorina littorea* ($t = 5.1, P < 0.0003, Fucus$ vs. *Ulva*), while *L. obtusata* showed little or no grazing on this species. In contrast, *Fucus vesiculosus* was highly preferred by *L. obtusata*. These results demonstrate clear resource partitioning between these two species. While *L. saxatilis* preferred *F. vesiculosus*, it showed low consumption of either macroalgal species. Instead, this herbivore prefers microalgal turfs growing in the high intertidal zone, thereby having limited interactions with the other two littorinid species. Our field and laboratory experiments highlight the complex interactions among these herbivorous gastropods for food and habitat resources.

*(Supported by B. Elizabeth Horner Fund in the Biological Sciences, Grantham Fund, Choate Fund)*

Advisor: Paulette Peckol, Biological Sciences
Figure 1. Mean (± SE) abundances (number/m²) of *Littorina obtusata* in high- and mid-intertidal areas after the manipulation of *L. littorea* densities at Pemaquid Point, Bristol, Maine (A) and Fort Wetherill, Jamestown, Rhode Island (B).
Figure 2. Mean (± SE) abundances (number/m²) of *Littorina littorea* in high- and mid-intertidal areas after the manipulation of *L. littorea* densities at Pemaquid Point, Bristol, Maine (A) and Fort Wetherill, Jamestown, Rhode Island (B).
Mapping Habitat Change and Sea Level Rise in the Waquoit Bay Estuary, Cape Cod, MA

Chloe Brownlie/2017

My internship at the Waquoit Bay National Estuarine Research Reserve (WBNERR) in Falmouth, MA included a variety of responsibilities in the lab and in the field, however my main project focused on using ArcGIS to map changes in pool development in the salt marshes in the Waquoit Bay estuary. The reserve has been working on this habitat change analysis for years, looking at the 4 main components of habitat change in the marsh: landward migration of the upland border, dune vegetation changes, seaward edge erosion, and pool development. Aerial photos were taken of the estuary in 2004 and 2012 and divided into fine segments by the NOAA Office of Coastal Management (OCM) to delineate different habitats in an unbiased manner. When I arrived, the 2012 files had been edited to account for the pools. I used 2014 groundtruthing data to classify unknown polygons from the 2012 aerial photos. When I arrived, the 2012 files had been edited to account for the pools. I used 2014 groundtruthing data to classify unknown polygons from the 2012 aerial photos. Then, I used data from WBNERR’s 2012 vegetation survey along with the aerial photos to create an accuracy matrix for the segmentation and groundtruthing.

Upon completing the accuracy matrix, I edited the 2004 aerial photos to locate and assign numbers to all the pools in the marshes surrounding Waquoit Bay, using the segmentation that had been done on the photos. Since there was no groundtruthing done for the 2004 photos, all unknown polygons will remain unknown. The next steps of this project include sending the new data layers I created for 2012 and 2004 to the OCM for them to run a change analysis on the pools in the marsh. From cursory comparison of the 2004 and 2012 photos, there are far more and larger pools in 2012 than in 2004. The change analysis will help quantify the changes in number and size of pools and identify what areas of the marsh are affected the most. My work on this project has finished the preparation of the 2004 and 2012 aerial photo datasets necessary for the 2004 to 2012 change analysis. Once completed, the change analysis will show whether some sections of the marsh are degrading faster than others. WBNERR will develop a plan to help the marsh in the face of these changes. When the next set of aerial photos are taken, the whole change analysis process will be repeated, comparing the future photos to the 2004 and 2012 datasets.

(Supported by the Agnes Shedd Andreae 1932 Research Internship Fund)

Advisors: L. David Smith, Biological Sciences, Environmental Science & Policy and Jordan Mora, National Oceanic and Atmospheric Administration (NOAA)
Assessing Macroinvertebrate and Mussel Health in Paradise Pond

Sarita Quimpo-Chiu/2018

Budget constraints have prompted studies to assess less expensive alternatives to the regular dredging of Paradise Pond. One such alternative is the simple opening of the pond’s sluice gates during high flow events to allow natural currents to carry sediment downstream.

An important consideration in using this technique is its effect on the health of the river ecosystem. This project aims to assess how river organisms are affected by this technique using the Before-After-Control-Impact design (BACI)\(^1\) method. The summer of 2016 was spent obtaining the “Before” sample.

We assessed the diversity and abundance of macroinvertebrates and mussels as a marker of river health. Aquatic macroinvertebrates (e.g. crayfish, worms, midges, etc.) were sampled at four sites along the Mill River via kick-sampling (Figure 1). Overall diversity and water quality at the sites were compared using the Shannon Diversity Index and Hilsenhoff’s Biotic Index (HBI)\(^2\). Water quality was evaluated by calculating the tolerance values of present macroinvertebrates, using tolerance values from the Biological Monitoring of Surface Waters in New York State.\(^3\) Mussel abundance and health were determined by measuring annual growth rings, lengths, and densities of mussels obtained via quadrat sampling, using the Manhan River as the control site.

No significant difference was found in taxa richness in either downstream and upstream sites, although the former is more diverse and even. Riffle habitats are significantly cleaner than run habitats. Upstream sites and riffle habitats have more environmentally sensitive orders of macroinvertebrates. The Mill River has a higher mussel density than the Manhan River, with 85.8% of sampled mussels alive at the former and 75.3% at the latter. Mill River mussels are also longer on average.

The comparison between this dataset and the “After” dataset to be obtained after pond sediment is allowed to flow downstream will hopefully show if this technique can be used as a viable alternative to dredging.

(Supported by the Smith College Facilities Management)

Advisor: Marney Pratt, Biological Sciences

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Figure 1. Sites sampled along the Mill River
The Effect of Polycyclic Aromatic Hydrocarbons (PAHs) on the Pharyngeal System

Gina Cho/2017, Emilie Jones/2018

Considered the largest oil spill in U.S. history and affecting a diversity of ecosystems, the Deepwater Horizon Disaster released 200 million gallons of crude oil into the Gulf of Mexico from its occurrence on April 22nd 2010. Crude oil components, specifically polycyclic aromatic hydrocarbons (PAH’s), are ubiquitous, yet hazardous, compounds. Naphthalene, a member of the PAH family, was found in especially high concentrations in crude oil released during the Deepwater Horizon Disaster. By using zebrafish as a model organism for vertebrate systems, our project has focused on how PAHs induce defects in craniofacial and cardiac structures, which arise from neural crest cells (NCCs), a type of multipotent stem cell that migrates through transient structures known as the pharyngeal pouches and arches. This summer has focused on identifying what specific genes that power pharyngeal system formation are affected by naphthalene.

Embryos treated with a 200 μM solution of naphthalene at 10 hpf were fixed at 30, 36, and 42 hpf and underwent in situ hybridization. We used probes for dlx2a, boc2a, boc2a, and jag1b, known genetic markers of the pharyngeal system. Imaging was performed using a dissection microscope.

Though naphthalene treated embryos labeled for boc2a and jag1b showed no difference in expression from the control embryos at any of the time-points, the treated embryos labeled for dlx2a (Figure 1) and boc2a showed reduced expression compared to the control embryos at all time-points. Thus, the in situ experiments suggested that though dlx2a, which promotes the formation of all arches and NCCs, and boc2a, which assists in the formation of all pouches, specifically the second pouch, are affected by naphthalene, boc2a, which leads to formation of the second pouch, and jag1b, which leads to the formation of pharyngeal endoderm, are not.

This project will be continued for Gina Cho’s thesis and Emilie Jones’s special studies for the 2016-2017 academic year. Though previous experiments have confirmed that naphthalene causes defects in craniofacial and cardiac structures of treated embryos, the specific molecular mechanisms leading to those defects from pharyngeal system defects are still unknown, and our project will focus on disclosing these mechanisms. Furthermore, as many more genes are responsible for pharyngeal system formation than the tested four, more in situ experiments will be performed to hone in on what genes are affected by the presence of naphthalene.

(Supported by the Howard Hughes Medical Institute)

Advisor: Michael Barresi, Biological Sciences

References:
Cytoplasmic dynein is a bipedal motor protein that walks on cytoskeletal filaments called microtubules. Dynein facilitates the transport of cellular cargoes, such as organelles and vesicles, inside eukaryotic cells. These cargoes are massive compared to dynein; therefore, hauling them requires a team of multiple dynein motor proteins. Research over the past decade has led to greater understanding of the mechanisms that govern individual dynein movement. However, less is known about how dynein motor proteins work collectively in groups to transport large intracellular cargoes.

To investigate the biophysics underlying the motility of motor ensembles, we have designed a DNA origami synthetic cargo. This allows us to control the number of dynein motors in the ensemble and vary the rigidity of the cargo chassis itself. Using total internal reflection fluorescence microscopy, we have observed ynein ensembles transporting these synthetic cargoes along microtubules in vitro. We find that ensemble motility depends on cargo rigidity – as the number of motors in the team hauling the cargo increases, ensembles transporting flexible cargoes move comparatively faster than a single motor, whereas ensembles transporting rigid cargoes move slower than a single motor. To explain this, we chemically induced individual motor pausing using an ATP analog called ATPγS.

We show that ensembles connected through flexible cargoes are less sensitive to the chemically induced pauses of individual motors within the ensemble. We conclude that cargo rigidity plays an important role in communicating and coordinating the states of motors, and consequently in the subsequent mechanisms of collective motility. The insensitivity of ensemble-driven cargoes to the pausing of individual motors contributes to the robustness and versatility of intracellular cargo transport. This work has been presented at the conference “Engineering Approaches to Biomolecular Motors: From in vitro to in vivo” and has culminated in a paper submitted for peer review.

(Supported by the Four College Biomath Collaboration)

Advisor: Nathan Derr, Biological Sciences

References:
Examining *Sciurus carolinensis* Over 150 Years

Isabella Fielding/2017

The eastern grey squirrel (*Sciurus carolinensis*) is a common feature of the landscape of the eastern United States. It is a species that has been viewed as both pest and pet but, more important to this study, has also sparked the interest of enough people for large collections of the species to have been accumulated across the United States, with specimens collected as early as the 1890’s. This study sought to identify relationships between body sizes, reproduction, locations, sex, and year of collection of eastern grey squirrels. Analyzing such relationships in a widespread yet local species such as *S. carolinensis* within a set time span can indicate how humans have impacted the landscape, either through rapid urbanization or climate change.

Data were collected from online museum databases or by visiting museums and recording the data located on specimen tags (Figure 1). Data collected included body size measurements, litter size, testes size, subspecies, and location and date collected. Relationships between variables were assessed through the computation of general linear models in Minitab 17.

While many results were obtained, there were a few that are especially noteworthy. Litter size decreased over time within the past 150 years, with the year accounting for 21.20% of variation in litter size. Also, significant relationships between sex, weight, and latitude were found. Specifically, there is a tighter relationship between weight and head body length in females than in males, and this sex difference is stronger at lower latitudes. Latitude and sex were found to account for 13.55% of the variation in weight to head and body ratios.

Smaller litter sizes over time may be an indication that humans are having a negative impact on *S. carolinensis*. Further data analysis, behavioral studies, and testing of female squirrels for traces of chemical ingestion may be a few ways to further explore this result. Also, while *S. carolinensis* are generally noted as not being sexually dimorphic, the results indicating relationships between sex, weight, and latitude challenge this idea. While this result does not have to do with the influence of humans on *S. carolinensis*, it is a reminder to challenge current knowledge to better understand the complexities of nature.

This study will be continued into the academic year as an honors thesis project in order to increase the database of *S. carolinensis* specimens, examine more variables such as habitat type, and further explore the relationships between data.

*(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)*

Advisor: Virginia Hayssen, Biological Sciences

References:


Investigating the Density of Rare Endemic Forest Plant Species in the Eastern US

Anna George/2017

Climate change, a looming threat to biodiversity today, is also thought to have caused large numbers of species to decline or go extinct in the past. Until recently, it has been somewhat of a challenge to study the some effects of past climate change on current biodiversity patterns due to the difficulty of defining species’ ranges. An emerging method for tackling range data collection for large numbers of species at once is to use the enormous pool of data on plants collected across their native ranges by botanists and preserved in herbaria at botanic gardens and universities. These records can allow scientists to study the geographic distributions of many species at once, and discover previously unknown patterns to biodiversity. My project this summer was part of a study on the biodiversity patterns of approximately 250 rare endemic woody and herbaceous plant species from the eastern United States. During previous unpublished research, Jesse Bellemare found that high plant diversity only existed south of the most recent glacial maximum in counties with large elevation ranges. In addition, he found that the herbaceous plants tended to have higher biodiversity further north, closer to the glacial maximum line than woody plants.

My job this summer was to clarify the biodiversity patterns and conduct further research on this group of plants. I created several different versions of biodiversity maps, correcting small errors in the data, and deciding how best to portray the biodiversity patterns in a paper. In addition, I determined the range sizes of each species and the mean of all herbaceous and woody species respectively. I found that the herbaceous species had a 54,000 km average range, while woody species had an 80,000 km average range. It is unclear so far why this difference is present, but ascertains that the ranges of herbaceous species are not further north simply because they are larger.

There are many more questions to be answered about the biodiversity of these species. I plan on studying this same group of approximately 250 rare endemic plant species from the eastern United States that I worked with this summer in an honors thesis. I will look at the seed-dispersal types of the herbaceous and woody species, and investigate their climate tolerances.

(Supported by the Strategic Environmental Research and Development Program)

Advisor: Jesse Bellemare, Biological Sciences

References:


Eastern Hemlock (Tsuga canadensis) forests in New England create a stable, cool and moist environment important to many organisms like the Red-backed salamander (Plethodon cinereus). However, Hemlocks are in decline due to two invasive insect pests: Hemlock Woolly Adelgid (Dreuxes tsugae, HWA) and the Elongate Hemlock Scale (Fiorinia externa, EHS). In western Massachusetts, hemlock decline is often followed by Black Birch (Betula lenta) growing in its place, a deciduous tree species which impacts the forest ecosystem in very different ways. The forest floor is thinner and breaks down more quickly because it is exposed to more sun making it drier.

The goals of this research were to look at P. cinereus distribution between plots of Hemlock and Black Birch, and how those bottom-up influences compared to top-down and intra-guild influences.

This research was conducted at Smith College’s MacLeish Field Station in Whately, MA and a site in Chesterfield, MA. Nine 10x15m forest plots in MacLeish, 3 birch and 6 hemlock; and 6 plots in Chesterfield, 3 birch and 3 hemlock have been established years beforehand. Each plot has 70 wooden cover boards spread across the forest floor. Once a month, a cover board observation survey was conducted at both sites where number of red-backed salamanders, carabid beetles, mammal tunnels as a proxy for short-tailed shrews’ presence, other notable organisms and soil moisture under the board were recorded. We found evidence of both bottom-up and top-down factors influencing salamander distribution. Red backs were almost always found in greater numbers in Hemlock forests with the exception of the MacLeish survey in May (Fig. 1), with the disparity in numbers increasing into the hotter, dryer months of June and July when hemlock trees’ ability to maintain a cooler and moister habitat serves as refuge for salamanders that can desiccate rapidly. When looking within a singular plot, however, red backs were less often found under boards that had mammal tunnels, but were regularly seen sharing boards with carabid beetles, intra-guild competitors that have been shown to be very territorial against salamanders, and vice versa. Most likely, both species rate the threat of competition less stressful then threat of predation, which the mammal tunnels imply. Exploring how red-backed salamanders’ landscape of fear and reaction to competitors in the absence of predation risk will carry on into the academic year, as will looking at if red backed salamanders can serve as indicator species for what to expect as hemlock forests decline and transition into mature birch forests.
Figure 1. The average number of red backed salamanders per all cover boards in a forest type within a study area. Both study stations, MacLeish and Chesterfield, have salamander averages shown once per month.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Advisor: Jesse Bellemare, Biological Sciences

References:
The Mill River Monitoring Project

Maya Hayden/2019

The Mill River deposits sediment that builds up over time in Paradise Pond, which needs to be dredged every 8-10 years. Due to the expense and disturbance of traditional dredging, another procedure was tried this summer. The pond was drawn down, and sediment was pushed into the fast moving water in the belief that the stream would move the sediment downstream. To understand the implications of this process on the ecosystem Dr. Marney Pratt, Sarita Chiu, and I collected macroinvertebrates and mussels data in the river. Both control and on-site impact data were collected for both organisms for the “before” of the monitoring project. The “after” segment is being done in the fall.

Mussels were sampled downstream of Paradise Pond (impact site), as well as in the Manhan River, which was used as a control. Mussels were collected, measured, and put back into the river. Secondly, kick net sampling was done at four sites on the Mill River for macroinvertebrates. An upstream and downstream riffle, and run, to insure the highest variety of organisms. The upstream was the control data, because it is not impacted by the sediment removal, and the downstream is the impact data, showing how the sediment effected the site. After being preserved, all the organisms collected were identified down to genus with dichotomous keys. After keying out each organism to the lowest identifiable taxonomic level, a water quality and Shannon diversity index can be done to determine the health of the stream.

The data collected this summer only shows the river before the movement of the sediment. After sampling has been done in the fall, conclusive results can be developed. After the data has been collected in the fall, we will be able to see clearly the impact that the sediment removal has had on the downstream ecosystem. Until then, we only have our “before” data. Hopefully, we will find that the procedure we have used to move the sediment will not have an effect on the life downstream, and we can continue to use this process in the future. During the course of this project I learned the detailed taxonomy of several different orders, including diptera, trichoptera, megaloptera. Learning how to use a dichotomous key was a challenge.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Adviser: Marney Pratt, Biological Sciences
Study and Identification of *Sphagnum* spp. Present at Hawley Bog and Correlations with Testate Amoebae

Mallory Kakley/AC 2017

*Sphagnum* moss is a bioengineer in bogs and fens in that it creates an acidic and eventually anoxic environment through the gradual leaching of calcium.1 However, there are many organisms including microorganisms that thrive in this environment. Some microorganisms, testate amoebae, are known to be top predators in these *Sphagnum* dominated environments. I sought to identify what species of *Sphagnum* moss are present at Hawley Bog in a comparative analysis of testate amoebae species present at correlating sites. There is an ecological relationship between *Sphagnum* species and testate amoebae that is understudied. I think that there are certain species of testate amoebae that correlate with certain species of *Sphagnum*. *Sphagnum* includes approximately 250–400 species and has a worldwide distribution.2 *Sphagnum* is also polymorphic with cryptic species and species that take express different morphs that are currently referred to as subspecies. DNA analysis through direct sequencing provides an accurate identification.

I sampled *Sphagnum* at Hawley Bog along three, twenty-one meter transects. The testate amoebae were counted and identified from each sample site by another student in Katzlab, Angela O’Donnell ‘18. I then worked with guidance from Jon Shaw’s lab at Duke University to identify what genes or regions of *Sphagnum* DNA would direct me to species identifications. I first used morphology to identify the species from the samples and selected primers based on the species I anticipated finding. I used a polymerase chain reaction (PCR) with known genetic data published through Genbank for references to amplify previously-characterized loci (both chloroplast and nuclear) for comparison to data on GenBank. I targeted a region known as trnG which is a transfer RNA gene found in the glycine of chloroplasts in *Sphagnum* spp. and another random region used for identification known as RAPD-F. The latter process is method referred to as the RAPD method or Random Amplification Polymorphic DNA. The results can vary greatly depending in the cycling conditions, concentration of the template DNA, and quality of the template DNA.3 I successfully identified three morphospecies through PCR and DNA direct sequencing. The species present so far are *S. magellanicum*, *S. capillifolium var. tenellum*, *S. rubellum*. There are still many more samples from this site that need to be identified. I will continue this project through my Senior Honors Thesis combining my data with the testate amoebae findings to determine if there are correlations of species.

(Supported by the Schultz Foundation)

Advisors: Laura Katz and Jesse Bellemare, Biological Sciences

References:

Migration Versus Mutualism: Can the Small-Ranged Endemic Plant Rhododendron catwabiense Form Specialized Ericoid Mycorrhizae with Soil Fungi North of its Native Range?

Elena Karlsen-Ayala/2016

The mutualistic interactions of plants with below-ground soil biota, such as mycorrhizal fungi, appear to be crucial for many species’ performance and local distribution. Such specialized relationships are also of concern at larger geographic scales in the face of anthropogenic climate change. For example, will rare or range-restricted plant species be able to successfully colonize new regions where co-adapted mutualistic partners from their native range might be absent?

In this study, we investigated the ecological interactions of a small-ranged endemic plant, Rhododendron catwabiense (Ericaceae), with soil biota from sites in the Northeast US, 800-1000 km beyond its native range in the Southeast. Members of the Ericaceae form highly specialized ericoid mycorrhizae with fungal mutualists, often allowing them to grow on nutrient-poor soils. To test how R. catwabiense might interact with soil biota beyond its native range, we inoculated 30 experimental mesocosms with organic layer material from the bases of two widespread, Northeast native Rhododendron species. Soil in half of the mesocosms were then sterilized to eliminate fungi and other soil biota. Fifty seeds were sown into each mesocosm, and seed germination, seedling growth rate, leaf tissue nutrient status, and colonization of roots by ericoid mycorrhizae were compared.

Germination rate was significantly higher (75%) in mesocosms with live inoculum compared to those that had been sterilized (55%). In the first two months of growth, sterilized soil seedlings grew significantly larger (mean largest leaf = 6.2 mm length) than those on live inoculum (4.1 mm). However, the high growth rate of seedlings on sterilized soil subsequently declined, while that of seedlings on live inoculum soil increased, resulting in comparable leaf size by the end of the growing season (10.3 vs. 10.6 mm). In addition, many seedlings on sterilized soil began to show qualitative signs of nutrient stress, such as leaf discoloration. Harvested and stained root samples revealed that seedlings on the live inoculated soil had successfully partnered with ericoid mycorrhizal fungi, despite the inoculum being sourced from habitats far beyond the native range of R. catwabiense. Overall, it appears that R. catwabiense can readily partner with new mycorrhizal mutualists far outside its native range. This result is important, as several recent studies have otherwise shown high degrees of local co-adaptation between plants and mycorrhizal fungi. The presence of other Rhododendron and Ericaceae plant species in new regions, and their associated ericoid mycorrhizal communities, might facilitate colonization by migrating species.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Advisor: Jesse Bellemare, Biological Sciences
Migration versus mutualism: Can the small-ranged endemic Rhododendron ponticum form specialized endomycorrhizal with soil fungi north of its native range?

Elena Kuprianova, Sara Chen, Marthe Varenier, and Irene Bethmann, Dept. of Biological Sciences, Smith College, Northampton, MA.
Competitive Advantages of Autumn Olive: Implications for Expanded Invasion in Response to Increasing Environmental Pressures

Windy Kelley/2017

Plants have been known to change biophysical traits in response to extreme variations in temperature and soil composition (Alameda and Villar, 2012; Lambers, Hans, et al. 2006; Pregitzer, Kurt S et al., 2000). Once landscapes are converted from their natural state, non-native plants seek out available resources and displace native species. Invasive plants may possess a specialized ability to maximize underground biomass that allows for them to capitalize on these disturbed habitats. How abiotic factors impact underground biophysical change is a necessary component to understanding the effects of climate change on growth success rates, competitive advantages over native communities and expanding geographic ranges of invasive plants.

We investigated how temperature, soil composition and characteristics impact the biophysical changes in roots and nodule structure of a dominant invasive plant, Elaeagnus umbellata (autumn olive). Differences in underground root architecture were investigated by collecting autumn olive biomass from seven disturbed habitats, starting in October 2014 through to August 2016. Multiple shrub and soil samples were collected from each of the sites along a natural temperature gradient in Western Massachusetts and west into Pennsylvania. These sites were expected to differ by soil composition and temperature. We analyzed shrub samples for root nodule size, abundance, and proportion of root weight, as well as rooting structure characteristics. Soil samples were analyzed for percent organic layer, estimated nitrogen release, pH values along with other soil factors.

Our research addressed how disturbance, changes in temperatures and soil characteristics may benefit autumn olive providing an elevated competitive advantage. The goals for collecting samples enabled an in depth exploration into expected variances in root characteristics between disturbed habitats.

Root and nodule characteristics were closely related and differed between study sites. Mean root nodule abundance, mean root maximum diameters and mean minimum diameters demonstrated significant differences between sites (Figure 1). These results highlight the ability of autumn olive to adapt to changing biotic and abiotic factors by channeling its energy to the root system. Mean seasonal temperatures demonstrate significant differences between sites highlighting autumn olive’s ability to direct energy to the most beneficial/required area of the plant’s system. During spring energy is directed to nodule size (Figure 2) however, root diameters are greater throughout the fall suggesting less energy is being directed towards nodule production (Figure 3). Soil analysis suggests older, less “disturbed” sites had significantly greater organic matter. These results suggest changing soil composition between differing types of disturbed habitats and that soil characteristics may play a larger role in the differences found between samples.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Advisor: Danielle Ignace, Biological Sciences

References:
Figure 1: Bar graph showing mean number of root nodules per site with mean number of root nodules on the y-axis and site represented by different colored bars on the x-axis.
Figure 2: Bar graph showing mean diameters of roots per season with mean diameter of roots (mm) on the y-axis and season represented by different colored bars on the x-axis. Minimum and maximum are labeled under bars, with the darker colors representing the maximum diameters and the light colors representing the minimum diameters.
Figure 3: Bar graph showing mean number of root nodules per season with mean number of roots nodules on the y-axis and season represented by different colored bars on the x-axis.
Assessing Standard Assays for Cytotoxicity of Dental Materials

Emmie Knobloch/2017

The cytotoxic effects of all medical devices and materials are a concern, but especially those in dental materials. Fillings and adhesives may remain in the mouth for decades, giving them ample opportunity for any possible cytotoxicity to take effect. Over time, residual uncured resin composite from fillings can leach monomers such as triethylene glycol dimethacrylate (TEGDMA), urethane dimethacrylate (UDMA), and 2-hydroxyethyl methacrylate (HEMA) into the body. Understanding the cytotoxic effects these monomers have on our cells is important for assessing the safety of both novel and existing dental materials.

Current standards for assessing the cytotoxicity of dental materials have been published by several organizations, including the American Dental Association (ADA), the International Organization for Standardization (ISO), and the American Society for Testing Materials (ASTM). While these standards do provide a reference point for the toxicity of dental materials, they do not accurately represent the environment in which these materials are used. Most suggest the use of animal cell lines, and they also expose these cells directly to solid materials that will realistically only ever come in contact with the tooth.

This project aims to provide an alternative to these standards by comparing the response of traditionally tested cell lines like mouse fibroblasts with those of more appropriate human gingival cells. Three cell lines (human gingival keratinocytes, human gingival fibroblasts, and mouse subcutaneous fibroblasts) were exposed to monomers that are known to leach from dental materials (HEMA, TEGDMA, UDMA, and TiO₂) in varying concentrations for either 24 or 72 hours. Their cytotoxicity was then measured using metabolic assays and fluorescent live/dead staining.

Results indicate that the minimum concentrations at which cytotoxic effects are observed from these monomers are significantly above an amount that could realistically be released, and so are not biologically relevant. We also observed an increase in cytotoxic effects over time, and a significant difference in response between cell lines, again highlighting the need for more biologically relevant standards (Figure 1). These results will eventually be coupled with the development of a microfluidic device to more accurately simulate gingival tissue and the environment of the oral cavity in which these materials are used, to some degree bridging the gap between traditional in vitro and in vivo testing.

(Supported by the National Institute of Standards and Technology)

Advisors: Diane Bienek, NIST and Nathan Derr, Biological Sciences

Figure 1: Live/dead staining shows that monomers exhibit greater cytotoxic effects over time.
Rapid Effects of G1 Injection on Sociosexual Behavioral and Preoptic Area Isotocin Expression in the Male Goldfish

Zoe Kohler-Boland/2017

Estrogen and isotocin (IT), a sex steroid and a neuropeptide, modulate sociosexual behavior in goldfish (*Carassius auratus*), possibly through activation of populations of preoptic area (POA) neurons. Previous results from the Mangiamele lab indicate that IT-producing POA cells express Gper1 (GPR30), a rapidly responding membrane-bound estrogen receptor. Thus, we hypothesize that the activation of Gper1, leading to an increase in IT production and/or release, is the primary mechanism underlying estrogenic behavioral effects. We tested this hypothesis by determining whether the application of G1, a Gper1 agonist, mimics the effects of estrogen administration and increases social approach behaviors, and whether POA neurons activated by G1 produce IT.

Sexually mature male goldfish injected either with G1 or a vehicle were given the choice to approach an empty stimulus tank or one containing a female fish. The duration of the proximity of male fish to each stimulus tank was recorded both before and after drug treatment during three ten-minute data collection periods. We used T-tests to test for differences in the duration of proximity to each stimulus tank between treatments and data collection periods. Contrary to the initial hypothesis, preliminary results suggest that G1-injected males less strongly prefer the occupied stimulus tank.

Immunocytochemistry (ICC) will be used to examine the expression of c-Fos and IT within the POA in response to G1. The production of c-Fos, an immediate early gene, is used to measure the activity of neurons. An anti-c-Fos antibody will be used to track POA neuron activation in response to G1 treatment. An anti-oxytocin antibody will be used to pinpoint IT expression. We predict that the number of neurons stained with both antibodies at predefined locations will differ between treatments (see Fig 1).

Results contrary to the initial hypothesis may be explained by the diversity of POA neurosecretory cells. Activation of morphologically and functionally distinct populations of POA neurons may show different patterns of co-labeling. For example, G1 may activate only parvocellular POA cells, which are associated with decreased sociality in several fish species. Investigations into correlations between the number of active IT-expressing neurons within different POA populations and sociality will be undertaken during the Fall 2016 term.

Figure 1. A green anti-c-Fos and a red anti-IT primary and secondary antibody pair were applied to immersion-fixed POA tissue. C-Fos is expressed primarily within the nucleus; IT, meanwhile, appears principally within the cytoplasm.

(Supported by the Howard Hughes Medical Institute)

Advisor: Lisa Mangiamele, Biological Sciences

References:


The Role of Reelin Signaling in Neurogenic Development of the Spinal Cord

Wiktoria Leks/2019

Reelin is a large secreted glycoprotein, crucial for directing cell migration in the developing brain through the establishment of cortical cellular layers and synaptic activity. Reelin performs intracellular signaling with neuron and glia cells by binding to two lipoprotein receptors, either Apolipoprotein-E receptor 2 (ApoER2), or Very low density lipoprotein receptor (Vldlr), which as a result activates the intracellular adaptor Disabled 1 (Dab1) by tyrosine phosphorylation. Activated Dab1 receives cues from the environment to regulate neurite growth cone extensions, cortical neuronal migration, cell fate specification and radial glial morphology.

The majority of research about Reelin focuses on cell migration with still images of the brain. However, little investigation has been done on the role of Reelin in conjunction with Reelin’s receptors, ApoER2 and Vldlr, which may affect cell differentiation of various cell types or radial glia and neuronal proliferation. The zebrafish spinal cord is a simple model to study neural development due to a limited number of neuron and glia cell type. We are currently using CRISPR/CAS9 technology to generate mutations in the genomic loci of zebrafish for Reelin, ApoER2, Vldlr and Dab1 to create a total loss of function approach. The CRISPR/CAS9 system induces double stranded breaks in an assigned location of the DNA, leading to an ideally unsuccessful repair process known as non homologous end joining. However, it takes time for CRISPR injected embryos to mature, create progenitor lines, and future generations which would then carry the homozygous recessive induced mutation.

While the CRISPR lines of zebrafish mature, a splice blocking Morpholino (MO) approach was used to characterize and predict potential phenotypes for a partial knockdown of ApoER2, Dab1, and Vldlr. The results for the ApoER2 MO (Fig A) shows an increase in Oligodendrocytes Progenitor Cells (OPCs) compared to wild type control. The increase in OPCs may suggest that ApoER2 is required as a suppressor of OPCs by promoting cell migration or cell differentiation out of the neural tube. The ApoER2 mutants also presented a physical phenotype of a shorter tail and microcephaly (Fig B). These preliminary findings will have to be affirmed by the CRISPR mutants and their phenotypes in future experiments.

Figure 1A: The ApoER2 designed splice blocking Morpholino targeting exon 12 and a PCR confirming a partial knockdown due to morphant bands appearing higher than the control.

Figure 1B: An observed glutamatergic neuron reduction and microcephaly phenotype in ApoER2 morphants compared to control.

(Supported by the B. Elizabeth Horner Fund in the Biological Sciences)

Advisor: Michael Barresi, Biological Sciences

References:
Incorporating Antibiotic Resistance in the Modeling of Tuberculosis in the US

Ellie Mainou / 2017

About one third of the world’s population is infected by Tuberculosis (TB), which is caused by the bacterium *Mycobacterium tuberculosis* (1). Latently infected individuals are neither symptomatic nor contagious. Individuals with active TB show symptoms, and may transmit the disease through the air. The greatest challenge concerning the disease is the emergence and spread of strains resistant to common antibiotics (e.g. isoniazid and rifampin), constituting treatment difficult and costly. So far, mathematical models (e.g. Hill *et. al*) assume drug-susceptible TB only (2). For this reason, I develop a predictive mathematical model of TB transmission in the US population that incorporates single- and multi-drug resistance.

We developed a compartmental model that includes the different groups of the population (susceptible, latently infected, actively infected, dead of TB), as well as all the possible ways in which an individual can move from one group to another. The model contains drug-susceptible, isoniazid-resistant, rifampin-resistant and multi-drug resistant strains. The change in the number of individuals in each compartment was described by a differential equation. The model was fitted to data obtained by the Center for Disease Control (CDC) for the years 2000-2012(1, 3).

Continuing prior work, I focused on understanding the behavior of the algorithm used and of the model. Firstly, I confirmed that the model is sensitive to the order according to which the parameter space is explored, changing the quality of the fits even by an order of magnitude. In addition, a distribution of parameter values was created, which shows for most parameters that values tend to cluster at the edges of the values range. Setting such a range based on epidemiological knowledge is not the cause of this behavior. Lastly, in an effort to improve results, by fitting the model to more data, namely for the years 1993-2013 (4). Surprisingly, it was found that fits were not improved. To examine how the error changes in correlation to the amount of data employed, I used the bootstrap method. For a total of 11 trials, I picked 2, 4, 5, 6, 8, 10, 12, 14, 15, 16, and 18 years from 1993-2013, 10 times for each case. The figure shows the results of the runs. As the number of years used a data increases, the error value increases as well, probably due to the addition of constraints to the model. However, that tendency seems to asymptote.

The major improvement of this model would be its simplification by specifying the values of certain parameters using established knowledge. The final goal of this project is to use this model as a means of identifying potential interventions to eradicate TB in the US.

*(Supported by the Howard Hughes Medical Institute)*

Advisor: Robert Dorit, Biological Sciences

References:


Utilizing Cre Recombinase to Improve Multi-fluorescence Techniques in S. cerevisiae

Brigit McDannell/2018

Dynein, a multi-functional motor protein essential in the transport of intracellular cargo on the microtubule cytoskeleton, helps regulate integral biophysical processes during cell division. In this work, we want to use Cre/lox recombination to gain information regarding motor protein life cycle to better elucidate dynein’s role and to track its movement, we used Cre recombinase to rearrange DNA sequences of dynein’s gene within site-specific sites (i.e., lox sites) to create a unique fusion protein (Fig.1).2 Within the DNA cassette encoding this fusion protein are lox recognition sites coupled with fluorescent protein genes such as GFP that tag specific dynein molecules. We hypothesize that recasting the fluorescent markers and altering genetic sequences at the protein level is a viable method for observing dynein behavior.

The transformation process starts by incorporating the exogenous Gal-Cre-Term gene into the yeast genome in plasmid form.2,4 Before the cassette carrying the fluorescent gene can be inserted, multiple preliminary protocol steps were required to ensure that the yeast cells express functional Gal-Cre-Term products. Using the program SeqBuilder, we constructed DNA and primer sequences synthesized by the bioengineering company IDT. Q5, KOD, and Taq PCR methods were then used to amplify genetic material. Gel electrophoresis imaging confirmed amplification by comparing band density. Both genomic and plasmid integration were used to produce varying transgenic strains of S. cerevisiae. In future work we will leverage Gibbon’s Assembly to assemble the full lox cassette with the Gal-Cre-Term genes in the correct order and region. Western Blot analysis and confocal microscopy will help visualize the multi-colored proteins that we have used to target dynein through the motor protein life cycle.

The results from the summer were fairly substantial. The gene mRuby3 was chosen as the fluorescent protein and the cassette was successfully synthesized via IDT. The strains with URA and Gal-Cre, genes were successfully transformed in lab, resulting in two individualized yeast lines. However additional strains such as the mRuby lox cassette must be fully integrated into the genome to ultimately tag dynein.

Once all strains have the lox cassette inserted, microscopic analysis will be used to visualize intracellular dynamics and color change across multiple conditions. Over this summer I learned complex and novel laboratory protocols and significantly improved upon my genetics and cellular biology background.

(Figure 1: Synthetic fluorescent cassette of mRuby3 and GFP for tagging dynein. Boxes with black dots represent stop codons.)

(Supported by the Blakeslee Fund in the Biological Sciences)

Advisor: Nathan Derr, Biological Sciences

References:

Fig: The figure shows the bootstrap results and depicts how the error value is associated with the size of the sample.
Visualization of Fimbriae and Flagella in *Escherichia Coli*

Cadence Miskimin/2017

CFT073 is a gram negative bacterium that is responsible for approximately 80% of hospital acquired urinary tract infections. This bacterium is a uropathogenic strain of *Escherichia coli* and has a high rate of recurrent infections, making antibiotic resistance a growing concern.1

A series of protocols were developed to visualize CFT073 with transmission electron microscopy (TEM). This pathogen is a BSL-2 biohazardous organism, and fixation of cells prior to staining or transporting to the microscope facility was addressed as a crucial component of protocol design. These fixation and negative staining protocols were developed for an ongoing visualization study of *E. coli* CFT073 based on previous gene expression research, which suggested that CFT073 is capable of temperature-induced changes in virulence factor gene expression, particularly of certain fimbriae required for surface adhesion during infection.2 Fixation and staining methods were tested and modified through repeated experiments followed by TEM visualization of cells. These methods were developed according to previous research3 followed by extensive trial and error. By varying the concentrations of glutaraldehyde, cacodylate buffer and water in the fixation mixture, a successful method was developed that fixed pathogenic cells and rendered them safe for contained transport. Concentrations of phosphotungstic acid in cacodylate buffer were also modified to achieve optimal staining of cell surface fimbriae.

The result of this protocol development was a final fixation and staining protocol that allowed visualization via TEM of fimbriae and flagella in *E. coli* K-12 and in uropathogenic strain CFT073. This protocol will be used in further visualization research for the White-Ziegler lab, specifically relating to the growth and virulence factor expression of CFT073 in 23˚ C and 37˚ C conditions.

(Supported by the Howard Hughes Medical Institute)

Advisor: Christine White-Ziegler, Biological Sciences

References:


Pictured: CWZ strain 246 (K-12 commensal strain) grown in 23° C in M9 minimal media and fixed with glutaraldehyde 2.5% in 1M cacodylate buffer. Stained with PTA 0.25% in water.
A Nanoscopic View of How Applied Forces and Cargo Rigidity Impact the Motility of Dynein Ensembles

Jessica Morgan/2017

Cytoplasmic dynein, hereafter referred to as dynein, is a homodimeric mechanoenzyme that moves towards the minus-end of microtubules. In most eukaryotes, dynein use retrograde transport cargo throughout the cell. Over the last few decades different groups have investigated the motile properties of both single dynein and teams of dynein. Previous work by Derr and colleagues have used DNA origami to create a synthetic molecular cargo that could be used to study the emergent behavior of microtubule-associated motor ensembles. Through this cargo structure, motors are able to communicate their motion and states of binding to microtubules to other motors in the ensemble. Few studies have looked at the impacts of external forces on the motion of motor protein assemblies using optical traps. To study the motile properties of dynein under load, when external forces are applied, the cargo structures created by Driller-Colangelo et al. and Derr et al. were employed. Additional oligo linker strands were added to the chassis structure such that the chassis could bind to a bead through a chemical bond between digoxigenin and antidigoxigenin. These beads were later trapped with optical tweezers. In our preliminary work, we investigated this new system and determined essential protocols for ensuring a single motor protein ensemble was attached to each individual bead. To do this, the bead-cargo-dynein structures were visualized using video-enhanced differential interference contrast (VE-DIC) using the methods of Guiteerez-Medina and Block.

Movies captured with VE-DIC were used to determine the desired concentration for single-motor-protein ensemble binding. Optical trap experiments investigating the impact of external forces on the motility of teams of dynein motors will be carried out in the 2016-2017 academic year as a senior thesis project.

(Supported by the Blakeslee Fund in the Biological Sciences)

Advisor: Nathan Derr, Biological Sciences

References:


Development of an Integrated Index of Significant Trees in the Smith College Arboretum

Taz Mueller/2018.

The Smith College campus exists both as a college landscape, and as a botanic garden. As such, when the fate of particular trees on campus come into question, the conflicting reasons on whether or not they can remain on campus are complex, and come from many different perspectives- for instance, the rarity and importance of the species, the way the tree contributes to the landscape, or the tree’s historical significance. My goal for this project was to develop a methodology for gathering all the information available on specific trees and to integrate the information into a single, cohesive portal that would allow someone considering a tree to understand its value from every possible point of view.

During the course of this research, I chose five pilot trees for the program, and collected as much information on them from every source possible. This included combing through College Archives, reviewing student and volunteer projects in the Botanic Garden, interviewing a Professor Emeritus who had taught biology and observed changes to the trees, and exploring the Botanic Garden plant database. I chose a GIS Story Map as the appropriate portal for the tree index, and constructed an interactive map that would display each tree, a historical photo, and a compendium of the available information about it.

Some of the information that was gathered included the measurements of the tree’s girth, height, and spread, the landscape character of the tree, the significance and rarity of its species, its proximity to above and belowground infrastructure, the places it has been historically noted (such as newspaper articles or brochures), miscellaneous special features. The planting dates, previously missing from some trees, were discerned from College Archives.

This research resulted in the creation of a public portal for new and previously inaccessible tree information, and will further the efforts of the Botanic Garden and the Campus Tree Committee to educate the Smith College community about the importance and history of each tree on campus. It will also contribute to the preservation of the trees in the face of threats such as construction and renovations to the campus buildings. I will continue this research as a Special Studies project in the fall, and the existing portal is built for new tree entries to be added during further student research.

(Supported by the Schultz Foundation)

Advisor: Gaby Immerman, Biological Sciences

Image: The pilot Story Map with the visible trees listed in a shortlist, and each marker for the tree shown on the map, along with an informational pop-up containing a summary, historic photo, and link to more information.
Testate amoebae are ecologically significant unicellular protists found in environments such as bogs and fens.\(^1\) Although testate amoebae are not uncommon and are regarded as important bioindicators, they are particularly understudied and their placement in phylogenies is often questionable.\(^2\) Accordingly, current projects seek to reveal insights into testate amoebae and ultimately contribute to a more precise phylogenetic organization. The genetics research projects I collaborated on are focused on assessing abundance and diversity of testates from Hawley Bog over time, generating transcriptome data from single testate cells, and reinforcing evidence for RNA editing in the testate amoeba genus, Hyalosphenia.

Sphagnum moss samples were collected from different sites at Hawley Bog, during three separate dates over the course of approximately two months. Samples were prepared using moss from each site and microscopes were used to morphologically identify and count testate taxa in order to determine spatial and temporal changes in testate communities. Furthermore, sixteen individual testate cells were picked from samples and prepared for whole transcriptome amplification (WTA) using a specialized kit from Clontech, followed by preliminary polymerase chain reaction (PCR), gel electrophoresis, and sequencing. Other objectives focused on continuing work on an RNA editing project that previously produced four cDNAs from testate amoeba cells and a successful PCR targeting COX1 gene.\(^3\) We attempted to clone preexisting COX1 PCR products and conducted new gradient PCRs using remaining cDNAs.

The results for testate communities in Hawley Bog revealed no clear diversity or abundance patterns. Nonetheless, the data suggests that the testate amoeba species, Hyalosphenia papilio, was usually, though not always, the most abundant taxa at sampled sites. Moreover, initial WTA attempts resulted in two sequences that potentially represent amoeba taxa. The majority of the remaining WTAs showed present bands in gels from preliminary PCRs and were sent for sequencing. Although many of the cloning attempts for the RNA editing project did not capture the desired gene, a functional COX1 positive control was generated. Subsequent attempts to replicate gradient COX1 PCRs showed promising evidence of bands in the gel electrophoresis.

Although testate amoebae continue to elude accurate phylogenetic placement, continuing this research could prove immensely useful to improving the understanding of these organisms and the tree of life as a whole. The opportunity to participate in this collaborative research has been truly enlightening and I am eager to continue with these projects in the near future.

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Gel from COX1 PCR, using prepared cloned material. Well 1 represents ladder, wells 2-4 depict COX1 positive control with 1:10, 1:50, and 1:100 dilutions. Wells 6-8 are replicates of wells 2-4, using different COX1 primers.
(Supported by National Science Foundation)

Advisor: Laura Katz, Biological Sciences

References:
Evolution in Gene Families in Ciliates

Olivia Pilling/2017

The Katz lab studies ciliates as a model to gain insight into how genome structure affects patterns of DNA sequence evolution. Previous research demonstrated that ciliates, a clade of unicellular eukaryotes marked by cilia in at least one stage of their life cycle and dimorphic nuclei, in the classes Spirotrechea, Armophorea, and Phyllopharyngea have rapid rates of protein evolution associated with highly processed somatic genomes (e.g. ‘gene-sized’ chromosomes)\(^1\). I am building on this work by investigating the protein evolution of three species of ciliates in the understudied class Heterotrichia. While Heterotrichia are not known to extensively fragment their macronuclear genomes, they do highly amplify their macronuclear genomes\(^3\); hence we hypothesize that ciliates in the class Heterotrichia have highly divergent paralogs for a variety of proteins. Our focus is on characterizing patterns of divergences among paralogs (i.e. genes that differ due to an ancient duplication event). I focus on the ciliates Blepharisma americanum, Spirostomum ambiguum, and Stentor sp., and six genes (Actin, \(\alpha\)-tubulin, \(\beta\)-tubulin, Ef1-\(\alpha\), H3 and H4).

Addressing our hypothesis on patterns of protein evolution in Heterotrichia required a combination of molecular and bioinformatic tools. I used a PCR based approach to analyze protein evolution and I am interpreting the resulting data in a molecular evolution framework. Preliminary analyses of the data suggest the majority of the paralogs are highly conserved at the amino acid level, even though they can be up to 20% divergent in DNA (Table 1). This suggests that these paralogs are old and under a high level of functional constraint. The few instances of divergent paralogs - the Ef1-\(\alpha\) gene in B. americanum, histone H4 in S.ambiguum, and histone H4 and \(\beta\)-tubulin in Stentor sp. - suggest either relaxed functional constraints or positive selection driving the evolution of these sequences.

These data provide insight into the number of paralogs per gene for each taxa as well as their rates of evolution. My next steps are to determine if the paralogs have an even copy number within the somatic macronucleus. I can do this by using a qPCR based approach to quantify the amount of DNA. The resulting data will compared to transcriptome and genome data to infer a correlation between copy number and expression level.

My SURF project also allowed me to learn more about research in my field through reading primary literature and attending a conference. I presented a poster on the preliminary findings at the Ciliate Molecular Biology Conference. Furthermore, I will be completing a senior Honors Thesis this year continuing my work on this project.

(Supported by the Schultz Foundation)

Advisor: Laura Katz, Biological Sciences

References:

Table 1. Our preliminary analyses reveal conservation of many paralogs in heterotrich ciliates, with the notable exceptions of Ef1-α in *B. americanum*, histone H4 in *S. ambiguum*, and histone H4 and (3-tubuhn in *Stentor*).

<table>
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<tr>
<th>Taxa</th>
<th>Gene</th>
<th># of Paralogs</th>
<th>%Identity (Nucleotide)</th>
<th>%Identity (Amino Acids)</th>
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Macronuclear Development of *Chilodonella uncinata*

Anna Rogers/2017

Ciliates are monophyletic clade of protists, characterized by dimorphic nuclei, the somatic macronucleus (MAC) and germline micronucleus (MIC).¹ In sexual reproduction, a MAC is generated from a germline MIC. The genomes of the two types of nuclei can be very different and much of the processing is species specific. In Chilodonella, a MAC with gene sized chromosomes that may have copy numbers up to 10,000 is developed from a MIC with a few long chromosomes.² Additionally, Chilodonella, among a few other distantly related ciliates, shows evidence of “unscrambling” in the MAC, meaning genes are fragmented and out of order in the MIC and reassembled during macronuclear development.³ I aim to characterize the chronology of development in Chilodonella uncinata and to use this information to analyze its unique relationship between DNA content and nuclear size. The volume of the developing MAC of Chilodonella uncinata is largest when the DNA content is lowest. A similar relationship between nuclear size and DNA content has also been witnessed in the distantly related ciliate Stylonychia lemnac.⁴ Phylogenetic analyses indicate independent origins of epigenetic unscrambling in these taxa.⁵ Detailed analysis of the macronuclear development of Chilodonella will elucidate similarities between development of this ciliate and other ciliates that unscramble and extensively fragment their genomes.

To better understand the chronology of nuclear development in Chilodonella, I developed protocols for live DAPI (4’,6-Diamidino-2-Phenylindole, Dihydrochloride). This nuclear stain is frequently employed on fixed cells. While developing cells can be distinguished from vegetative cells on a fixed slide, each cell is only a snapshot of its stage of development. Live cell imaging would better aide the analysis of data from fixed cells. Though DAPI has been used for live imaging on some species of ciliate, DAPI is a semi-permeant stain and thus does not enter all cells readily or has too high of toxicity for some species. A protocol for Chilodonella has not previously been developed. A range of dilutions was used and cells were checked every 15 minutes for vitality and fluorescence. A concentration of 2.5 µg/mL was determined as optimal for fluorescence while still low enough to not kill the cells. From here we will develop protocols to isolate or immobilize individual Chilodonella cells during development in order to capture images. When the chronology of development is better established, DAPI staining on fixed cells can be used to quantify the size and DNA content of the developing MAC.

(Supported by the National Science Foundation)

Advisor: Laura Katz, Biological Sciences
References:


Vernal pools are transient freshwater systems that are fed by rainfall and snowmelt in the autumn and spring and experience a period of dryness each summer. Despite the high frequency of vernal pools in temperate forest landscapes, literature on microbial communities in vernal pools is very limited and existing studies have consistently neglected protists, despite their importance in nutrient cycling. The small size and shallowness of vernal pools makes them susceptible to dynamic changes in abiotic factors, including pH and temperature.

Since microbial communities have been shown to be sensitive to abiotic changes in other habitats, we hypothesize that the microbial communities in vernal pools will alter rapidly in response to the constant fluctuation of environmental factors. Freshwater systems are reported to have distinctive seasonal communities, with photosynthetic members having a higher abundance in the spring and summer. Hence we further expect a striking difference in community profiles between sampling months, particularly with an increase in cell count of photosynthetic protists.

Our study used two distinct methodological approaches: analysis of DNA and RNA community profiles through high-throughput sequencing (HTS) and enumeration of reverse-filtered samples by light microscopy. We extracted DNA (total community) and RNA (metabolically active communities) from aquatic samples through serial filtration on the nano and pico sized communities. We also concentrated water collected from the vernal pool and enumerated morphotypes. Ultimately, we plan to incorporate fluorescent in-situ hybridization (FISH) to strengthen data from HTS sequencing, and to identify organisms with fluorescent probes.

Our preliminary data are consistent with our hypothesis, as the abundance of autotrophic protists increased during the warmer months (Figure 1). We are currently finishing HTS data generation, and analyses of these data will provide species-specific identifications based on the genetic sequence of the small ribosomal subunit. We can corroborate the frequency of HTS reads (a proxy for species) with abundance as represented by physical counts obtained through enumeration.

As previous vernal pool studies have focused on bacteria and fungi, our analysis of protist biodiversity in the vernal pool is a pioneer study in the field. Our findings in the study will be presented at Collaborations event for the Smith College community. We will continue our work on protists in the vernal pool throughout the academic year with the aim of publishing our data.

Figure 1: Comparison of averaged enumeration counts of heterotrophic and autotrophic cells from reverse-filtered samples collected 3/9/16 through 6/10/16. Data from two sampling sites were pooled and averaged for each of the months represented.
(Supported by the National Science Foundation and the Schultz Foundation.)

Advisor: Laura Katz, Biological Sciences

References


Goldfish (Carassius auratus) communicate using chemicals called pheromones that function as external signals when released into the water. One important female pheromone is PGF2a, which increases male sexual behavior towards females by binding to receptors on cell surfaces. Although the precise mechanisms of action of PGF2a are unknown, some studies hypothesize that PGF2a increases the sensitivity of male goldfish visual pathways that guide approach responses towards females. This indicates that males may rely on visual signals for courtship and not, as previously believed, on chemical cues alone. My research is aimed to determine whether a male goldfish exposed to PGF2a displayed a preference for female goldfish when presented with chemically isolated but visually available male and female stimulus fish.

Prior to PGF2a exposure, experimental male fish were injected with Ovaprim, a gonadotropin and dopamine inhibitor cocktail that ensures peak sexual maturation, facilitates reproductive behaviors, and increases motivation. Experimental males were exposed to a PGF2a plume while partitions allowed visual but not chemical communication with male and female stimulus fish. The time spent in proximity to the stimulus fish was tracked in Ethovision and analyzed using t-tests and a two-way ANOVA in Prism.

Males significantly preferred interaction with males over females, but only following PGF2a exposure [sex * treatment, F(1,32) = 4.1, p = 0.041] (Figure 1). There was no significant preference without PGF2a (p = 0.43), though there was a marginally insignificant preference for males in the PGF2a choice experiment (p = 0.052). This contrasts with my hypothesis and previous studies that have reported male preference for females in similar choice experiments. One potential explanation is that male testosterone levels exceeded a certain threshold and caused males to display aggressive behaviors towards other males rather than chasing females. Since goldfish are external fertilizers, switching between aggressive and mating behaviors through rapid hormonal control increases the likelihood of fertilization and thus may be evolutionary advantageous. Alternatively, males may require additional female pheromonal cues or other visual signals to make a definitive choice. For instance, goldfish have UV cones that they may use to distinguish between sexes by observing intensity of UV reflectance or patterns in the facial region. Future experiments will therefore analyze testosterone levels in male fish, adjust the experimental paradigm, and explore UV reflectance and patterns to further contribute to the discussion surrounding the largely unknown communication system of goldfish and other aquatic species.

Figure 1. Males prefer females in the context of PGF2a [F(1,32) = 4.1, p = 0.041] and there was marginal insignificance in preference for males in the PGF2a experiment (p = 0.052). % social time was calculated by dividing duration near stimulus fish over total social time. Black bars denote male fish and gray bars denote female fish.

(Supported by the Howard Hughes Medical Institute)

Advisor: Lisa Mangiamele, Biological Sciences
References:


Exploring the Reelin Signaling Pathway and Its Role in the Early Development of the Spinal Cord

Amelia Stapleton/2019

Autism Spectrum Disorder is a progressively prevalent condition characterized by many developmental, behavioral, cognitive, and psychological symptoms. Several genes have been associated with ASD, some of which play a role in the Reelin Signaling Pathway. This pathway plays a major role in appropriate development of the central nervous system through the binding of two lipoprotein receptors, Apolipoprotein-E Receptor 2 (ApoER2) and Very low density lipoprotein receptor (Vldlr) which then go on to activate Disabled 1 (Dab1) by tyrosine phosphorylation. This now activated Dab1 facilitates many intracellular processes including radial migration, cone extension, neuronal cell fate, as well as neuronal cell morphology and branching. With the help of previous research done in BIO159Y and projects conducted by Katrina Anderson ’18, further research was conducted to continue exploration into this pathway and its associations to ASD as well as its contributions to early CNS development.

A loss of function approach was taken using the Crispr/Cas9 system, on ApoER2 and both Dab1 paralogs. The Crispr/Cas9 system works to create a double-stranded break in the allocated gene which will then hopefully succumb to an unsuccessful repair, also known as non homologous end joining. Zebrafish were used as our model system due to their ease in reproduction and fast development rate. These ApoER2 and Dab1a/b Crispr-injected progeny were then put on the system and grown up to further screen through a process of incrossing and outcrossing. While these progeny were being screened, an ApoER2 splice blocking morpholino was injected into embryos followed by in situ hybridization using seven different probes in order to analyze possible phenotypes that could be displayed in our knockouts. These embryos were then analyzed using cryostat techniques and other imaging methods to look for expression within the CNS, specifically the spinal cord. In addition, Crispr G1 Reelin mutants were screened through a fin clipping process to confirm already suspected heterozygous mutations.

Through a process of incrossing and outcrossing, mutants for ApoER2 and Dab1a/b were found therefore confirming the designed Crisprs. A 17 bp deletion was also confirmed in both male and female G1 Reelin mutants. There was also a smaller deletion discovered in the Reelin hets (Figure 1). The ApoER2 morpholino injections also reaped extensive expression data for all of the probes suggesting that ApoER2 plays a large role in the patterning of neuronal spinal cord cells.

Future plans include designing a VLDLR Crispr as well as continuing the incross and outcrossing process of G0 ApoER2 and Dab1 mutants. The Reelin heterozygous mutants will also be incrossed with the hopes of producing a homozygous mutant in a quarter of the population. There are also plans to use transgenic reporter lines in order to look at specific cell development, such as radial glial cells.

Figure 1: Reelin hets: 17 bp deletion and smaller bp deletion found.

(Supported by the Schultz Foundation)

Advisor: Michael Barresi, Biological Sciences.

References:

Transcriptional Studies of the Tpx-2 Gene in *Brugia malayi*: Optimizing Nuclear Protein Extraction Needed for Promoter-Binding Protein Identification

Yael Tsitohay/2017

Lymphatic Filariasis is a mosquito-borne parasitic infection caused by three species of filarial parasites: *Wuchereria bancrofti*, *Brugia malayi*, and *Brugia timori*.\(^1\) My research project focuses on the molecular study of the thioredoxin peroxidase-2 (tpx-2) gene in *B. malayi*, a gene known to be up-regulated when the parasite enters the human host from the mosquito vector during the L3 infective larval stage. Thioredoxin peroxidase proteins in general are used by the parasite to protect itself against the host’s immune response by detoxification of oxygen radicals. Previous honors students in the Williams lab (Iju Shakya ‘13 and Krithika Venkataraman ‘15) have conducted their honors theses on the tpx-2 gene and its mechanisms of up-regulation. The tpx-2 promoter region has been successfully amplified and cloned (Shakya’s honors thesis), and identification of tpx-2 proteins for transcriptional factor studies has been previously explored (Venkataraman’s honors thesis). However, due to the tough nature of filarial worm cuticles, nuclear proteins have proven to be of low yields in past experiments. Hence, the nuclear proteins extraction technique must be refined in order to optimize the yield of nuclear protein necessary for transcriptional studies of the gene.

This summer, as a continuation of my special study from the 2015-2016 academic year, I worked on optimizing nuclear protein extraction in L3 stage *B. malayi*. The worms were stained with DAPI and lysed using the Qiagen Tissue Lyser II, and fluorescence microscopy was used to check for the presence of intact nuclei before proceeding to total nuclear lysis. Nuclear protein extraction was done using the Active Motif Nuclear Extract Kit®. A total of five trials were implemented, and the lysis time was varied throughout the trials. Microscopic image results showed that with increased duration of lysis, more free nuclei were observed. Nuclear protein fractions were collected from each trial and stored at -80°C.

Continuation of this work will be done during the fall 2016 semester as special studies. Future work will consist of further verification and quantification of enriched nuclear protein via 2-D gel electrophoresis and Bradford assay, respectively. Then, the identification of transcriptional factors via binding studies will be done in order to further elucidate the studies of tpx-2 gene transcriptional studies.

*(Supported by the Blakeslee Fund in the Biological Sciences)*

Advisor: Steven Williams, Biological Sciences

References:

Wnt, The spinal frontier: The role of Wnt5b in Radial Glial Proliferation and Differentiation During Zebrafish Spinal Cord Development

Carla Vélez/Grad, Julia Kim/2019, Dana Wood/2019, Sophie Chase/2018

During embryonic development, the central nervous system is built through the proper regulation of neural stem cell proliferation and differentiation. In vertebrate organisms, radial glial cells serve as the resident neural stem cell, giving rise to several neuronal cell types including interneurons, motor neurons and oligodendrocyte precursor cells. Here, we are investigating the role of the non-canonical Wnt5b signaling protein in radial glial development. We hypothesize that Wnt5b cross talks with the canonical Wnt/β-catenin pathway and specifically functions to negatively repress β-catenin signaling. By taking advantage of both transgenic and mutant zebrafish and the use of drugs, we show that loss and gain of wnt5b function results in the increase and decrease of radial glial cell numbers. These data suggest that Wnt5b-mediated attenuation of Wnt/β-catenin signaling serves to reduce the amount of radial glial proliferation during spinal cord development. Furthermore, we are employing mathematical modeling to guide our predictions of Wnt5b as a secreted morphogen that patterns neural stem cell proliferation and differentiation. We intend to use our results and this Wnt5b model to better understand the role of neural stem cell regulation in spinal cord development and disease.

Figure 1. The effects of Wnt5b on Neural Stem Cell proliferation and differentiation. Loss of Wnt5b leads to an increase of Gfap+ radial glia and a decrease of Huc/d+ differentiated neurons. Upon treatment with β-catenin antagonistic and agonistic drugs, results are similar to those observed in previous analyses. These data lead to the suggestion that wnt5b functions as a negative regulator of Wnt/β-catenin signaling.

(Supported by the Howard Hughes Medical Institute, Schultz Foundation, National Institutes of Health and the Blakeslee fund in the Biological Sciences)

Advisor: Michael Barresi, Biological Sciences

References:
Species-Specific Assay for Seal Heartworm (*Acanthocheilonema spirocauda*)

Josselyn Vergara/2017

Heartworm and lungworm parasitic infections in phocid seals are a significant cause of parasitic anorexia, fatigue, heart and lung problems, and potential death. Nematodes obtained from marine mammal stranding agency and aquarium necropsies were identified using genetic barcodes. This method relied on small, single bar-coding genes amplified by PCR. The most widely used were the nuclear internal transcribed spacer 2 (ITS2), the mitochondrial gene cytochrome oxidase subunit 1 (COI), and the small subunit ribosomal RNA (SSU) as a third “tie-breaker” gene. However, genetic barcode data is insufficient to identify nematodes and a greater resolution is required for confirming species identification.

In order to study seal heartworm infections in novel hosts like porpoises and grey seal (*Halichoerus grypus*); a species specific assay that relies on a quantitative real time PCR to target non-coding genetic elements was optimized. Two versions of the assay were used: TaqMan and SYBR. The first one being more sensitive than the second as it uses a fluorescently labeled probe. By using an optimized version of SYBR it was possible to have the first diagnostic assay for seal heartworm. The use of this assay reveals that heartworms from harbor porpoise (*Phocoena phocoena*) that were identified as seal heartworm in the barcodes are not seal heartworms. In addition, a positive result for grey seal was obtained when this case has never been reported before. Nematodes may have recently spread to grey seal as a result of the overall rise in seal heartworm infections and global warming.

The next steps include the study of the seal louse (*Echinophthirius horridus*) as a vector of seal heartworm infection as louse is positive for seal heartworm, as shown by our diagnostic assay. This means that lice can be used as a form of xenomonitoring allowing infected seals to be treated before their death. Additionally, it would be ideal to test blood samples from infected seals to treat live, sick animals.

(Supported by the Howard Hughes Medical Institute)

Advisor: Steven Williams, Biological Sciences

References:
4 Marcogliese, D J. The impact of climate change on the parasites and infectious diseases of aquatic animals. St Lawrence Center, 2008: p 44.
Species-specific Diagnostic Assay Confirms the First Case of Seal Heartworm (*Acanthocheilonema spirocauda*) in a Grey Seal (*Halichoerus grypus*)

Kalani Williams/2018 and Caroline Keroack/2016 G

Seal heartworm (*Acanthocheilonema spirocauda*) is a filarial nematode without a definitive host species; it infects most species of phocid seals in the Northern hemisphere.1 Symptoms include anorexia, fatigue, dehydration, and coughing or bronchospasms.1 Morphological identification of nematodes found in necropsied animals has historically been used to understand the parasitic burden in marine mammals. Unfortunately morphological identification is often not possible due either to the condition of the parasite or to the lack of specialists with sufficient expertise to make the identification. When we obtain new nematode samples, our standard procedure is to tentatively identify the species by sequencing three single barcoding genes: COX1, ITS2, and SSU. For definitive species identification, single gene barcodes are not sufficiently specific.3 Preliminary barcoding data obtained from parasite samples collected from various stranding agencies and aquariums identified nematodes from both the grey seal (*Halichoerus grypus*, a phocid seal) and the harbor porpoise (*Phocoena phocoena*, a cetacean) as seal heartworm. Both of these would be novel host species for this filarial nematode.

To confirm the identity of these infections, a species-specific diagnostic assay for seal heartworm is required. We developed such an assay using an established pipeline, which included sequencing the full genome of seal heartworm.5,6 We have now optimized this quantitative PCR assay and confirmed the identity of the nematode found in the grey seal to be seal heartworm (Figure 1). These data suggest that seal heartworm does, in fact, parasitize the grey seal. This result makes sense because the grey seal is one of the few phocid seals in the Northern hemisphere that had not yet been found with this infection.2 None of the nematodes from harbor porpoises that had been tentatively identified as seal heartworm were positive with our new qPCR assay, suggesting that the seal heartworm of phocid seals has not extended its range to include cetaceans.

In the future we plan to diagnose infections within live animals. Diagnostic tests will move away from using whole nematodes to using either blood samples or xenomonitoring. We have already had success screening the seal louse (*Echinophthirius horridus*) with our qPCR assay: lice from an infected seal are positive while those from uninfected seals are negative. The next step will be to optimize them for clinical use with blood to quantitate the parasite burden for animals in rehabilitation to aid in diagnosis and treatment.

![Amplification Plot](image1.png)

Figure 1. *Acanthocheilonema spirocauda* (seal heartworm) qPCR diagnostic assay with a positive nematode sample from a *Halichoerus grypus* (grey seal) host.

(Supported by the Blakeslee Fund in the Biological Sciences)

Advisors: Robert Dorit, Steven Williams, Biological Sciences

References:

Caused by mosquito-borne virus, dengue fever affects 50 to 528 million people annually. Dengue virus is a RNA virus that belongs to Flaviviridae family, which also includes West Nile virus, Japanese encephalitis and Zika virus. During early stage, dengue fever may share similar symptoms with infections of other Flaviviruses. However, traditional diagnostic method that relies on virus-specific antibody detection is not effective during early stage, since it usually takes 5-7 days for human immune system to produce IgM and IgG in response to an infection. PCR test is a new diagnostic method that is proved to be effective in the first seven days of infection. Yet, current PCR tests bear many defects, such as only valid for one subtype of Dengue virus or cannot distinguish between Dengue and Zika virus. Therefore, this summer, I focused on designing a quick and accurate PCR-based diagnostic tool that is applicable to all serotypes of Dengue virus.

Designing an ideal primer is the key for a PCR test. First, all sequences of Dengue virus that are available are aligned by MAFFT. Then a consensus sequence was calculated in order to design primers using IDT Primer Request Tool. The genome of Dengue virus has about 11000 nucleotide bases, which can be divide into four main parts, each coding for 5'UTR, 3 proteins that constitute the virus particle, 7 nonstructural proteins and 3'UTR. The primers that I have found share similar characteristic with primers that has been tested to be not effective enough in previous papers about PCR based diagnostic method of Dengue virus. Thus, my target has been shift from the whole sequence to a single region. Nonstructural protein gene 1 (NS1) and nonstructural protein gene 5 (NS5) are two immune-relevant genes. Since immunological traits of dengue are used in the diagnosis of Dengue, NS1 and NS5 might also be possible targets in genetic test. Therefore, consensus sequences of NS1 and NS5 were calculated. However, because NS1 and NS5 are not highly conserved, no possible primer was found.

Some recent researches have found that some genes that code for certain amino acid are well conserved. These genes might be new targets for the improvement of PCR test of dengue virus.

**Characteristics of the primers**

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(Supported by the Howard Hughes Medical Institute)

Advisor: Steven Williams, Biological Sciences
References:


Optimization of a Chan-Lam Copper-Mediated Cross Coupling of Carbon Nucleophiles and Boronic Acid

Suzanne Abreu/2017

The addition of methyl groups to bioactive compounds often results in a boost in drug potency, mainly due to the conformational changes the molecule undergoes by the addition of this group.1 Previously, Itami discovered C-H arylation using copper salts to catalyze the reaction between nucleophilic C-H bonds and arylboronic acids.2 In the Gorin lab, we came up with a reaction for C-H methylation using bench-stable reagents and reaction conditions extrapolated from the literature.2 Using N-methylindole (NMI) as our substrate, the methylated product was selectively afforded in 20% yield (fig. 1). An internal standard, dimethyl pimelate (DMP) was used to calculate yield through GC-MS data analysis. Our goal was to discover and optimize conditions for C-H methylation of bioactive arenes.

We first examined the role of air in our reaction. With the exclusion of air, the reaction still made 20% product. This suggested that oxygen was, surprisingly, not the final acceptor of electrons in our catalytic cycle, since its presence/absence had no effect on product yield. Thus, we began to screen strong oxidants, but the addition of these oxidants did not improve product yield. Different solvents such as polar protic/aprotic solvents were screened, but isopropyl acetate proved to be the best solvent for the reaction, with a maintained yield of 20%. Different methyl sources were used, but methyl boronic acid gave the best yields. We also used reagents that formed methyl boronic acid slowly throughout the reaction so that it wouldn’t be consumed quickly, but it also did not improve our reaction yields.

Moreover, different copper salts were tested, but 0.2 equivalents of copper trifluoroacetate was the least harsh. However, using 1 equivalent of the same copper quickly degraded both product and substrate. After screening different additives (i.e. acids, bases, a sacrificial alkyne), we concluded that water stabilized N-methylindole during the reaction, but it still produced 20% yield, suggesting that running the reaction longer while using water and high equivalents of copper might produce higher yields of product. Product degradation studies were conducted to monitor how much product degraded overtime. We found that the reaction yield plateaued at 20% yield and decreased over time, suggesting that 1 equivalent of copper must eventually be used to overcome a 20% yield. Currently, we have selected two new aromatic substrates to C-H methylate under the same reaction conditions. We aim to conduct similar optimization studies on those substrates as well as continue testing a few remaining promising additives on N-methylindole.

(Supported by the Schultz Foundation and the Smith College Chemistry Department Alumnae Gift Fund)

Advisor: David Gorin, Chemistry

Fig 1. Reaction scheme of the Chan-Lam cross coupling reaction of N-methylindole and methyl boronic acid.

References:


Copper-Mediated C-H Methylation of N-Heterocycles with Methylboronic Acid

Mairead Bartlett/2018

C-H methylation is a useful organic reaction, replacing a carbon-bound hydrogen with a methyl group. In pharmaceuticals, this alteration can have profound effects on a drug’s solubility and half-life, thereby affecting its potency. However, current methodologies often require hazardous reagents and/or need to be performed early in the synthesis to maintain regioselectivity in the final product. The potential rewards of improving C-H methylation led us to investigate Chan-Lam cross coupling conditions typically used for arylation as a method of methylation on biologically-relevant substrates.

Chan-Lam cross-couplings use copper catalysts to pair a nucleophile and boronic acid. We aim to improve the yields of this reaction with methylboronic acid to rival traditional methylation methods, and to make yields comparable to those of typical Chan-Lam cross couplings performed with arylboronic acids. This would be the first example of C-H methylation using a Chan-Lam.

Figure 1: Typical Reaction Conditions for the Methylation of N-Methylindole at Beginning of Summer

We investigated C-H methylation of N-methylindole using copper trifluoroacetate under air, which gave a regioselective methylated indole. Variables including the solvent, copper source, methyl source, additives, oxidants, and starting substrate were examined using GC/MS spectrometry yield quantification. One challenge encountered was that product degraded under the reaction conditions, requiring us to find conditions that promoted product formation while limiting product decomposition. Copper triflate and copper trifluoroacetate both successfully produced product, but the original copper trifluoroacetate degraded the final product more slowly. Methylboronic acid in 2.0 equivalent was demonstrated to be the best balance of product formation and degradation. Basic and acidic additives both considerably slowed the reaction, but water and boric acid showed promise as additives due to their “protective” nature of the final product. Oxidant studies demonstrated that oxygen was not acting as the oxidant as expected, but did not further elucidate a potential oxidant in our system. Alternative substrates were tested in our standard conditions, some of which appeared to yield product, but the yields did not appear higher than those we achieved with n-methylindole.

Further studies must be conducted to study the effect and mechanism of boric acid and water on both product formation and degradation. Discovering the final acceptor of electrons is of utmost importance to fully understand the reaction, which will hopefully be transferrable to other substrates in the future. I plan to return to the project next summer after which I hope to complete an honors thesis working on methylation.

(Supported by the Schultz Foundation)

Advisor: David Gorin, Chemistry

References:

Exploring Aerosol Growth as a Function of Chemical Composition

Tara Bhat/2018

The purpose of this research project was to explore aerosol growth and chemical composition through analyzing absorbance values. An investigation was done to assess the impact of 1-propanol on aerosol formation by comparing different concentrations of 1-propanol added to 0.17 M glyoxal and 3 M ammonium sulfate solutions of glyoxal and ammonium sulfate. The goal was to determine what products were formed with this interaction and compare the effects with other alcohols. Solutions in the concentration range of 0.01 M to 5 M were made and tested every hour in the Cary 100. Each timestamp was graphed at absorbance value 277 nm which is the absorbance maximum. The results of 1-propanol absorbance values over time showed similar results to 2-propanol values, which has a similar molecular structure. It was expected that the smaller the concentration of alcohol, the closer to the stock the trend would be, since the solution would have more of the stock solution in it. This was supported by the results of 5, 4, 3, 2, and 1 M. However, 0.5 M and 0.25 M results were below the stock range. Comparison to the 2-propanol results from earlier in the year showed similar trends. The results showed that the smaller amount of alcohol in the solution did not always result in trends close to the stock. It was found that overall, 1-propanol behaved similarly with 2-propanol. This research will be continued as a special studies project in the remaining two school years. Further research would try smaller concentrations of 1-propanol and 2-propanol and compare them in the same experiment. In addition, measurements over multiple days will be taken with the Cary 300 to see if the trend continues.

(Supported by the Schultz Foundation)

Advisor: Andrew Berke, Chemistry
Development of a Tandem Diels-Alder/Pauson-Khand Reaction Strategy for the Synthesis of Tetracycles

Katie Blackford/2017

The development of new strategies to promote Diels-Alder reactions will make forming important cyclic compounds, such as steroids, more efficient. With the ultimate goal of determining whether a cobalt-complexed alkyne can act as an electron-donating group and promote the Diels-Alder reaction, the Shea lab has worked towards developing a novel one-pot tandem Diels-Alder/Pauson-Khand strategy applicable to the synthesis of steroids. Since 2006, Shea Lab researchers have attempted to synthesize tetraenyne 8, the acyclic precursor that will be converted to steroid backbone 11 via the tandem reaction (Scheme 1). It was likely synthesized by Elsa Hinds (’13), but full characterization of the miniscule amount produced was impossible. Several of the reactions shown in Scheme 1 were optimized by Natalie Vaninov (’14) and Zulema Peralta (’15), but tetraenyne 8 has yet to be successfully synthesized with certainty.

Scheme 1. Proposed synthesis of tetracyclic steroid backbone 11 via a one-pot tandem Diels-Alder/Pauson-Khand reaction.

Research this summer focused on the optimization of the Suzuki cross-coupling reaction used to produce dienyne 6, as inseparable mixtures of the desired dienyne and impurities were originally produced. Several combinations of palladium catalysts and ligands were tested, and pure dienyne 6 was produced using two of these combinations, but these results were not reproducible. Further modifications to the reaction again yielded material that could not be purified. Thus, it was concluded that the Suzuki reaction was not feasible, and a new method of producing the dienyne should be found.

The first potential new method of producing the dienyne that will be explored this fall involves coupling vinyl iodide 3 and a bromoenyne using a Negishi reaction. Once a viable, reproducible method of obtaining pure dienyne has been found, progress towards tetraenyne 8 can continue. The tandem Diels-Alder/Pauson-Khand reaction will then be investigated.

(Supported by the Committee on Faculty Compensation and Development, (CFCD)

Advisor: Kevin Shea, Chemistry

References:

Chemical Patterning of Nanoscale Surfaces: Comparing the Partial and Absolute Etching of Nanoscale Hillocks on Si(100) Surfaces

Kelsey Mack/2017, Xinyuan Chen/2018, Claire Vinson/2019

The chemical and topographical functionalization of silicon surfaces is important in various fields, including the study of microorganism growth. Being able to further understand how multifunctionalization affects the surface of a silicon wafer can give us the ability to create a broad array of surfaces with control over both topographical and chemical characteristics. This work is a continuation of the work done in the Queeney Lab by Madeleine Beasley (2014) and Minhee Kim (2015) in an effort to learn more about multifunctionalized Si{100} surfaces.

Sample surfaces were prepared through an SC-1 and SC-2 cleaning process to remove impurities before being treated with an HF etchant. The samples then underwent either a 5 or 24-hour etch in argon purged water to form the desired nanotopographical features, namely hillocks, before being multifunctionalized through 1 hour oxidation and 15 minute hydrosilylation. Surface infrared spectroscopy (FTIR) was used to analyze the SiHx, SiOx, and CHx regions.

By analyzing the SiHx region after the samples had been etched, we saw that the 5-hour etched surfaces had a higher proportion of strained {100} oriented Si-H bonds, while the 24-hour etched surfaces had more prominent peaks that correlated to unstrained Si{111} and {110} species, as shown in Figure 1. This data confirms earlier research that suggests the 24-hour etched surface was more homogeneous than the 5-hour etched surface due to the uniform peaks of the fully etched surface. The partially etched surface is not going to have as consistent a surface because of the larger patches of surface that are not etched.

Difference spectra provide information about what species on the surface disappear after oxidation. By knowing what peaks appear and disappear, we can confirm whether the tops or the sides of the nanohillocks receive more oxidation. Figure 2 shows the difference spectra of both the 5-hour and 24-hour etched surfaces after oxidation, revealing which Si-H bonds disappeared through this process. The presence of downward peaks suggests the disappearance of SiH2 on Si{100}, {111} and {110} after oxidation. The peaks corresponding to Si{110} species are the most prominent, indicating that the sides of the hillocks were oxidized more readily than the tops.

Evidence for hydrosilylation is shown in Figure 1, with the presence of three prominent peaks in both spectra that correlate to three species of CHx bonds. While the 24-hour etched multifunctionalized surfaces had a higher integration in the CH region than the 5-hour etched multifunctionalized surfaces, Figure 1 suggests that the 5-hour etched surfaces were more crystalline due to denser packing of alkyl chains. Because the peaks for the 24-hour etched surfaces are red-shifted compared to the 5-hour etched surfaces, it can be said that the 5-hour etched surfaces were more crystalline and homogeneous than the 24-hour etched surfaces.

In order to definitively comment on the differences between 5 and 24-hour multifunctionalized Si(100) surfaces, the experiments described above should be repeated, with a greater emphasis on contact angle goniometry to confirm our results. Further research will be conducted throughout the upcoming semester through special studies as well as in preparation for a thesis written by Kelsey Mack.

(Supported by the Katherine C. Hauch 1921 Fund)

Advisor: Katherine Queeney, Chemistry

References:


Figure 1. On the left is an FTIR spectrum of the SiH region of a 5-hour etched surface compared to a 24-hour etched surface. On the right is an FTIR spectrum of the CH region of the 5-hour etched surface compared to the 24-hour etched surface.

Figure 2. FTIR difference spectra of the 5-hour etched oxidized surface compared to the 24-hour etched oxidized surface.
Investigation of the Base Pair Opening for Spiroiminodihydantoin (Sp) DNA Lesions

Luojun Dong/2017

Oxidative damage to DNA has been shown to be relevant to carcinogenesis, aging, and neurological disorders. Many oxidative lesions derive from guanine because it has a low reduction potential. 8-Oxoguanine (8-oxoG) is one of the guanine lesions that has been well studied. While 8-oxoG is similar in size and structure to guanine, it is highly mutagenic, prone to GC→TA transversions. 8-oxoG is also highly reactive toward further oxidation. Through one-electron oxidation, 8-oxoG can be oxidized into the spirominodihydantoin (Sp) lesion. To obtain more insight into the enzyme repair mechanism of the Sp lesion, we measured the opening rates (aka imino proton exchange rates) of a normal G-C base pair and a Sp-C base pair in 11-mer DNA duplexes by 1D NMR.

As DNA is a dynamic macromolecule, the hydrogen bonds between bases are continuously breaking (opening) and reforming (closing). Even in stable duplexes some “breathing” occurs, allowing hydrogen bonds to open and exchange with protons in the solvent, which in this case is water. My hypothesis was that at higher pH and higher base (ammonium chloride) concentration, the imino protons in all base pairs would exchange with magnetized water at a faster rate. Hence, with the enhanced exchange rates, we would be able to better observe the different opening behavior between the normal G-C base pair and the Sp-C base pair. This summer I was able to complete the first base pair opening experiment of the G-C control sequence with five base titration points. I found that the base pairs closer to the center of the duplex have slower exchange rate than those terminal base pairs. Another conclusion based on this preliminary data is that the active base concentration should not exceed 0.024 M. More base titration points are need for a more accurate base pair opening rate extrapolation.

This research experiment was very helpful because I not only gained more knowledge about DNA, but also learned many useful laboratory and analytical techniques, such as advanced level operation of the 500 MHz NMR. During the academic year, I will continue on the data collection and improvement of the base pair opening experiments.

(Supported by the Committee on Faculty Compensation and Development, (CFCD)

Advisors: Cristina Suarez, Chemistry; Elizabeth Jamieson, Biochemistry

Reference:
Figure 1. 1D NMR spectrum of the base pair opening experiment of the G-C Control sequence at pH=8 and 10°C.

Figure 2. The intensities of imino proton peaks are plotted as a function of mixing time (s) for the G-C control sequence at [0.017M] active ammonium chloride. The imino peaks are numbered from left to right based on the 1D spectrum above. The peak intensities decrease modestly as the imino protons exchange with inverted water protons and then increase as they relax back to their equilibrium value.4
Self-Folding Multi-Walled Hydrogel Tube: A Candidate for Cell Encapsulation and Controlled Drug Release

Ana Paola Garcia/2017

Hydrogels, three-dimensional, cross-linked polymer networks, have emerged as functional materials for numerous biomedical applications. The ability to manipulate the composition, shape, surface morphology, and porosity of hydrogel-based scaffolds has ultimately created attractive approaches to overcoming the various challenges of designing such materials. One such advance involves the development of tubular, multi-membrane hydrogels as promising carriers for drug delivery and tissue engineering. While existing literature describes the self-assembly of various polymer materials and designs, such approaches lack the chemical flexibility and biocompatibility necessary for advanced tissue scaffolds.

Hence, the project's aim was to develop a strategy for inducing a rolling shape transformation in a planar poly(2-vinyl-4,4'-dimethylazlactone) (PVDMA)-based hydrogel, cross-linked using Jeffamine with a molecular weight (MW) of either 600 or 2000 g mol$^{-1}$, termed Jeffamine-600 and -2000 respectively. Preliminary cell studies on these gels have demonstrated their biocompatibility. In addition, our studies on their swelling behavior in water showed that the Jeffamine-2000 cross-linked gels had an increased capacity to uptake water when compared to the Jeffamine-600 gels. Based on these observations, we hypothesized that a bi-layered, hydrogel patch consisting of two layers with different stiffness and swelling ratios would self-fold into a tubular structure when placed in water (Figure 1A).

In brief, to generate this hydrogel, 2-vinyl-4,4'-dimethylazlactone (VDMA) monomer was prepared via a two-step synthesis from 2-methylalanine, a modified amino acid. The monomer then underwent a free radical polymerization to become PVDMA. In a vial, a solution of the synthesized PVDMA (8 wt%) with the respective cross-linker (Jeffamine-600 or -2000; 0.01-0.05 equiv.) was prepared in DMSO to achieve hydrogel formation. The self-folding bi-layered hydrogel was assembled by first preparing a thin, 25% Jeffamine-600 cross-linked gel. The higher MW Jeffamine-2000 was then used to prepare a second, 10% cross-linked hydrogel layer over the first (Figure 1A). Each hydrogel thickness was kept constant at 200 μm, and no physical separation was noted between the two layers. The subsequent immersion of the bi-layered gel in water triggered self-folding into the target tubular structure (Figure 1B).

In accordance with the initial hypothesis, the driving force behind self-assembly was the differential swelling of the two layers within the hydrogel patch. As each layer swelled in the water, a stress was introduced at the interface, which was relieved through rolling up. Overall, we envision that these self-assembling hydrogel tubes, well-suited for cell or drug introduction, will be used to fabricate functional tubular tissues, such as the trachea and blood vessels.

(Supported by the Schultz Foundation)

Advisor: Maren Buck, Chemistry

References:

Figure 1. A). Schematic showing the bi-layered hydrogel, with each of the layer cross-linked using Jeffamine with either -600 or -2000 molecular weight. The distinct swelling behavior of each layer ultimately allowed the planar gel to self-roll when placed in water. B). Images of the formed, multi-walled hydrogel tubes with different spiraling patterns and diameters.
Antibiotic resistance is a rapidly growing problem. Over the past year about 2 million people were diagnosed with antibacterial resistant infections, at least 23,000 of whom died from their infections.1

However, over recent years a promising new class of antibiotics has emerged, known as lipopeptides. This group of antibiotics is much harder for bacterial cells to resist, due to its indirect method of attack. These molecules integrate themselves into bacterial cell walls, forming ion channels. This disrupts the ion concentration gradient within the cell, leading to bacterial cell death. If we can enhance this group of antibiotics we may be able to target a multitude resistant infections. My research is focused on creating a completely new molecule that would not only be a potential antibiotic but also hold other positive effects that are normally not present in cyclic lipopeptides.

The cyclic polymer design was inspired by biological structures isolated from marine life containing oxazole and thiazole groups. Both groups are five membered rings that are believed to have the ability to transport metal ions, which could greatly increase their medicinal qualities.2 An oxazole group can be easily made from serine and phenylalanine, thus I designed an eight residue cyclic peptide composed of four amino acids alternating in position. The oxazole rings are formed through coupling and oxidation of the amino acids. This synthesis was done using a solid phase strategy, a very common means of peptide synthesis. Solid-phase synthesis enables a drastic reduction to the synthesis time as compared to traditional synthesis in solution form. It is done by using small bead like objects as anchors (resin) on each of the numerous peptides formed. This summer I sought out to analyze my initial product and resynthesize.

I have analyzed my first product using a planar lipid membrane (PLM) system and shown that it is an efficient, sodium-selective ion channel (Figure 1). However, we have not been able to determine the exact structure of the polymer by using typical means of analysis. This past summer, I was able to resynthesis a molecule of similar functionality and structure. We will continue with further analysis this coming semester to identify the exact structure of our first and second product.

![Figure 1. PLM data from the sodium-selective ion channel](image)

(Supported by the Howard Hughes Medical Institute)

Advisor: David Bickar, Chemistry

References:
Alpha Group Influence on Intrinsic Barrier and Reaction Rate Estimation Using Marcus Theory

Nancy (Xingyi) Guan/2019

The goal of this experiment is to investigate how the rate of the process in Figure 1 is influenced by the change of alpha group and how well the rate of substitution reaction can be estimated using Marcus Theory. Using Gaussian 09, energies of each molecule in its separate, ion pair and transition state were obtained, and the reaction activation barriers were then calculated.

In this research, the intrinsic barrier of an interested molecule was found to be significantly influenced by its alpha group. We argue that this is due to the interaction between the $P_x$ orbitals of X-C-X system and the $P_x$ orbital or pi bond on the alpha groups. This argument is supported by the fact that as the oxygen $P_x$ orbital of OCH$_3$ in alpha position turns from parallel to perpendicular position, the activation energy lowers by 10 kcal/mole. The same phenomenon was also observed in the alkene alpha group case. As shown in Figure 2, when the attacking/leaving group is fixed, most activation energies follow the pattern that the more electron donating is the alpha group, the lower the intrinsic barrier. However, certain anomalies were observed. For instance, the activation energies of some fluorine points were higher than expected. We observed that the $P_x$ orbital on the two fluorine are tilted, and we doubt that this is due to the shortness of C-F bond, but more work needs to be done to see why the orbital interaction is distorted.

Marcus Theory suggests that the activation energy of substitution reaction can be estimated using the two intrinsic barrier and the heat of reaction via the following equation.

$$\Delta E_{X,Y} = \frac{1}{2}[\Delta E_{X,X} + \Delta E_{Y,Y}] + \frac{1}{2}\Delta E^0 + \frac{(\Delta E)^2}{8[\Delta E^*_{X,X} + \Delta E^*_{Y,Y}]} \quad (3)$$

In the system I worked on, it was found that the equation works quite well to estimate the activation energy from ion pair to transition state, but not so well for that from separate starting molecules to transition state.

(Supported by the Committee on Faculty Compensation and Development, (CFCD))

Advisor: Robert Linck, Chemistry
Figure 1. The interested self reaction. The interested molecule is of CH₂AX form, where A represents the alpha group and X stands for the attacking/leaving group in the substitution reaction.

Figure 2. The activation energy of self reaction with 4 different attacking/leaving groups. The alpha group from left to right are planar NH₂, carbonyl, OCH₃, amide, alkene, H, betamethoxide and twisted methoxide.
Lymphatic filariasis (LF) is a neglected tropical parasitic disease that affects 120 million people worldwide. Current treatments for LF can only eliminate parasites in the microfilarial stage of their life cycle, but the unaffected adult nematodes can cause severe side effects in the host, including permanent disabilities. For this reason, it is imperative that more effective treatments be developed.

A class of compounds called neurolenins, extracted from the plant Neurolaena lobata, have already demonstrated promising anti-filarial activity against both microfilarial and adult nematodes. The goal of this project is to synthesize analogs of the naturally occurring neurolenins and administer them to Brugia malayi nematodes, one of three species that cause LF, in order to find compounds that could be viable candidates to treat LF.

Work this summer expanded on some of the ways in which neurolenin D has already been functionalized. A series of reactions revealed that epimerization at the secondary alcohol stereocenter can be achieved in either acidic or basic conditions, although the latter resulted in significantly better yields. Since neurolenin D is biologically inactive, the epimerized neurolenin D was acetylated to form an epimer of the biologically active neurolenin B.

Previously, neurolenin D was esterified at the secondary alcohol using isovaleric acid. This esterified compound showed promising bioactivity but was partially insoluble in the solution required for testing. Three new esterification reactions were run this summer in hopes of producing an esterified compound with better solubility. All three anhydrides tested esterified neurolenin D at the secondary alcohol, and in the case of propionic anhydride, the tertiary alcohol was esterified as well.

The five new compounds produced this summer will be tested by the Williams lab against B. malayi nematodes. The results from these tests will guide future reactions, which may include esterifying the epimerized neurolenin D. So far, reactions have targeted primarily the secondary alcohol, but there are still many functional groups on neurolenin D that have yet to be altered.

(Supported by the Howard Hughes Medical Institute)

Advisor: Kevin Shea, Chemistry

References:

Propofol (2,6-diisopropylphenol) is a commonly used general anesthetic that binds GABA<sub>\alpha</sub> receptors. Though effective in causing rapid onset and offset of action, there are adverse side-effects associated with propofol that have spawned efforts to discover novel anesthetics with fewer drawbacks. Past studies by Adam Hall analyzed the activity of cyclohexanol analogs as anesthetics and suggested that cyclohexanols with short aliphatic chains act as potential anesthetics. The Hall lab is now interested in testing single stereoisomers of these cyclohexanol analogues in efforts to study the stereoselectivity of these GABA<sub>\alpha</sub> receptors as well as to search for compounds with improved potency.

The pharmacodynamics of propofol as an anesthetic has been studied by analyzing the activity of a diverse range of propofol analogues with short aliphatic chains. Hall's lab previously demonstrated anesthetic activity of mixtures of 2,6-dialkylcyclohexanol isomers. The goal of the current project is to study single isomers, specifically by synthesizing and purifying the isomers of 2,6-dibenzylocyclohexanol, a cyclohexanol analogue with bulkier groups in the ortho positions than previously tested.

Last year, Naina Zaiman '16 and Julia Yun '16 were able to synthesize 2,6-dibenzylocyclohexanol but failed to isolate pure cis,cis- and trans,trans- cyclohexanol isomers. The reaction scheme presented in Figure 1 illustrates the successful three-step synthesis of 2,6-dibenzylocyclohexanol, initiated by a double aldol condensation reaction and followed by hydrogenation of the conjugated enone. The final reduction step, as previously attempted on the cis cyclohexanone, was also successfully completed on the trans ketone to yield four total isomers of the cyclohexanol. Future work for this project includes investigating the synthesis of the 2,6-diisopropyl derivatives and preparation of unsymmetrical analogs.

(Supported by the Schultz Foundation)

Advisor: Kevin Shea, Chemistry

References


Manganese pentacarbonyl compounds, Mn(CO)\textsubscript{5}X, are important in understanding insertion reactions. They are observed to distort from octahedral geometry depending on the nature of the ligand X. Our calculations show that in compounds of appropriate symmetry, there are two equatorial X-Mn-CO angles. In addition, in compounds with a big distortion the yz-plane carbonyls actually bend down from the X ligand. To explain this phenomenon of bifurcation we evaluated the change in energy of highest occupied orbitals with predominantly d character in a simple compound, Mn(CO)\textsubscript{5}NH\textsubscript{2}, as the XMn-CO\textsubscript{yz} (\(\alpha\)) angle is varied (Figure 1). The XMnCO\textsubscript{yz} angle was kept constant at 70°. Much to our surprise, the Hückel calculations showed that the Mn d\textsubscript{yz} orbital is stabilized as the \(\alpha\) angle increases (Figure 2). The change in energy of the d\textsubscript{yz} orbital that occurs even when the XMnCO\textsubscript{yz} angle is kept constant suggests that orbital interactions, other than simple MnCO\textsubscript{yz} \(\pi^*\) type donation, may be present. We propose an interaction between Mn d\textsubscript{xz} orbital and CO\textsubscript{yz} \(\pi^*\), which we refer to as “pseudo-delta bond”, that may be responsible for the stabilization of d\textsubscript{yz} orbital (Figure 3). As seen from Figure 3B, higher \(\alpha\) angles lead to constructive overlap between the \(\pi^*\) of yz-plane carbonyls and manganese d\textsubscript{yz} orbitals. The “pseudo-delta” interaction is also evident when analyzing the composition of the Mn orbital with predominantly d\textsubscript{yz} character. Our calculations show that at higher \(\alpha\) angles (i.e bifurcated compounds) the yz-plane carbonyls exhibit bonding with d\textsubscript{yz} orbital. In nonbifurcated compounds, for example with \(\alpha = 80°\), this bonding is almost nonexistent. We hope to continue this research during the upcoming academic year and find other evidence of this interaction.

**(Supported by the Schultz Foundation)**

Advisor: Robert Linck, Chemistry
The Research conducted this summer investigated how aerosol mimicking solutions interact with atmospherically relevant alcohols. Aerosols are an atmospheric cooling agent, yet little is known about secondary organic aerosols (SOAs), which are produced from both anthropogenic and biogenic sources. By researching the composition and behavior of aerosols, the accuracy of predictive global warming models can be enhanced; along with potential pollutant hazards.

Over the course of the summer, an aerosol mimicking solution (ammonium sulfate, ultra-purified water, glyoxal) was combined with various concentrations of atmospherically relevant alcohols. These solutions were then analyzed using UV-Vis absorbance spectrometer. The intensity that the solution absorbs light the peak (277 nm) which is associated with the formation of high weight molecular oligomers which indicate whether an alcohol additive effects oligomerization. Data obtained for tert-butanol, 1-propanol, and methanol was then fit and extrapolated in order to analyze whether alcohol structure or concentration affects oligomerization.

Although no clear trend was acquired, both tert-butanol and methanol increased absorbance with certain concentrations of alcohol while other concentrations had either less of an effect, or none at all (see graphs below). One potential oligomerization mechanism is micelle-like, meaning that, like micelles the structure and polarity of the alcohol would impact the arrangement of the product. Future research to further test this theory could include DOSY NMR, extended four day absorbance spectra using the Cary 300 UV-Vis, and the inclusion of other atmospherically relevant alcohols.

(Supported by the Howard Hughes Medical Institute)

Advisor: Andrew Berke, Chemistry
Symptoms of Parkinson’s disease can only be observed when approximately 70% of the dopaminergic neurons in substantia nigra are lost. Diagnosis of the disease prior to huge loss of dopaminergic neurons may help slow the progress of the disease and provide better patient care. Dopamine concentration in peripheral nervous system decreases in early stages of Parkinson’s disease. Concentration of salivary DJ-1 protein in Parkinson’s disease is much lower than the “normal” population. Since saliva is easily accessible, a salivary dopamine assay would make an effective diagnostic tool. This research aims to develop a consistent and reproducible dopamine assay.

3,4-dihydroxybenzylamine (DHBA) and dopamine have almost identical molecular structure. Therefore, DHBA and dopamine have similar retention time in high performance liquid chromatography (HPLC). DHBA is used to find the unknown concentration of dopamine in a saliva sample. Artificial saliva (ECD buffer and dopamine) has been purified using WCX column and analyzed using HPLC. The dopamine signal detected is converted into dopamine concentration.

The ratio of DHBA and DA are very similar and consistent in all the test runs. More research will be conducted in the future with saliva of Parkinson’s patients on a drug holiday.

References:


Cobalt(III) pentaaamine, (Co(NH₃)₅)³⁺, can bind to a nitrite group (NO₂⁻) in the axial position in two ways. One is the oxygen bonded complex (nitrito), and the other is the nitrogen bonded complex (nitro). Experiments have shown that the nitrito complex undergoes an intermolecular process forming the more stable nitro complex, where O-Co bond breaks and N-Co bond forms. A paper by Jackson et al. also discovered a faster process of O to O isomerization, where the oxygens on the nitrito exchange with each other¹.

Previous studies used Gaussian 09 to find the structures and energies of the nitrito and nitro complexes of cobalt, and the transition states of O to N and O to O isomerizations using cc-pVTZ basis set. In this study, the nitrite group of the nitrito and nitro complexes were rotated along the z-axis to find the most stable states of the two complexes, and the activation energy of the O to N and O to O transitions was then calculated. The results showed that the activation energy of the O to O transition was about half of the O to N transition, meaning that the O to O transition is much faster. The axial ammonia group was replaced with H₂O, CO, PH₃, F, Cl, NH₂, H, and CH₃⁻ respectively to see how the axial ligands affect the activation energy. The result showed that the complex with more negative axial group had lower activation energy in both the O to O and the N to O transitions. The hypothesis was that the more negatively charged group donated more electron density to the cobalt, making the Co-O bond break more easily. However, the hypothesis needs to be tested with ADF.

(Supported by the Schultz Foundation)

Advisor: Robert Linck, Chemistry

Reference:
Characterizing the Structure of Lesioned DNA Duplexes Using 2D NOESY Spectroscopy

Heather Giguere/2018J and Leigh Tanji/2018

Oxidative damage to the nitrogenous bases of DNA can lead to mutations, carcinogenesis, and even apoptosis. Guanine is particularly susceptible to the formation of such lesions due to its low reduction potential and can easily be transformed into either one of a diastereomeric hyperoxidized spiroiminodihydantoin lesion (Sp1 or Sp2). These highly mutagenic lesions can compromise the stability and configuration of DNA duplexes. The goal of this research is to understand the structural effects of Sp lesions on DNA. To this end, two different 11-mer DNA duplexes derived from the control strand (Figure 1) were analyzed using two dimensional Nuclear Overhauser Effect Spectroscopy (NOESY). One of the derivatives consists of an Sp lesion at the G6 position (Sp1 and Sp2). The other replaces C17 with a guanine base pair, resulting in a G-G mismatch. The synthetic aspect of this project was carried out by another group of students, prior to 2D NMR analysis.

5'- C2 A2 T6 C6 G6 C7 T8 A9 C10 C11-3' 
3'-G22 G21 T20 A19 G18 C17 G16 A15 T14 G13 G12-5'

Figure 1. Control 11-mer DNA duplex sequence.

NOESY spectra were obtained for the G-C, G-G and one of the Sp-C diastereomers samples on a Bruker 500 NMR spectrometer at pH 7 and 8 °C. The resulting data were analyzed using the NMR peak assignment program SPARKY. Figure 2 shows one of the NOESY spectra collected for the G-C control with the regions that we analyzed highlighted on it. Methods for peak assignment included using the "NOE walk" (region C), which allowed for the sequential assignment of each nucleobase's aromatic protons, and the H1' proton of each sugar. Assignments in other regions were made possible through pattern recognition of chemical shifts, and reference to previous literature findings. 

Figure 2. NOESY spectrum of G-C 11-mer at pH 7 and 8 °C showing cross-peak regions of interest
Using the above methods, all desired regions of the control were successfully assigned, and the experimental assignments were found to be comparable to those of the literature (Figure 2). While both the Sp1 and the G-G mismatch spectra remain works in progress, significant headway was made in their assignments, including the discovery of certain chemical shift patterns which will allow for an easier assignment of future spectra. For example, H2’ protons always appear upfield of H2” protons. The Sp1-C data reveal significant chemical shift changes within a three base pair distance of the lesion or mismatch. In the Sp1-C data specifically, the lesion disrupts the NOE walk, and we do not see cross peaks connecting C5 to C7. Furthermore, imino peak assignments appear different than those previously reported in the literature, which has brought about important questions which merit further investigation. The G-G cross peaks associated with base pairs C5, G6, C7, G17, and G16 had shifted compared to corresponding G-C control peaks. Expressively, the peaks describing the imino-iminos walk across DNA strands and the interactions between G or T imino with the opposite amino, shifts to a lower frequency.

Going forward, there will be a need to collect more NOESY data before unambiguously assigning certain resonances, including the imino peaks of the Sp1-C and all the resonances of an Sp-G double strands. This project is expected to continue during the 2016-2017 school year.

(Supported by the Schultz Foundation and the Howard Hughes Medical Institute)

Advisors: Cristina Suarez and Elizabeth Jamieson, Chemistry

References:


Investigating the Role of Succinic and Acetic Acids in Aerosol Mimicking Solutions

Lily Timpane/2017

Atmospheric aerosols impact both human health, and the earth's climate in significant ways. Aerosols are implicated in a range of health concerns related to airway inflammation, and cardiovascular risk factors. Due to their minute size, ultrafine particles scatter light, reflecting it back into space and thereby cooling the atmosphere. Despite this, their net impact on the planet's radiative budget remains uncertain and much more research into the mechanisms and products of their atmospheric reactions is needed.

Last year, Claire Keller's research explored the reactivity of aerosol mimicking solutions, specifically focusing on the effects of simple alcohols and acids on the kinetics and growth of the glyoxal-ammonium sulfate system. Her findings indicated that the addition of succinic acid to a stock solution of ammonium sulfate, glyoxal and ultra-pure water resulted in an increased rate of reaction for the system. She also found that the rate of reactivity of the system was dependent on pH, with a higher pH resulting in an increased rate of reactivity.

My research sought to further investigate these findings, initially through replicating Claire's experiments with succinic and acetic acid. Additionally, I investigated the effects of varying the starting pH of stock solutions in similar experiments. And finally I was able to create a baseline of kinetics and pH data for each acid in the glyoxal-ammonium sulfate system.

The resulting findings supported the hypothesis that the rate of reactivity of the system is pH dependent. However, results also indicated that both succinic acid and acetic acid accelerate the system in some way that is not pH-related. When the starting pH of stock solution was matched with acid-containing stock, both acids were found to increase the reactivity of the system. Given this, the question remains, what is causing the observed increase in reactivity?

I hope to continue my research during the upcoming academic year. I would like to explore this question as well as study the effects of similarly structured acids on the glyoxal-ammonium sulfate system. Ideally the results of this research will provide illuminating clues about the specific reactions taking place within the system, as well as contribute to the eventual identification of the products of these reactions.

(Supported by the Howard Hughes Medical Institute)

Advisor: Andrew Berke, Chemistry

References:


Anesthetics play an important role in the field of medicine for pain management. One of the most commonly used anesthetics is propofol (Figure 1). While it provides significant advantages for hypnosis and rapid onset of action and recovery, its shortcomings include serious hypotension and respiratory depression, especially in the elderly and critically ill. The importance of propofol as a general anesthetic and its drawbacks have stimulated the development of novel anesthetics, specifically analogues of propofol that possess anesthetic potency, yet with fewer side effects. Bearing a structure similar to propofol, 2,6-disubstituted cyclohexanol derivatives are potential propofol replacements since they do not possess a benzene ring which may account for propofol's toxicity. Professor Adam Hall's neuroscience lab has studied isomers of some 2,6-disubstituted cyclohexanols for anesthetic properties, and some of them have shown positive results.

Previous work done in the Shea lab includes the successful synthesis and isolation of 2,6-dimethylcyclohexanol and 2,6-diethylcyclohexanol, and according to the studies from Hall lab, the cis-cis isomer of 2,6-dimethylcyclohexanol was found to be the most promising. 

Research this summer focused on synthesizing and purifying 2,6-diisobutylcyclohexanol in hopes of finding more potent 2,6-disubstituted cyclohexanols. Although the first step of the scheme (Figure 2) was achieved and 1 was successfully synthesized and purified, the second step, hydrogenation of this product, was unsuccessful after several different trials. Thus, 2 became our target while an efficient way to hydrogenate 1 was explored. The reaction scheme for the synthesis of 2 is shown in Figure 3. The reduction of 1 to 2 was successful.

Future work for this project involves synthesis and purification of isomers of both 2,6-diisobutylcyclohexanol and 2,6-diisobutylidenecyclohexanol. In addition, different aldehydes and reactants will be used to gain various 2,6-disubstituted cyclohexanols and other substituted cyclohexanols that are similar to the structures of propofol analogues that Krasowski et. al has reported to be anesthetically potent.

(Supported by the Schultz Foundation)

Advisor: Kevin Shea, Chemistry
References:


Tissue engineering research aims to offer alternative treatments for tissue and organ damage, a major health problem.¹ Hydrogels, hydrated polymeric matrices, are a promising tool for guiding and supporting regenerating tissue.² Essential to biocompatibility in different tissue systems is the addition of features that make hydrogels resemble the topographies and mechanics inherent in the extracellular matrix (ECM), which is the primary scaffold for cellular support and organization in tissues.³ Poly(2-vinyl-4,4’-dimethylazlactone) (PVDMA) is a promising polymer characterized by its reactive functional groups, which allow the polymer to be functionalized post-synthesis in rapid click-type reactions with amines, alcohols, and thiols, which can impart biocompatibility to hydrogels.⁴ Combining PVDMA with diamines results in the hydrogels we are interested in studying.

To test the implementation of ECM topography and mechanics on hydrogels, topographical and chemical patterning experiments were conducted. Topographically patterned gels were synthesized by combining PVDMA with Jeffamine-600 in DMSO, casting the mixture onto nanopatterned PDMS stamps (stripes, wells, and posts), then allowing the polymer to crosslink. The effect of crosslinking percent on pattern size was investigated as well as solvent dependent pattern swelling. Patterns were visualized by functionalizing the gel with a fluorophore and imaging them on an upright fluorescence microscope. For chemical patterning, a fluorophore was cast onto nanopatterned PDMS stamps, then held flat against hydrogels with a dried surface for one minute, then carefully removed and the gel rinsed. Topographically and chemically patterned gels were created by chemically patterning gels imparted with topographical features. Visualization of the 3D features of the gels was performed on a confocal microscope.

Hydrogels easily molded to the PDMS stamp to make topographical patterns, but extended optimization in methodology and alterations in wt % was necessary to get reproducible data for quantification, for which further experiments are necessary. Optimized patterns were imaged with confocal microscopy, taking a series through the gels and creating 3D renderings with ImageJ.⁵ Significant progress was made in selectively chemically functionalizing the tops of topographical patterns and consistently patterning flat gels.

The capability of fine-tuning PVDMA hydrogels makes them an exciting addition to the field of tissue engineering. The possibilities of an easily altered, biomimetic hydrogel can be applied to a vast range of tissue types and applications in these tissues. I will continue this research in my senior thesis, ideally reaching a point of testing topographically and chemically patterned gels in cell studies.

(Supported by the Howard Hughes Medical Institute)

Advisor: Maren Buck, Chemistry

References:

¹Nature Biotechnology. 1999. 17, 506–510. doi:10.1038/80103
Figure 1: Left 3D rendering of a topographically patterned gel. Right A selectively chemically patterned topographical gel.
Site-selective chemistry has always been an enthusiastically researched topic in past decades, for the capability of binding and modifying an interested target in a complicated environment without disturbing other potential competitors is highly demanded in the fields of biological sciences. For instance, chemical labeling is an essential tool for monitoring the behavior of interested biological molecules in vivo; though proteins can be genetically modified to emit fluorescence to be detected, the labeling of small molecules and native proteins requires the tag to be attached to target directly by a highly selective reaction. The inactivation of signaling molecules involved in bacteria quorum sensing by selective ester hydrolysis is a promising application of intermolecular selectivity as well.

The Gorin lab proposed to use DNA-small molecule catalysts (DCats): DNA aptamers are covalently linked to small-molecule catalysts. DNA displays great stability¹, high selectivity, and high affinity. Hypothetically, DNA-aptamer binds exclusively to the targeted substrate, which mimic the working mechanism of enzymes, to increase the efficient concentration of catalyst and catalyze that reaction only. Previously in our lab, some proof-of-concept experiments have been performed to validate the hypotheses. During the summer SURF time, in collaboration with Derr lab at Smith College, total internal reflection fluorescence (TIRF) technique was employed to explore the binding kinetics of DCats with the chosen substrate cholic acid (CA). For the sake of TIRF experiment, cyanine5-labeled cholic acid and biotin-tagged DNA-aptamer were synthesized. The synthesis products were characterized by proton and carbon NMR and mass spectroscopy. By the end of the summer, all required pieces were prepared and confirmed for the sake of future study.

(Supported by the National Science Foundation)

Advisor: David Gorin, Chemistry

(Endnotes)

Chemical tools are increasingly applied to investigate biological systems. By selectively transforming one target molecule in the biological mixture, we could control the biological function of the target molecule. However, the complexity of biological systems presents a challenge to achieving chemical reactions of a specific target molecule without disturbing the function of other biomolecules. In the presence of the abundant functionality in living systems, the introduced molecule could unselectively trigger a large amount of undesirable reactions.

To address this challenge, we propose a potentially selective and efficient method using the DCats: DNA-small molecule catalyst conjugates. Consisting of a DNA aptamer covalently linked to a small molecule catalyst, the DCat hypothetically functions the same way as a modular enzyme. In a modular enzyme, the site to achieve selectivity in substrate binding is separate from the site to achieve reactivity, where the transformation of the substrate takes place. In DCats, the DNA aptamer is a short sequence of single-stranded DNA, capable of binding specifically to its target molecule. This specific binding interaction brings the target molecule in close proximity to the small molecule catalyst, which will subsequently attack the target molecule and achieve the desired transformation.

As further investigation on kinetics could allow us to gain insight into the mechanism of DCat, I performed fluorescent assays to determine the initial reaction rates, which were used for kinetics modeling. Since Michaelis-Menten kinetics is one of the widely used model for enzyme kinetics, and DCat was designed as a synthetic enzyme, we proposed that Michaelis-Menten kinetics would be a good model for the DCat catalyzed reactions. As a proof of concept, hydrolysis of cholic acid umbelliferone ester was studied. The hydrolysis of this non-fluorescent substrate generates a fluorescent signal. By measuring the rate at which the fluorescent signal increases as the reaction progresses, I determined the initial rates of the reaction in the presence of 5 µM DCat and different concentrations of substrate. The initial rates were fitted to Michaelis-Menten equation and an R² value of 91% was obtained, indicating a good fit. The Michaelis-Menten constant, Km was found to be 24.4 ± 8.0 µM, and Kcat was found to be 0.594 ± 0.062 h⁻¹. These kinetic parameters provide important information for catalytic efficiency and selectivity of DCats. I will continue doing more kinetic analysis to confirm the validity of these kinetic parameters.

(Supported by the National Science Foundation)

Advisor: David Gorin, Chemistry
Dating Historical Handwritten Syriac Manuscripts Using Junction Detection

Stephanie Xie/2018

Estimating the ages of undated historical documents is a difficult problem to solve, as little is known about ancient dating and transcription conventions. However, further analysis indicates that there are specific patterns in handwriting unique to different time periods, which can be determined through a method called junction detection. Junctions are the regions formed by the intersections of character strokes, and provide information useful in characterizing handwriting styles. Since junction detection has been successfully used to date medieval writing, we wanted to see whether the same technique would work for ancient Syriac. We duplicated their work, but the results show that the technique does not generalize to our data.

The manuscript pages (Fig. 1) were split into three groups: training, testing, and validation. The junctions of the training data were used to create a Self-Organizing Map (SOM), a data visualization technique that preserves the topological structure within the training junctions. In the first experiment, the training junctions were grouped by manuscripts, and each manuscript was split in half. For each half, we computed the hit histograms, which show how the best matching units of a data set are distributed on a SOM (Fig. 2), and then calculated how often the histograms in the first half matched the ones in the second half. The experiment was repeated, this time with all data sets. In the second experiment, the validation data was used to date the training data, and these dates were then compared to the actual dates of the training manuscripts. Finally, we created a movie that displayed each manuscript's histograms in chronological order, but there appeared to be no significant patterns in handwriting changes over the years.

For the first part of the first experiment, the likelihood of matching histograms was 0.7436, indicating the method was relatively accurate; however, when taking all of the data sets into account, the rate was significantly lower at 0.4086. This could be due to the testing and validation sets containing images that had not been corrected for water damage, resulting in loss of data and therefore increased inaccuracy. The RMSE of the second experiment was 178.3342, so the computed dates were off by 178 years.

The results from both experiments and the movie indicate that junction detection is not a viable method for dating handwritten Syriac manuscripts. More effective methods of historical document dating will be explored in the future.

(Supported by the Schultz Foundation)

Advisor: Nicholas Howe, Computer Science

References:


Fig. 1. Binarized image of a manuscript page.

Fig. 2. Hit histogram of Fig. 1. Warm colors indicate where the input data lines up on the SOM.
My research assistantship in Smith College’s Design Thinking Initiative involved assessing the learning process and acquisition of creative confidence in the first cohort of students of IDP 316 [Critical] Design Thinking Studio, from spring 2016. Creative confidence is the ability to think outside the box and use that ingenuity to actualize ideas into products. Through the research, we hope to determine in what ways the students became more creatively confident and how they think they will apply these skills from the course to their area of work, be it engineering, architecture, social justice work or other fields.

I transcribed the interviews that Dr. Zaza Kabayadondo, co-director of the Design Thinking Initiative, conducted with each of the students last semester. Upon completing the transcriptions, both Dr. Kabayadondo and I made a first round of coding the transcribed interviews. We looked for themes and key takeaways within each student’s interview and between interviews. After separately completing the initial coding, we met and compiled a collective list of emergent codes, or categories of qualitative data from the interviews. Once we had done this, we re-examined the transcripts and the emergent codes, working together to make a list of super-codes that accurately and fully represented the findings from the data. After the super-codes had been finalized, I conducted a second round of coding, re-reading all of the transcripts and assigning super-codes where I found they represented the data.

Finally, I created a spreadsheet with the full transcript for each of the interviews and the corresponding super-codes I had assigned. Since my research was only six weeks long, this is where my part ended. From this point, Dr. Kabayadondo will also complete a second round of coding and then continue on to analyze the coded data from the interviews. Since she is the main instructor for IDP 316 [Critical] Design Thinking Studio, Dr. Kabayadondo will take the findings of this research and use it to craft the course for the second cohort of students, who will meet Fall 2016.

This research assistantship was my second experience collecting qualitative data and coding it. In addition to getting practice with this sort of research, the chance to work fairly independently has been a challenge that will surely prepare me for more similar work in the future. I am interested in design and environmental justice work that is strongly based in a connected community – through education and harnessing people-power towards change making. Because of these interests, it is very likely that the qualitative research experience from this summer will come in use again soon – in my next few years at Smith and afterwards – in order to analyze and grow from the experiences of community members or educational program participants.

(Supported by the Branta Foundation)

Advisor: Zaza Kabayadondo, Co-Director of the Design Thinking Initiative
Cancer is a worldwide epidemic, killing 8.2 million people in 2012.¹ A cure is still being pursued, but research has veered from cytotoxic chemotherapies, to specific therapies targeting molecular pathways that aid in cancer survival.² Targeted therapies for many cancer types—such as pancreatic, ovarian, triple negative breast cancer, and lung cancer—have yet to become available. Mesothelin (MSLN), a tumor cell surface biomarker of the four cancer types, is currently being investigated as a potential target for therapeutics and diagnostics. MSLN is known to bind to tumor cell surface biomarker MUC16, leading to cancer cell motility and invasiveness.³ The goal of this project is to engineer a protein that binds MSLN, blocking this interaction and serving as a potential diagnostic and therapeutic of cancer types expressing MSLN.

Previously in the lab, directed evolution was used on a fibronectin III (Fn3) scaffold to create a candidate protein, anti-MSLN 1.4.1, with promising MSLN-binding capabilities. This summer, a soluble form of the anti-MSLN 1.4.1 was produced. Using yeast surface display (YSD) technology (see Figure 1) and flow cytometry, the $K_D$ of anti-MSLN 1.4.1 and soluble MSLN was found from triplicate titration binding assay data. In the future, the anti-MSLN protein will be conjugated to a polymer drug delivery system.

The polymer, currently being engineered by Professor Maren Buck's lab at Smith College, is most stable in DMSO. Several DMSO incubation experiments were run to test the stability of an Fn3 variant in DMSO. With a formerly-engineered Streptavidin 488-binding Fn3 variant expressed on the surface of yeast and soluble antibody Streptavidin 488, preliminary data was collected on the denaturing of the Fn3 variant in DMSO concentrations ranging from zero to 100 percent DMSO. In addition, a DNA primer of a cysteine tag was created for use in chemically binding anti-MSLN 1.4.1 and the polymer.

The $K_D$ range computed from the YSD system (Figure 1) is 400-1000nM. The target $K_D$ range is 1-5nM (the $K_D$ of MSLN and MUC16), so the sorting of higher affinity candidates will continue. The Streptavidin-binding clone is stable in up to 80 percent DMSO, and only shows minimal reduction in binding in up to 100% DMSO, allowing for the future conjugation of a polymer to an Fn3 variant in high concentrations of DMSO.

**Figure 1: Yeast Surface Display (YSD) system.** YSD was used to display anti-MSLN 1.4.1 on the surface of EBY100 yeast for titration binding assays with soluble MSLN. Stars represent fluorescent signal analyzed by flow cytometry.
References:

Uptake of *E. coli* by *B. calyciflorus* in Natural Systems

Britney Blokker/2017

Ecosystems are more resilient when water is pollutant-free and clean water is a necessity to human life. Filter feeding organisms have the potential to remove water contaminants therefore increasing the quality of water sources they are present in. My project aims to develop an understanding of uptake rates of *E. coli* K12 in *Brachionus calyciflorus*, a species of rotifer. This information can be used in the future to develop a model to predict performance of filter feeding organisms in natural systems.

A new rotifer culture was started using 3-5000 dry resting eggs in synthetic freshwater. The batch was grown until it was stable enough to maintain life after extracting high volumes of the culture to use in experiments. At this point feeding experiments were conducted to measure the uptake rate of *E. coli* K12. Beakers with zooplankton were spiked with the contaminant and water samples were taken at set intervals for up to 36 hours. These water samples underwent membrane filtration, then the filters were placed on selective mTEC agar plates before being incubated and analyzed. By analyzing the plates it was possible to determine the concentration of *E. coli* in the water at the time of the sample. No uptake was observed during the *B. calyciflorus* feeding experiments. One potential reason for lack of uptake was that the concentration of *E. coli* was too high and the concentration of rotifers was too low.

Moving forward I will address the problems faced during the feeding experiments. I plan to increase the concentration of rotifers in the control volumes. In addition to more feeding experiments I will be enumerating cell uptake rates of *B. calyciflorus* using fluorescent latex beads as surrogate cells. By better understanding how zooplankton feed on *E. coli* we can begin to predict how they can be used to enhance water quality.

*(Supported by the Smith College Committee on Faculty Compensation and Development (CFCD)*

Advisor: Niveen Ismail, Engineering

Brachionus calyciflorus viewed under 20x

Fluorescent microbeads on filter
Over Smith College's 2016 SURF program, research was conducted to design and build a device capable of solar azimuth orientation. Once refined, this device will enhance the flight capabilities of Controlled Meteorological (CMET) balloon systems. CMET balloons are altitude controlled balloon systems capable of multiday flights. Longer flights provide more opportunities to collect useful meteorological data; however, battery life is frequently the limiting factor of flight duration. Although equipped with solar panels used to recharge batteries, CMET balloons lack the physical control systems necessary to align their solar panels with the sunlight's angle of incidence; therefore, the solar panels may not always maximize their photovoltaic potential. This prototype aims to align CMET solar panels with the azimuth angle to increase battery life, and thereby prolong flights.

The prototype consisted of a microcontroller, servo, and light sensor adhered to the center of a rectangular balsawood platform with small fans mounted at each end, all of which was suspended by fishing line to simulate the mechanism's suspension during flight. (fig 1) A LED utilized as a capacitor acted as the light sensor and was mounted on the servo. The microcontroller was programed to rotate the servo 180° taking light measurements every 10°, then calculate the maximum light measurement observed and corresponding angle. Based on PID control principles, the microcontroller sent pulses of power to the fans causing the platform to rotate towards the direction of maximum light. This process was programed in an infinite loop, and by continuously having aligned itself with most intense light source, the device eventually aligned itself with the solar azimuth angle.

Although the solar azimuth angle can be successfully located by the prototype, the fan propulsion system cannot consistently position the orientation device successfully. The foremost issue preventing successful azimuth orientation is replacement the servo with a stationary array of sensors. Currently, the rotation of the servo causes jostling and impedes the intended fan propulsion. A stationary array of sensors would eliminate this unwanted motion and allow for further refinement to the fan PID control system.

Although this research was guided by previously developed solar tracking systems, novel attempts to design an elegant, minimal application have been uniquely challenging. The basic functionality of the solar orientation mechanism has been achieved and successive necessary steps have been determined, but much work remains before flight tests are worthwhile.

(Supported by the Schultz Foundation)

Advisor: Paul Voss, Engineering
Understanding Three-phase Systems, Synchronous Machines, Transformers and PowerWorld Simulation

Alysha de Silva/2018

Three-phase systems, synchronous machines and transformers are some main components to be understood when delving into Power Systems. This is important background information for an electrical engineer looking into this specified field. Furthermore, once that material has been covered, applying it to PowerWorld Simulator becomes more appropriate for commercial use.

Power System Analysis and Design\(^1\) was read to obtain background information on these topics and to observe Case Studies which explained the real life applications. Synchronous machines are motors used in power systems for power factor correction, maintaining constant speed over load variances, and for reduced maintenance cost. On the other spectrum, Transformers are used in transmission networks of higher voltages to convert them to more suitable, lower voltages using step up and step down applications. Figure 1 below shows a sample case created to understand how the generators (sun, barrel and star) produce power for the buses (bold, straight lines) and 400MW, 200Mvar loads (thin arrows). The line flow pie charts indicate the percentage MVA Rating out of 2000 that is flowing to its respective bus/load. The arrows along the transmission line show the direction of power flow.

Further study would involve exploring a country or city and modelling its power grid, thus integrating it into PowerWorld. This would be beneficial to the region, as the simulator is capable of determining scenarios that could result in blackouts.

![PowerWorld Simulation](image)

Figure 1 showing PowerWorld Simulation with four loads connected to six buses, powered by three generators.

(Supported by the National Science Foundation)

Advisor: Judith Cardell, Engineering

References:


\(^2\)Power System Modeling Using PowerWorld Simulator and Add-ons, 2008 PowerWorld Corporation
Cancer therapy is headed in a new direction that diverts from the traditional non-specific killing of all rapidly dividing cells and is transitioning to targeted therapies that use drugs designed to recognize specific biomarkers that are prevalent, if not unique, to the cancer cell. One example of a biomarker that is currently under investigation is Mesothelin (MSLN). MSLN is over expressed on the surfaces of triple-negative breast, ovarian, pancreatic, and lung cancers. The interaction and downstream signaling between MSLN and MUC16, another tumor biomarker, has been implicated in the progression, aggressiveness, and metastasis of the cancer. Our laboratory focuses on engineering small proteins that will specifically bind MSLN and either disrupt the MSLN-MUC16 interaction or deliver drug conjugates to the site of the tumor. The appeal of the proteins currently being engineered is that their small size allows better penetration of solid tumors than the traditional antibody.

After multiple rounds of error prone PCR and fluorescent-activated cell sorting (FACS) using a fibronectin scaffold, we currently have a preliminary candidate (αMSLN1.4.1) that has shown minimal binding to MSLN. We have begun to characterize αMSLN1.4.1, with special focus on any therapeutic activity. Although the goal is to get specific binding to MSLN, it is important to note if the addition of the protein alone can cause cancer cell death. To test this in vitro, Cell Counting Kit-8 (CCK-8) was utilized to quantify viable cells by detecting dehydrogenase activity. Ovarian Carcinoma cells (OVCAR-3) were used in all experiments because they have been shown to express both MSLN and MUC16 on their surface. Shown below in Fig. 1 are the preliminary results obtained from the first experiment. Over a time scale of 48 hours, cell viability was monitored to see if the addition of αMSLN1.4.1 would affect the OVCAR-3 cells. A comparison of αMSLN1.4.1 with the original fibronectin scaffold and Mitomycin-C, an anti-tumor molecule, shows promising activity.

While no conclusions can be made about the mechanism and efficacy of our engineered proteins, this preliminary data has sparked many questions on how to improve the current assay, which additional in vitro experiments might be useful in elucidating the mechanisms, and future directions that will lead to in vivo experiments using mouse tumor xenograft models. I plan on investigating some of these questions through a Special Studies in the upcoming academic year.

**Figure 1: Cytotoxicity Activity.** CCK-8 was used in 96-well microplate containing OVCAR-3 cells to determine cell viability. Controls: the wildtype fibronectin scaffold Fn3-WT (+), anti-tumor antibiotic Mitomycin C (-). A survival rate below 100% indicates cell death, while a rate above 100% indicates stimulation of cell growth.
(Supported by the National Institutes of Health)

Advisor: Sarah Moore, Engineering

References:


Since the advancement of technology, end-of-use electronic equipment, also known as electronic waste (e-waste), has been considered as the fastest growing part of the municipal waste stream. Its complexity makes it different from other waste such as paper and bottles. As “green” and sustainability have drawn significant attention recently, e-waste recycling has also been a major focus in agency regulations and technological research fields. The objective of this project is to investigate both the technological and socio-economic considerations of metal recycling from e-waste.

Through reading literature about e-waste and metal recycling, I first searched for conflict metals, a group of metal resources fueling conflicts in multiple areas. I investigated the environmental and societal impacts of conflict minerals, as well as their recycling rates and common recycling techniques. The current situations and challenges of metal recycling in general were then assessed, mainly through the lens of laws and policies established recently and historically, as well as the global e-waste flow.

As I analyze the global e-waste flow stream, I have found that e-waste is associated not only with environmental and health impacts, but also with severe societal implications because of their reliance on precious metals and rare earth elements. From the metal resource perspective, commonly known metals such as gold, zinc, iron, and copper have higher recycling rates than others such as tantalum and tungsten. For e-waste management, organizations and government agencies have been proposing amendments to their previous policies and laws based on emerging managing challenges. The global e-waste flow is also more nuanced than one would usually think, as this field includes trade, environmental issues, as well as social considerations.

To this end, this project needs further revision for the analysis, which will contribute to the background narrative of an ongoing project on environmental modeling and metal recycling.

(Supported by the Schultz Foundation)

Advisor: Paramjeet Pati, Engineering
Zooplankton Abundance and *E. coli* Concentrations in an Ephemeral Stream at the MacLeish Field Station

Christine Hart/2016

Clean freshwater is a limited resource that is in high demand. There are numerous types of pollutants that can lead to impaired water bodies. One type of microbial pollutant is the fecal indicator bacteria, *Escherichia coli* (*E. coli*). *E. coli* is regulated by the US EPA and high densities of *E. coli* indicate the possible presence of more dangerous gut microbes. In addition, depending on the strain, *E. coli* can itself be pathogenic and harmful to humans. In general, high *E. coli* concentrations can be indicative of water quality issues in a water body. Another measure of overall water quality health is zooplankton abundance and diversity. Select species of zooplankton feed by pushing the surrounding water over permeable tissue to absorb food in a mechanism known as filter-feeding. This method of feeding has been previously demonstrated, under certain conditions, to remove pollutants like *E. coli*.1

During this summer, I quantified the natural levels of both *E. coli* and zooplankton at an unnamed tributary of Jimmy Nolan Brook at the Smith College MacLeish field station. The upstream portion of this tributary is forested and the downstream portion is through cow pasture, where inputs of fecal matter are expected to be found in the stream. Water samples were collected during 5 visits to the field site. *E. coli* was enumerated in each sample via sample dilution, membrane filtration, and plating on a selective media. Zooplankton was also quantified and identified using a Sedgewick-rafter cell and a compound light microscope. Other water quality parameters collected were dissolved oxygen, conductivity, temperature, pH, and turbidity.

Across all sample time points, the *E. coli* levels were high in the downstream region. On average the *E. coli* concentrations found in the cow pasture were over a 100 times higher than the EPA standard for recreational water of 126 colony forming units/100ml.2 In contrast, the upstream concentrations of *E. coli* remain below or close to this standard. The zooplankton data still requires more analysis and is currently inconclusive. More work must be done to identify and quantify the different species of zooplankton found within the samples and determine if a correlation exists between *E. coli* concentration and zooplankton densities and types.

*(Supported by the Schultz Foundation)*

Advisor: Niveen Ismail, Engineering

References:
Transparent solar panels absorb all the energy from the sun except visible light. This means that the light we can detect would pass through the solar cell, and the rest would be absorbed by the solar cell—we’d never miss it. Transparency is such a unique characteristic that will greatly expand our use of renewable energy and decrease overall carbon emissions.

This research project involved a literature review of transparent and conventional solar panels, tours of conventional solar panels, and programming the Arduino to track solar cell efficiency. Emphasis on literature was required to fully understand the idea behind transparent solar panels and how they function. Shown in Figure 2 are the key components of a single transparent cell. The thickest layer toward the left is the transparent substrate being coated (i.e. glass), and the right shows multiple layers of photovoltaic (PV) coating. Between those layers of coating are two active absorptive semiconductors that are excited by sunlight and interact, creating an electric field that causes current to flow. Surrounding those layers are electrodes connected to an external circuit, carrying the current out of the device. Because both electrodes are transparent, there is a layer that sends certain wavelengths back for a second time through the active layers.

Currently, efficiency is about 2%, but theoretical analysis suggests a level of efficiency comparable to existing commercial solar panels. The benefits of transparent solar panels are endless—solar cells in all the windows in a skyscraper, for instance, could cut down a large percentage of the electricity needs of a building. Transparent solar panels are an amazing feat of engineering with endless applications, and it is exciting to see continual development.

Figure 1: The absorption levels (black line) of a conventional PV cell versus a transparent PV cell.

Figure 2: Key components in the transparent photovoltaic device.

(Supported by the National Science Foundation)

Advisor: Judith Cardell, Engineering

References:


Human Hearing and Wideband Acoustic Immittance (WAI) Measurements

Jingping Nie/2017

The research has a particular focus on human hearing and wideband acoustic immittance (WAI) measurements, a family of power-based and impedance-based acoustic measurements. Results from the WAI measurements can not only provide diagnostic information on the tympanic membrane and middle ear via a noninvasive method but also help determine ear-canal geometry, which has a range of practical and theoretical implications. Current ongoing work at Smith College involves the measurement of WAI on normal ears with the two most common instruments (Titan, Mimosa), an experimental instrument (ER10x), and control of age, gender, and race of subjects. I have completed Collaborative Institutional Training Initiative (CITI) training for data collection on human subjects, and as of today I have successfully measured over 100 people.

The objective of my work is to perform WAI measurements on human subjects and conduct theoretical and experimental work on acoustic horns. The ultimate goal is to compute time-domain reflectance (TDR) from WAI measurements. To do that, I physically built acoustic horns models by generating their CAD files with known cross-section area functions using SolidWorks and a 3D printer in Center for Design and Fabrication in Smith. Subsequently, WAI measurements were made on the acoustic horns. MATLAB scripts adopted from Dr. Stephen Neely were studied and modified to try to calculate the TDR.

In the following research, WAI measurements made on the horns will be used to calculate the TDR, and cross-section area functions can be then computed from TDR. This step targets at developing and verifying the methods of TDR calculation by comparing the calculated functions with the known ones. The verified method will be applied to the WAI measurements on human ears. Further verification of the TDR methods might be made by comparing the TDR derived ear-canal cross-section area functions of each subject with their 3D-scanned ear molds files.

(Supported by the National Institutes of Health)

Advisor: Susan Voss, Engineering
Determining the Uptake Rate of E. coli by *Daphnia magna*

Sarah Price/2018 and Mariah Ollive/2018

Filter feeding organisms, such as zooplankton, can potentially be used in treatment wetlands and other natural systems to improve water quality. Zooplankton such as *Daphnia*, may have the ability to remove different particulate based contaminants in the water column, such as *E. coli* and algae. This summer we focused on studying the uptake rate of *E. coli* by *Daphnia magna* through various laboratory based feeding experiments.

We tested the effectiveness of the *D. magna* removing a laboratory strain of *E. coli* from water through filter feeding. We completed a total of ten 36 hour experiments. Figure 1 shows the general setup of our experiments. Over the 36 hour duration, *E. coli* levels were reduced by 2 orders of magnitude. This 2-log removal of *E. coli* translates to up to 99% removal of this microbial pollutant. In addition to testing removal of *E. coli* at room temperature, we compared the impacts of two temperatures of uptakes rates. This final experiment compared removal of *E. coli* at 15°C and 20°C (room temperature). Initial analysis of data indicates that *Daphnia* at 15°C removed slightly more *E. coli* than *Daphnia* at 20°C, but statistical analysis and additional data points are needed to determine if the apparent difference in uptake is significant.

Based on the results gathered from the experiments, *Daphnia* shows the potential to be an effective addition in natural systems reduce levels *E. coli* from fresh water sources. In the future we will be studying the efficiency of *Daphnia* removing different types of particulate matter under varying environmental conditions. We will be continuing the research beyond the summer and continue research through special studies. The data that we collect and analyze will be important in understanding and modeling how *Daphnia* can be used in engineered treatment wetlands or other natural systems to improve performance.

(Supported by the Shultz Foundation)

Advisor: Niveen Ismail, Engineering

![Diagram](image)

Figure 1. Beaker A (control) had no *Daphnia* and showed a constant concentration of *E. coli* throughout the experiment. Beakers B, C and D (experimental) each had 40 *Daphnia* and all showed a significant decrease in *E. coli* concentrations by the end of the experiment (up to 2-log removal of *E. coli*).

Reference:

Impact of Reactor Parameters on Product Characteristics in Biomass Torrefaction

Anna Partridge/2016

Forest residues that are currently left on hillsides or burned, can be transformed through the thermochemical process of torrefaction into energy rich fuel sources. Torrefaction increases the bulk density of the wood chips while also increasing their energy content so that they may be co-fired with coal after pelletization. This research explores the impacts of process variables on the ultimate chemical composition of the torrefied product. The impacts of controllable parameters on product characteristics can determine how feedstocks are chosen and utilized to improve economic feasibility and environmental sustainability, while providing a reliable renewable energy source.

Torrefaction experiments were conducted in the summer of 2015 at Big Lagoon, California under the direction and supervision of the Schatz Energy Research Center (SERC). Mass yield, moisture content, temperature, and residence time were measured on site during these tests. SERC later measured energy yield using bomb calorimetry and proximate analysis using a thermogravimetric analyzer in the lab. Analysis was performed in RStudio to assess the correlation coefficients and statistical significance of each variable in linear regression models.

The proximate analysis of the feedstock was found to be impacted most significantly by the mass yield of the product, an indication of degree of torrefaction. As the degree of torrefaction increased, the volatile matter decreased and the fixed carbon content increased on a dry mass basis. The ash content did not change significantly with mass yield. The reactor temperature is linearly correlated with the mass yield and therefore also showed a linear correlation with the proximate analysis compositions of volatile matter and fixed carbon, but not ash. The residence time, moisture content of the feedstock and the feedstock species did not have a statistically significant impact on the variation in the proximate analysis data.

The results indicate that the four feedstocks considered in this study, redwood, douglas fir, tan oak, and hardwood slash respond similarly to torrefaction and that the feedstock type is not a significant predictor of the product proximate analysis. This, as well as the lack of correlation between moisture content and product composition, indicates that the physical properties of the incoming feed are not significant variables that impact the torrefaction process. Rather, temperature inside the reactor is the most significant variable impacting the mass yield and the mass yield plays the most important role in predicting the chemical composition. This extension of my Smith honors thesis will directly contribute to a journal paper compiled by researchers in the Picker Engineering Program and at SERC.

(Supported by the Schultz Foundation)

Advisor: Denise McKahn, Engineering

References:


Van Krevelen diagram indicating atomic ratio for torrefied wood in comparison to coal for current work and Nachenius et al.2 data.
Engineering Proteins to Deliver Therapeutics Across the Blood Brain Barrier

Anisha Tyagi/2018

The blood brain barrier is a highly selectively permeable membrane that is an obstacle to drug delivery in the brain. Of more than 3000 documented in Comprehensive Medicinal Chemistry, 95% of the drugs do not treat central nervous system diseases because the blood brain barrier does not allow any large molecule drugs cross into the brain. In order to overcome the drug delivery barrier, this project aims to engineer proteins that will target the transferrin receptor on the blood brain barrier to carry therapeutic cargo into the brain.

Previous work in lab identified 6 engineered anti-transferrin receptor non-antibody proteins (anti-TfR sequence 6, sequence 1, A14, A19, A22 and B7) to be made using yeast expression systems for further testing. To purify these proteins, nickel affinity chromatography and size exclusion chromatography were used. To shift engineered proteins’ expression to bacteria, bacterial plasmid DNA was ligated with inserts of these engineered proteins. From there, plasmid DNA was sequenced to confirm proper insert ligation. Along with setting up bacterial expression systems for engineered proteins, human colorectal adenocarcinoma cell line Caco-2 was cultured for use in future assays with engineered protein.

Of the engineered proteins, anti-TfR sequence 6 was not purified after yeast soluble expression. Multiple attempts at nickel affinity chromatography were unsuccessful at eluting anti-TfR sequence 6 from yeast growth supernatant. Further efforts were then made to shift expression into bacterial systems. Currently, anti-TfR sequence A14 expression and anti-TfR sequence B7 expression have been confirmed in bacterial protein expression vectors.

A possible explanation for the unsuccessful purification of anti-TfR sequence 6 is its instability in large quantities. The protein may have precipitated out of supernatant at early stages before nickel affinity chromatography, evidenced by a lack of protein in supernatant before the chromatography and after filtration in Figure 1.

This research will continue in the Fall of 2016 as a special studies project with renewed efforts at making engineered protein with bacteria.

Figure 1. Protein gel after second attempt at nickel affinity chromatography. Anti-TfR sequence is ~ 15 kDa. No band in any lanes correspond to a protein of ~15 kDa. (Lane 1—wild type MSLN ~10 kDa, Lane 2—supernatant before purification and after filtration, Lane 3— flow through, Lanes 5-6— 2 elutions)

(Supported by the Howard Hughes Medical Institute)

Advisor: Sarah Moore, Engineering

Reference:

Manipulation and Application of the WAI Database

Tinli Yarrington/2018

Wideband acoustic immittance (WAI) measures are used to describe how sounds flows through the middle ear. Specific WAI quantities include the absorbance and the impedance, which are both functions of frequency. They are used to determine when middle-ear function is normal and to develop a database for WAI measurements across race, gender, and age. All these factors help predict hearing issues in the future as well as compare how they affect the measurements.

The goal of my summer work was to (1) enter data into the database, (2) design methods to graph the data within the database, (3) and to update and manage the website that hosts the database.

One of my roles was to manage the data that was added to the database. I mastered MySQL, the software used for the database, so that I could convert measurements from other researchers into the proper format for the database. I then worked on a brochure that described how to maneuver around the database using MySQL, such as adding, removing, accessing, and updating subject information and measurements.

In addition to managing the database, I worked in Matlab to create graphs of the data within the database. I wrote a program that contained multiple options for the user to graph data from the database. The user can either select a single subject, a subset of subjects based on age, gender, race, and ethnicity, or all subjects from the database. The program then displays frequency on the x-axis for all the graphs, with an absorbance, impedance magnitude, and impedance angle graph for both the left and right ear of the subjects. The programs were added to the database website so that other researchers could easily create the graphs themselves. To complete this program and make it available for other auditory researchers, I mastered Matlab and Wordpress (which involves HTML and CSS) software.

The final products of my research this summer were an updated database, where other researchers can add and easily access measurements, and programs that can generate graphs for easy analysis. For the upcoming year, I hope to reach out to more researchers and add more measurements to the database as well as perfect my Matlab code so the graphs are more clear and easy to interpret.

(Supported by the Schultz Foundation)

Advisor: Susan Voss, Engineering

Figure 1. Output from Matlab code of measurements for Subject 9

Figure 2. Example of MySQL code editing
Historically, the electricity market was mainly governed by vertically integrated utilities (IOU). Vertical integration means that the company has control over the entire supply chain, which typically involves generation, transmission, distribution and marketing of the electricity. This kind of electricity market remained to be a successful structure until the appearances of the advancement of technologies and the deregulation of many other industries in the 1990s. Prior to the 1990s, electricity was primarily generated by coal and nuclear powered plants, which introduced heavy contaminants to the air. Consequently, people began to seek more environmentally friendly methods to generate electricity such as using natural-gas-powered generation units, which could operate on a smaller scale. As a result, the non-utility generators began to appear in the electricity market. As more and more generation units emerged, competition arose among them, as they were not under regulation. The market finally became a deregulated market. Independent system operator (ISO) characterizes the deregulated electricity market, which serves as an intermediary agent between the supply side and the demand side of the market and determines the market-clearing price for each time period.

Electric load forecasting has always been an essential part in the operation of deregulated market for its role in the decision of market clearing price. An inaccurate forecasting will make the ISO come up with a flawed plan, which may in turn result in a waste/deficiency of money and power in the market. For decades, researchers have been working very hard to forecast the electric load as accurate as possible, and they have tried various statistical methods, such as multiple regression and stochastic time series. However, these methods principally use linear models. Due to the nonlinear nature of the electric load, a nonlinear model is expected. Therefore, artificial neural network has become a popular method in electric load forecasting for its exceptional performance in nonlinear modeling.

This summer, I studied the artificial neural network and built a network to forecast the electric load in New England control area. The network did not perform perfectly, but it worked. The next step will be improving the performance of the network by incorporating more input factors and using data from a smaller area. I will continue this project as a special studies next semester.

(Supported by the National Science Foundation)

Advisor: Judith Cardell, Engineering

References:

Severin Borenstein and James Bushnell. The U.S. Electricity Industry after 20 Years of Restructuring. Energy Institute at Haas.
Jeff Lien. Electricity restructuring: What has worked, what has not, and what is next. Economic Analysis Group.

Figure 1: Load forecasting results in ISO New England Control area on various dates.
The objective of our research is to build a converter that receives a digital S/PDIF input value and sends out an 8-bit parallel TTL signal. Currently, Professor Barbara Shinn-Cunningham’s lab, an auditory lab at Boston University, uses a TDT device as a medium to send signals from a computer to their EEG device. However, the TDT device is very expensive and cannot send multi-channel signals. To cut down the cost as well as to enable multi-channel signals to be sent to the EEG device, a S/PDIF to TTL converter was built and tested, building on instructions by Bram Van Dun and Cong-Van Nguyen.¹

Because the DAC they used was not manufactured any more, we ordered a DAC with different chips and made some modifications on the board, mainly on the chip CS8416. Changes are shown in Figure 1. We tested the board by sending different hexadecimal numbers, and we did get the correct output sometimes, but it turned out that it did not function as stable as we expected. Thus, the next step will be to look for patterns from the recorded outputs which used different triggers and different hexadecimal numbers. Ultimately, the hexadecimal numbers sent should match the corresponding binary numbers.

This summer research experience was an excellent opportunity for both of us to apply what we learned at Smith to a real world problem. We were also inspired by graduate students and postdoctoral researchers in Professor Shinn-Cunningham’s lab and had clearer goals for our future career.

(Supported by the Howard Hughes Medical Institute)

Advisors: Susan Voss, Engineering, and Barbara Shinn-Cunningham, Biomedical Engineering, Boston University

Reference:

This summer six Smith students participated in the seventeenth iteration of the Coral Reef Ed-Ventures Program. Coral Reef Ed-Ventures is based in San Pedro Town on the island of Ambergris Caye off the northeast coast of Belize. Originally founded by Paulette Peckol, Al Curran, and Susan Etheridge, Coral Reef Ed-Ventures was created in partnership with Hol Chan Marine Reserve as a way to both conduct research and engage the local community. In the current program Smith students provide two free summer camps to children on the island and conduct research on mangrove and coral reef habitats.

This year in camp, students learned how all the ecosystems of San Pedro are connected, and why conservation of each is necessary. For the older campers, we conducted a biodiversity survey of a populated beach and a mangrove habitat. Advanced camp students also took a trip with a crocodile conservation organization called ACES. Youth camp attendees learned about different ecosystems, culminating in a glass bottom boat trip so that students could see firsthand all that they had learned about in class. The end of youth camp also featured a graduation ceremony for campers and their families, where campers presented projects about a marine topic that had interested them.

Smith students participated in a number of research projects with professors to survey coral reef, soft coral, and mangrove health. They flew drone missions with Scott Gilman in the Environmental Science and Policy Program’s Spatial Analysis Lab to generate aerial images of the reef and mangrove research sites.

Coral research was conducted using scuba and snorkel at Mexico Rocks, a site that very recently received protected status in the Hol Chan Marine Reserve system. The goals of the long-term surveys at Mexico Rocks were to quantify live coral cover and soft coral abundance and diversity and track changes over time.

Similarly, mangrove health and biodiversity surveys were conducted at Belizean Estates, an area that is undergoing intensive land development in response to Ambergris Caye’s rising tourism pressure and population increase. Over time, this research can show how the construction in Belizean Estates affects the natural ecosystem. Aspects of this year’s research will be analyzed and presented as a part of various special studies in the coming semesters.

(Supported by the Environmental Science & Policy Program (ES&P) and Agnes Shedd Andreae 1932 Research Internship Fund, B. Elizabeth Horner Fund in the Biological Sciences and Margaret A. Walsh Grantham Fund, and a gift from Linda Salisbury ’78)

Advisors: L. David Smith, Denise Lello, Biological Sciences, Al Curran, Geosciences, and Miguel Alamilla, Jr., Hol Chan Marine Reserve, with help from Shannon Audley-Piotrowski (Education and Child Studies), Scott Gilman and Jon Caris (Spatial Analysis Lab), and Anne Wibiralske (ES&P).
Estuarine Stewardship and Diamondback Terrapin Conservation in the Chesapeake Bay

Claudia Deeg/2017

This summer, I was part of the stewardship team working at the Chesapeake Bay branch of NOAA’s National Estuarine Research Reserve program (CBNERR), located at the Virginia Institute of Marine Science (VIMS). CBNERR program conducts continuous monitoring projects that observe short-term and long-term changes in the viability of estuarine ecosystems providing important information for developing effective land management plans and conservation policies. I participated in a variety of monitoring activities at the four CBNERR research sites, including habitat mapping, elevation surveys, vegetation surveys, collecting groundwater well data, and taking sediment cores. In the lab I learned how to process sediment cores, determine organic matter content, and run grain size analysis. Additionally, I worked with other branches of the CBNERR office with weather and water quality monitoring. I also helped survey submerged aquatic vegetation. Overall, I experienced the full range of research activities at a long-term habitat monitoring program such as CBNERR.

In addition to my stewardship work, I created a protocol for establishing a citizen nest protection program of diamondback terrapins (Malaclemys terrapin), a.k.a. Team Terrapin. The primary role of citizen volunteers will be to locate nests and install nest protectors. M. terrapin is the only species of turtle which lives in estuarine waters, and is found along most of the east coast of the United States. Diamondback terrapins are threatened by unnaturally high depredation rates of nests and drowning in crab pots. Historically, these turtles were hunted to make turtle soup. This delicacy lost popularity during prohibition era since sherry was a key ingredient in the soup. To provide a strong foundation for the citizen monitoring protocol, I researched M. terrapin life history and nesting habitats in the scientific literature and on reputable websites. I investigated numerous other terrapin conservation programs and interviewed their leaders on how they attained success. Additionally, I identified local groups and organizations who we hope to involve with the project. For a species with a “very high conservation need” in Virginia, this project, particularly the nest protection component, will be vital in conserving diamondback terrapins. The CBNERR staff will launch this program in summer 2017. At the end of my internship I was offered a continuing fellowship with VIMS so that I can continue refining this protocol and helping assist with the start of the Team Terrapin project.

(Supported by the Agnes Shedd Andreae 1932 Research Internship Fund)

Advisors: Anne Wibiralske, Environmental Science & Policy, and Scott Lerberg, Chesapeake Bay National Estuarine Research Reserve Site

References:

On the southern coast of Oregon lies the small crabbing town of Charleston, built at the mouth of the Coos Estuary. In 1974 the South Slough National Estuarine Research Reserve (NERR) was established to protect lands around the South Slough of the Coos Estuary and to provide a site for research and long-term monitoring of this estuarine ecosystem. Understanding the coastal ecosystem dynamics is essential for maintaining the health of the Coos Estuary, allowing it to sustain the major industries of Coos County, crabbing and logging.

My internship focused on the Reserve’s Sentinel Sites program, though I had the opportunity to assist with a variety of other projects. South Slough NERR is one of the many research reserves around the country where NOAA mandates the study of how sea level change influences the vegetation of estuarine ecosystems. These studies contribute to NOAA’s Sentinel Site Program. Throughout South Slough, research sites have been selected where vegetation differs along an elevation gradient as well as a salinity gradient.

My first project was helping to create a field plant identification guide, working from a list of 70 plant species identified from previous biomonitoring in the marsh. The field guide was put to the test as the other interns and I learned the biomonitoring methods, and then again as we taught volunteers. At each of the research sites, we determined the percent cover of all species in a randomly selected 1m² plot. We then determined the stem density and count for the three most common species in the plot. We collected water quality data from loggers in groundwater wells, and we installed new wells at some of the sites that did not have them yet. Accretion rates were measured in a few ways to improve accuracy, with each method determining how much soil had built up since the set table, feldspar, or rebar had been installed.

In the lab, my time was split between data entry (using Access), assisting with the graphic design for the 2016-2021 management plan (using InDesign), editing a map of vegetation spread in a creek overrun by invasive reed canary grass (using ArcGIS), and creating graphs to show the trends of the data collected for the Sentinel Sites program. I was often in the field helping with other research projects including fish seining, lamprey shocking, and eelgrass counting, as well as assisting with educational programs hosted by South Slough. The work that I did this summer contributes to the ongoing monitoring of the Coos Estuary that supports education and decision making in response to rising sea level.

(Supported by the Agnes Shedd Andreae 1932 Research Internship Fund)

Advisors: Anne Wibiralske, Environmental Science & Policy and Jenni Schmitt, Watershed Monitoring Coordinator, South Slough National Estuarine Research Reserve
Communications internship at the National Oceanic and Atmospheric Administration

Sable Liggera/2017

For my internship with the National Oceanic and Atmospheric Administration’s (NOAA) Coral Reef Conservation Program (CRCP) as a communications intern, my main role was to write material for the CRCP’s website and social media sites. This material took the form of web features as well as designing content for the CRCP’s posts on Instagram, Twitter, and Facebook. I conducted interviews, typically with scientists and managers of other environmental organizations working on new projects regarding coral reef biology and reef resilience. A large part of my position was ensuring that the new studies I was learning and writing about from my interviewed subjects were translated in a way that would be easy for a general audience to understand, without losing the accuracy or details pertaining to the subject at hand. When drafting social media posts, I also balanced presenting new data and studies in an interesting and concise way to attract new viewers. Along with writing the content for web and social media sites, I went through all of the CRCP’s camera and video footage and organized these resources so they could easily be accessed in future web content as well as, in the case of the video footage, be used in a NOAA coral reef documentary that is currently in production.

In addition to writing about the work of NOAA and NOAA-affiliated scientists, I also wrote features promoting the CRCP’s work and promotion of other organizations. This project is a special feature highlighting NOAA’s cooperation and backing of other international marine protected areas (MPA). Here, I collaborated both with the CRCP’s and the Gulf and Caribbean Fisheries Institute’s (GCFI) communications officers in designing a theme for the project and conducting interviews.

Through my internship with NOAA I gained insight on the importance of the interconnections among environmental protection agencies to fully address the transboundary nature of environmental issues. I also gained appreciation for how crucial the work of communications officers are in translating scientific research in a way that resonates with and catches the attention of a general audience.

(Supported by the Agnes Shedd Andrae 1932 Research Internship Fund)

Advisors: Anne Wibiralske, Environmental Science & Policy, and Alicia Clarke, Senior Communications & Outreach Specialist at NOAA Coral Reef Conservation Program
Under natural flow conditions, every stream system carries sediment downstream from areas of active erosion. Dams impede this process by greatly reducing the speed of water, resulting in sedimentation behind the dam. Allowing this deposition to continue leads to the filling in of any lake, natural or artificial. Traditional dredging can, however, be a large cost not only to the institution but also the environment. As a cheaper, more convenient, and more environmentally responsible alternative to dredging, Paradise Pond at Smith College was allowed to effectively dredge itself. Following partial drawdown of the pond, sediment from the pond bottom was moved by a bulldozer into high-energy sections of the main stream channel.

The area to be excavated was determined using analysis of orthophotos taken by drones in partnership with the Smith College Spatial Analysis Lab (SAL). Photos were compiled into a digital surface model (DSM) of the exposed pond bottom using Pix4D software. This DSM was then subtracted from a plane at a set elevation, determined using a guess-and-check method and the raster calculator function of ArcMap, to meet permit limitations of 1000 cubic yards of sediment moving downstream. Working with Pix4D and ArcMap during the summer allowed my teammates and me to develop skills in geographic information systems (GIS) and spatial analysis. Following two days of excavation, additional drone missions were flown to determine the exact volume of sediment lost from the excavation site. This volume is approximately equal to that of the sediment relocated to above the sluice gate, as sediment losses through the open gate are expected to be minimal.

Initial removal of sediment was successful, as the pond bottom was stable enough to support the weight of the bulldozer and sediment was eroded efficiently by the high-energy stream. This served both as an effective form of sediment transport and a lesson on how transport relates to the varying energy level of water systems. In future plans for the fall, a high-flow rain event will be used to evacuate the sediment moved to above the dam through the sluice gate. This event will determine the viability of the sluicing method for future pond maintenance.

(Supported by the Pond Project)

Advisor: Robert Newton, Geosciences
Figure 1. Orthophoto taken July 7, 2016 by the drones of the SAL before the excavation of the pond bottom.

Figure 2. Orthophoto taken July 14, 2016 following the excavation, lighter areas show where sediment was removed.
Before Excavation 07/07/16:    After Excavation 07/14/16:

**Figure 1.** Orthomosaic photos of Paradise Pond captured with a DJI Phantom 3 Quadcopter drone and the corresponding sparse Digital Surface Models (DSM) constructed in Pix4D software for pre and post-excavation flights.
Sediment Monitoring in Paradise Pond

Emma Harnisch/2018

Various methods have been employed to manage sediment accumulation in Paradise Pond. Accumulation occurs due to the presence of the dam, which disrupts the natural flow of the Mill River and creates Paradise Pond. In previous years, the preferred method of sediment removal has been dredging—excavating and moving loads of pond sediment to a landfill. This process occurs every 8-10 years disturbs our local ecosystem and remains extremely costly to Smith College.

The alternative method of lowering the water level of the pond to create a flowing river, manually moving approximately 1000 cubic yards of sediment from area of increased build-up, and naturally moving the excavated sediment downstream of the dam was tested this summer. Preparation for this experimental sediment transport began in the summer of 2015 and continued this summer before its implementation. Geographic Information Systems (GIS) was utilized to find area of sediment build-up and create a bathymetric map of the pond bottom. Chemical analyses of both pond sediment and water were measured and evaluated.

To create an accurate map of the pond, required to find the sites of sediment excavation and movement, the campus pond was surveyed using a Leica Total Station. Several drone missions also captured images and data to accurately approximate the location and amount of sediment to transport. Data from the survey and drone missions allowed us to map the sediment elevations on the pond bottom using ArcGIS and Pix4D software. The below map shows the amount and location of sediment moved after the water level was lowered.

Sediment in this area was also cored using a drive core and a Uwitec Gravity Corer. Sediment cores were tested for heavy metals including lead by Inductively Coupled Plasma (ICP-OES) after 2.5cm extractions were digested with nitric and hydrochloric acids. Basic water chemistry analysis of the pond, upstream and downstream, sites was taken in the field—this included pH and conductivity to continue our baseline study of the Mill River. Subsequent to these preliminary analyses, approximately 1000 cubic yards of sediment was excavated and placed upstream of the island to encourage its movement downstream.

This baseline study has helped improve our understanding of sediment flow in Paradise Pond and the Mill River, and how we can move away from dredging methods used in the past. From here, we will observe the transport of sediment over time after its relocation within the pond.

(Supported by the Schultz Foundation)

Advisor: Robert Newton, Geosciences
Ecological and Chemostratigraphic Analysis of Ordovician Coral Reefs in the Steinvika Formation, Southern Norway

Emma Roth/2017

The Ordovician saw a drastic change in reef ecology. Prior to this time, prominent reef builders included sponges and microbes. During the Ordovician, a shift occurred and corals became prominent reef builders. Previous work has shown this change resulted in a higher structural complexity within the reefs. Work completed last year on Ordovician coral reefs in Newfoundland has given preliminary evidence that the coral reefs also had a higher ecological complexity, with a higher abundance and diversity of organisms. The analysis of the Steinvika reefs investigates whether this increase in ecological complexity is a worldwide trend.

Field work was conducted to collect samples of the coral reefs. The reefs are located in the Bunaes member of the 41-m thick Steinvika Limestone Formation. We sampled from two patch reefs and one laterally extensive reef. For all three reefs we also collected samples of surrounding sediments. A total of 37 reef samples were taken. In addition to the reef samples, we also described and sampled 33 m of stratigraphy at a 1 m scale. In the lab, a petrographic thin section was made for each reef sample and powders were drilled from the carbonate stratigraphic samples and sent for carbon isotope analysis.

Initial analysis shows there are some key differences between the reefs of Newfoundland and Norway. The most striking difference is in the surrounding sediments. The Newfoundland reefs were surrounded by sediments with relatively little fossil material (12% of points counted) compared to the reef core (37% of points counted). The Norway patch reefs, however, were surrounded by encrinite, a grainstone made almost entirely of crinoid skeletal debris. Another key difference is the presence of a laterally extensive reef in Norway. No laterally extensive reefs were found in Newfoundland.

The differences between the reefs of Newfoundland and Norway may indicate that they were formed in different depositional environments. The grainstone surrounding the Norway reefs may be evidence of a higher energy environment. I will be continuing this project as an honors thesis. The thin sections will be point counted to gather quantitative data on the skeletal make up of the Steinvika reefs, and the carbon isotope data will be analyzed to better understand the global environment in which the reefs were formed.

(Supported by the Schultz Foundation)

Advisor: Sara Pruss, Geosciences

References:


A field photo of one of the two patch reefs sampled from the Steinvika Formation.
Using Drone-Based Photogrammetry to Quantify Sediment Removal from Paradise Pond

Lizzie Sturtevant/2018

Every 8-10 years, accumulated sediment needs to be removed from Paradise Pond to preserve the look and recreational functionality of this central campus landmark. Sediment accumulation occurs in the pond in response to decreased water velocity due to the presence of the dam that disrupts the natural downstream transport of sediment by the Mill River.

In previous years, costly dredging operations required the excavation and transport of the sediment to a nearby landfill. An alternative method of sediment removal has been proposed that minimizes both cost and environmental impact using the natural flow of the river to flush sediment downstream during high flow events. This is a two-step process where sediment is first moved from the upstream portion of the pond down to the area just above the dam by a combination of mechanical excavation and stream transport during a partial pond drawdown. The accumulated sediment is then released at a later time when the sluice gate at the base of the dam is opened during a high flow event.

In July of 2016, an experiment was conducted to test the feasibility of the first step of this process. The pond was drawn down and sediment was moved with a bulldozer into the newly formed channel to be transported down in front of the sluice gate. A DJI Phantom 3 quadcopter drone was used to collect low altitude (50m) air photos that were analyzed using Pix4D “Structure from motion” software to obtain high resolution Digital Elevation Models (DEM’s) of the pond bottom both before and after excavation of the sediment. Vertical resolution of the resulting DEM’s was generally less than 5cm due in large part to the use of an array of up to 20 high precision control points established using a Leica Total Station.

The use of photogrammetry in this study has greatly improved our ability to quantify the movement and accumulation of sediment in Paradise Pond. With the establishment of ground control points and only 2-3 drone flights, we can collect enough data to map the entire pond basin in less than an hour. Previously, all depth and elevation data were collected using a depth sounder and total station over the course of several days in the field. Moving forward, we will continue to gain experience using Pix4D to improve the precision and efficiency of our data processing and will be able to precisely track the effectiveness of this method of sediment removal.

(Supported by the Smith College Facilities Management)

Advisor: Robert Newton, Geosciences

Figure 1. Orthomosaic photos of Paradise Pond captured with a DJI Phantom 3 Quadcopter drone and the corresponding sparse Digital Surface Models (DSM) constructed in Pix4D software for pre and post-extraction flights.
Japanese barberry is an invasive species that came to the United States in 1864. Since its early arrival it has spread wildly; the plant has been a favored ornamental due to its resistance to deer grazing and brilliant fall foliage and berries. However, Japanese barberry has a number of negative effects beyond crowding out native species, including: increased tick populations, raised soil pH and nitrogen content, attracting invasive European earthworms and intensified soil erosion. While management methods have been tested and reported, I attempted to test and establish the most effective mitigation strategy for Japanese barberry at MacLeish Field Station.

I started the project by mapping the Japanese barberry using GIS, which collected information on the plants location and data indicating the age, number of stems, sunlight, and proximity of the Japanese barberry to native species for each individual stand. From these files I created maps of the invasive at the Field Station based on both stand size (the number of stems) and density within 1.5 x 1.5 meter grid squares. These maps displayed a total of 292 stands within the 50 x 150 meter area tested. This area was then divided into five sections with approximately equal stands of Japanese barberry. From there treatments were designed and implemented.

Known treatments of Japanese barberry are burning, herbicides and manual pulling. Pulling is the most cost-effective and simplistic treatment, while the others have their own issues and benefits. For a primary treatment I burned the base of each cluster due to both ease of practice and the effects of burning, which weakened the thorns of the plants for easier handling later. Four of the five treatment areas were burned, while one was pulled manually as a control area. These areas were then left for a month until the secondary treatment was due.

The divided areas were mapped again to record that only 216 stands remained after the pulling and burning. The secondary treatments of two herbicides (spot-sprayed), burning and manual pulling were then applied to the four sites still containing Japanese barberry. After a month the population will be mapped once more using GIS to test each treatment’s effectiveness. As it stands burning proved to be the least difficult treatment to carry out, but continued research will use data to determine which methods were most sound.

(Supported by the Center for the Environment, Ecological Design & Sustainability, (CEEDS)

Advisor: Reid Bertone-Johnson, Landscape Studies

Reference:

1, Zouhar, Kris. 2008. Berberis thunbergii
Japanese barberry stands mapped proportionally using GIS.
A graph $G$ with order $n$ and size $m$ is a $(k, l)$-sparse graph if every subgraph with order $n'$ for $n'$ less than or equal to $n$ has at most $kn'-l$ edges. For any graph $(V, E)$, if adding an edge $e$ such that $e$ is not in $E$ violates $(k, l)$-sparsity, then we say this graph is a $(k, l)$-maximum graph. Given a $(k, l)$-maximum graph, if adding an edge $e$ such that $e$ is not in $E$ forces some edge $e'$ out of the maximum edge set, then we say the new graph is a $(k, l)$-maximal graph. My major interest is the ratio of edges in $(k, l)$-maximum graph and edges in $(k, l)$-maximal graph with $l$ strictly greater than $2k$. I work on narrowing down the upperbound and lowerbound of the ratio, and also work on proving some $(k, l)$-sparse graph properties. To find the upperbound and the lowerbound, I use 1 spindle strategy and multiple spindles strategy to construct $(k, l)$-sparse graphs with $l = nk$ for $n$ is even, $n$ is odd, $n$ is non-integer, etc. Next, I prove these constructions do not violate $(k, l)$-sparsity. In the process of constructing maximal and maximum graphs, I also prove some $(k, l)$-sparse graph properties as lemmas.

As a result, I find the upperbounds for $(k=2, l=2m)$-sparse graphs with $m$ is even or $m$ is odd, and I find the lowerbounds for $(k, l=2m-1)$-sparse graphs for some $m$ greater or equal to 3 and $(k, l=2k)$-sparse graphs. In addition, I conclude the upperbound for any $(k, l)$-sparse graphs is greater or equal to $\frac{1}{2}$. Furthermore, I prove 7 lemmas. For example, the union of 2 node disjoint $(k, l)$-sparse graphs is still $(k, l)$-sparse.

For now, I have constructed a lot of graph examples, and I look forward to generalizing a stronger statement for random $(k, l)$-sparse graphs. In addition, future work could continue to connect current results to the pebble game algorithms.

(Supported by the Susan M. Rambo 1905 Fund in Mathematics and Statistics)

Advisor: Gwen Spencer, Mathematics and Statistics
This research is a continuation of work that began in spring 2016 with Professor Benjamin Baumer. The objective is to learn more about ‘etl’ package to allow R users to work with medium data that are big and changing through time.

RStudio is a free and open source integrated development environment (IDE) for R, a programming language for statistical computing and graphics, and CRAN is a network of ftp and web servers around the world that store identical, up-to-date, versions of code and documentation for R.

R packages provide users with software that extends the core functionality of R, as well as data that illustrates the use of that functionality. However, by design the type of data that can be contained in an R package on CRAN is limited. First, packages are designed to be small, so that the amount of data stored in a package is supposed to be less than 5 megabytes. Furthermore, these data are static, in that CRAN allows only monthly releases. Alternative package repositories -- such as GitHub -- are also limited in their ability to store and deliver data that could be changing in real-time to R users.

The ‘etl’ package provides a CRAN-friendly framework that allows R users to work with medium data in a responsible and responsive manner. It leverages the ‘dplyr’ package to facilitate Extract-Load-Transfer (ETL) operations that bring real-time data into local or remote databases controllable by R users. The suite of ‘etl’-dependent packages brings the world of medium data -- too big to store in memory, but not so big that it won’t fit on a hard drive -- to a much wider audience.

SQL is a programming language that is designed for managing data held in a relational database management system. The ‘etl’ package helps people with no SQL experience to do data analyzation with medium data.

(Supported by the Susan M. Rambo 1905 Fund)

Advisor: Benjamin Baumer, Mathematics & Statistics
NYC OpenData - NYC 311 Phone Call Information

Wencong (Priscilla) Li

07/01/2016

NYC 311 Open Data

3-1-1 is a telephone number used in the United States for citizens to get access to non-emergency municipal services. The nyc311 package provides an interface to NYC 311 Phone Call information from NYC OpenData. Because these are medium data, the nyc311 packages leverages the etl package for creating and maintaining an SQL database.

Getting started:

Install packages

The etl package provides the generic framework for the nyc311 package. Since the nyc311 package currently lives on GitHub and not on CRAN, you have to install it using devtools.

```r
install.packages("devtools")
devtools::install_github("beongul/nyc311")
```
Basis of Splines on the $A_n$ Root Lattice

He Yun/2017

Splines on the $A_n$ root lattice form the equivariant cohomology ring for a certain affine Springer fiber. Therefore, it is important to study the algebraic structure of the space of splines on such lattice. The previous result shows that all every space of splines has a basis, so the main goal of this project is to find a basis of the above-mentioned spaces.

We were able to determine an upper-triangular basis for the spline space of the $A_2$ root lattice. First we turn the root lattice into an infinite graph with lattice points as vertices, and label an edge with the label of the root to which it is parallel. Second we order the vertices in the graph by choosing arbitrarily an origin and spiral out. The $n^{th}$ element in the basis has $n\!-\!1$ leading zeros and the $n^{th}$ entry is guaranteed to be nonzero. The basis includes the trivial spline (with no leading zero), one degree one class (with one leading zero), and a number of degree two and degree three classes (with two or more leading zeros). The distribution of degree two and three basis splines is shown in the figure below, with the green vertex (the circled vertex) being the origin, red vertices (vertices on the lines bounded by the dotted lines) having degree two, and all the others degree three.

We also found a formula for calculating the number of roots contained in an $m$-dimensional subspace of the $A_n$ root lattice, which can provide information on the minimal degrees of basis splines.

Currently we are looking for a basis of the $A_3$ spline space, and hope to generalize the method to the $A_n$ space.

(Supported by the Schultz Foundation)

Advisor: Julianna Tymoczko, Mathematics and Statistics
This project was a continuation of work that had begun spring 2016. The objective was to write a R software package called ETL, which stands for extract, transform and load. The tool helps to bring data together from public sources, transform into a standard format, and load into the database. The focus of the research was on medium data, which is too big for the software, but could fit into the memory of computer. Since most ETL tools are aimed at small and big data, there was a need for developing a new ETL tool that is specifically for medium size data. In particular, I was working on the data from citibike. In order to build the package, I first pulled data from public API, the official data source of citibike. Then, I wrote codes that covered the automatic downloading process. My programming, problem solving and independent working skills have been greatly improved after the research. Working with another teammate has also been beneficial. By the end, I was able to build a R package that allows users to download the citibike data automatically on their local machine. At the same time, the process was efficient because the files were kept in their drives instead of the software. The work I developed would help people who are interested in citibike data perform analysis easily. However, due to the time limit, I wasn't able to give the package too much flexibility. The ideal scenario would be to give users freedom to choose the specific date of interest. Thus, it would be exciting and rewarding to keep working on the project this coming academic year.

(Supported by the Schultz Foundation)

Advisor: Benjamin Baumer, Mathematics and Statistics
R package based on ETL framework to interface with NYC CitiBike data

citibike

R package based on ETL framework to interface with NYC CitiBike data
citibike is a R package that contains NYC CitiBike trip data from CitiBike website, and it allows users to upload multiple years of monthly citibike trip data to a local SQL database.

CitiBike updates New York City citibike data once per season, so users can get access to trip information from 2013-07 to 2016-05.

This package uses the etl database framework.

Please see the vignette to get started.
Python-based Instrument Drivers for Data Acquisition and Instrument Control

Margaret Allard/2019

Using a computer to collect data makes it possible to obtain large quantities of accurate data in a short period of time. However, when highly specialized instruments are needed, one must create new software in order to communicate with the instrument. I wrote Python-based instrument drivers that, paired with a commercial data acquisition software package called Labber, allow the lab to collect and analyze data more accurately and efficiently. One reason instrument drivers are helpful is that they allow people in the lab to communicate with the instruments without knowing its quirks or the language that the instrument speaks. The instrument drivers allow for users to talk to the instruments through a user friendly interface instead of communicating with them directly. Instrument drivers can also be written to make conversions as data is being collected. One example of that is how when we want to monitor the temperature of a cryogen in an experiment, we sometimes do so by getting a voltage reading from a pressure gauge monitoring the vapor pressure of the gas. The driver can then convert the voltage into a pressure, and then use knowledge of the liquid-gas phase boundary to convert the pressure into a temperature. They can also be used to more easily send calibration data that instruments can store in memory.

With the drivers I wrote, students and researchers can now more efficiently use our equipment to perform experiments. The instruments I wrote drivers for included power sources, function generators, multimeters, temperature controllers, resistance bridges and oscilloscopes. Many of these will be used for lab experiments and in the advanced physics lab classroom. The commercial data acquisition software package uses these drivers to set and read instruments, and to collect data. A user can instruct the computer to sweep a value on one instrument and then read how the values of other instruments change with that. Then it produces graphs and data points that can be exported. As an example, see the image below, which shows a graph that was produced by reading information from a digital storage oscilloscope.

(Supported by the Schultz Foundation)

Advisor: Nathaneal Fortune, Physics
Simulating Damping of the Excitation in Trapped Bose-Einstein Condensates

Shuyao Cathy Gu/2017

Bose-Einstein condensates (BEC) is a state of matter in which most of the boson particles occupy the lowest energy state, providing a chance for us to study the quantum mechanical effects on a macroscopic scale. Our research focuses on finding an algorithm that simulates the oscillation modes of trapped BEC at nonzero temperature. Compared to our old algorithm which is viable at zero temperature, this will extend our scope of the investigation to a physically realizable region.

Gross Pitaevskii equation (GPE), widely used to describe the behavior of BEC, strictly holds true at zero temperature. While experiments with BEC (at a tiny temperature above zero) indicate the existence of damping, GPE offers no explanation. Fortunately, one of its modified versions, Dissipative Gross Pitaevskii equation (DGPE), is compatible with nonzero temperatures by including a damping coefficient. DGPE predicts that, after an excitation, BEC will go through damped oscillation until reaching the equilibrium. We found it the ideal model for our system.

We simulate the condensate in a harmonic trap at breathing mode \((l = 0, n = 1)\), where the condensate is spherically symmetric. By varying the damping parameter and the size of the trap, we control the temperature and the energy of the system. At the outset of the simulation, an excitation is given by making a sudden small change in the size of the trapping potential. We then use time evolution algorithm to solve for DGPE at each time step at each point to get the behavior of the condensate. Our simulation results show damped oscillations as expected, with excitation frequencies consistent with previous theoretical predictions. Furthermore, this result indicates that we have created a robust and promising algorithm for modeling the behavior of BEC at nonzero temperatures in spherical potentials.

In the coming academic year, we will use the same algorithm to mimic the behavior of BEC in shell potentials, in which the condensate is in hollow spherical shell shape instead of solid spheres. The performance of this kind of BEC at nonzero temperature has not been studied yet, and this algorithm offers us an excellent opportunity to observe the excitation modes of the condensates.

(Supported by the National Science Foundation)

Advisor: Courtney Lannert, Physics

References:


Figure 1: Oscillation patterns of the wave function probability density in the center of the condensate at different temperatures.
The energy levels of the beryllium atom (atomic number four) are currently known to a far lower precision than that of other elements in the same group. Improved experimental measurements of energy level transitions could be used to confirm theoretical predictions and improve our knowledge of the atom. Our lab is attempting to perform spectroscopy on beryllium. To do this, we use a well-stabilized tunable laser to scan over a small range of frequencies near the energy level transitions of interest, to determine the precise frequency at which the atoms absorb the light.

To examine the feasibility of and potential sources of error in beryllium spectroscopy, we first performed a similar experiment on cesium. The advantage of cesium is that it has several very strong energy level transitions at known frequencies, so the absorption peaks are much larger and easier to measure. We used a technique called saturation spectroscopy which sends two counter-propagating beams through the atomic sample. When an atom’s absorption spectrum contains two or more nearby absorption peaks, which cesium’s does, saturation spectroscopy produces additional “crossover” peaks exactly halfway between the two saturated absorption peaks. According to initial estimates, the crossover peaks were not halfway between the transition peaks within error. This led to the conclusion that there was a non-random systematic source of error which the model did not account for because it assumed only random error. Changing beam alignment, averaging time, or beam power did not significantly affect the error, implying that non-random error was unavoidable within the experimental design and would have to be accounted for in measurements. Next, we attempted to detect a spectroscopy signal for beryllium. The scans were unexpectedly noisy, leading us to move and realign all of the optics on one optical table, to eliminate noise from tiny shifts in relative table heights. The move decreased the noise in our measurements; however, there was still no measurable amount of absorption by the beryllium sample.

The attempted beryllium scans indicated that the beryllium absorption signal was weaker than hoped. The next step is to lock the laser, which will make it possible to average over multiple scans, potentially revealing a small signal not visible on one scan. If there still isn’t a visible absorption signal, we will have to find a way to increase the amount of absorption, such as using an optical cavity to reflect the light through the beryllium sample repeatedly.

(Supported by the Schultz Foundation)

Advisor: William Williams, Physics
Modeling Scalar Fields in the Early Universe

Lauren Stanley/2018

In a world of classical physics, the universe is made entirely of particles and a few forces, such as gravity, that are governed by Newton's laws. However, with the advancement of physics, fields were introduced as a separate object that was not made of particles. A field, such as an electric or magnetic field, is a quantity that has a value throughout all of space. In this research project, we are primarily concerned with classical field theory.

In cosmology, the most widely recognized theory about the very beginning of the universe is Inflation Theory. This theory states that in order to have a universe like the one we observe, there must have been a point in time around 14 billion years ago when the universe was undergoing exponential expansion, known as ‘inflation.’ Since the rate at which the universe expands is determined by the energy density of the universe, in order to undergo exponential expansion, the universe must have been dominated by a scalar field, the only known mechanism with the right energy density. This research project was focused on modeling scalar fields according to the equations derived from the scalar field equations of motion, and exploring scalar fields that were minimally coupled and conformally coupled to gravity. Using a Mathematica program, 2D models were generated for several different potentials exploring cases where the geometry of the universe is flat, and 3D models where the universe could be open, flat or closed. Models were also generated which showed the infinite limits of the scalar field each case could be explored more fully. By generating these models, we can start to understand how scalar fields behave and what conditions lead to inflation.

The result of the work this summer was completed models for 2D and 3D graphs which were minimally coupled to gravity for a quadratic potential, a constant potential, and a quadratic plus a constant potential. In the conformally coupled cases I completed 2D models for the constant and quadratic plus constant potentials. I also have incomplete models for the 2D quadratic and 3D models for each potential.

These models show a little more clearly the sort of behavior we can expect from a scalar field, and which potentials will lead more easily to inflation and which will require more fine tuning. Hopefully, with more models we can start to evaluate which potentials are most likely representative of our universe.

(Supported by the Committee on Faculty Compensation and Development, (CFCD))

Advisor: Gary Felder, Physics

Figure 1: Models for a Quadratic Potential. From left to right, 2D minimally coupled, 3D minimally coupled, 2D conformally coupled (incomplete), and 3D conformally coupled (incomplete).
Inspired by the jeweler's refractometer,¹ we have designed a laser refractometer capable of measuring refractive indices of mineralogical samples with high precision, approaching 1 part in $10^5$. The refractometer uses a He-Ne laser of wavelength 632.8 nm to illuminate the planar surface of a glass hemisphere made of SF-11 Schott glass with index 1.77862 at 632.8 nm. The laser beam enters the curved hemispherical surface from below and is focused to a diffraction-limited spot size of 5 µm at the center of the planar surface. All the rays in the focused beam enter the hemisphere along a diagonal and therefore suffer no refraction at the surface. The mineralogical sample to be measured (typically a thin section) sits on top of the planar hemisphere surface. Rays striking the hemisphere-sample boundary at shallow angles above the critical angle for total internal reflection (TIR) are 100% reflected, whereas rays entering more steeply are only partially reflected, producing a dark-bright boundary in the reflected beam. The position of the TIR boundary in the out-going beam is measured using a linear CCD array with 7-µm pixel resolution, and Snell's law then gives the desired index in terms of the critical angle.

We have used a set of known isotropic index standards to calibrate our instrument and correlate pixel number with angle. Subsequently we have successfully measured the indices of a number of other standards treating them as unknowns. Because of the tightly focused beam, our instrument also has high spatial resolution, so the index of individual grains within a sample can be measured. The instrument is non-destructive and will accommodate virtually any sample with a polished surface. The hemisphere/sample assembly sits in a rotation platform so that the crystal orientation can be smoothly varied. The refractometer design can thus accommodate anisotropic crystals, with the goal of measuring two (uniaxial crystals) or three (biaxial crystals) principal refractive indices on a single surface with high precision.

(Supported by the Schultz Foundation)

Advisor: Doreen Weinberger, Physics

Reference:

Does That Pose Become You? Testing the Effect of Body Posture on Self-concept

Alexa Barriga / 2016

Self-concept refers to the accessibility of traits, roles, relationships, and beliefs that comprise one’s meaningful, core psychological self. Self-concept expansion predicts a range of adaptive outcomes such as job satisfaction and commitment, a heightened persistence on cognitive and physical tasks, and an increase in self-efficacy. An intriguing possible but as yet not studied cause of self-concept expansion is the posing of one’s body expansively, i.e., “power posing.” In Study 1 (N = 65), we investigated if body expansion, compared to body contraction, would result in differences in self-concept size. We closely followed the same experimental method as Carney, Cuddy and Yap’s well-known 2010 study on power posing and found that posing had an effect of moderate magnitude (d = .58) on self-concept size in college women as measured by the Twenty Statements Test. Participants who were randomly assigned to hold expanded poses under the guise of a cover story centered around testing the function of wireless electrodes, wrote significantly more self-statements than those who assumed contracted positions (Mexpanded = 18.16, SD = 3.25) and (Mcontracted = 16.03, SD = 4.10) conditions, F(1, 63) = 5.33, p = .024. In pre-registered Study 2, we tested if this finding was replicable and extended this research by aiming to characterize the process by which it occurred. Again, one hundred and twenty-eight women students were randomly assigned to hold either expanded or contracted postures. They completed surveys measuring two general classes of potential mediators (“broaden-and-build” and “narrow-and-disrupt”), four indices of self-concept size, and a measure on body self-objectification, which we explored as a potential moderator. Results showed that posture did not affect any of the self-concept size measures, nor was it moderated by self-objectification. Although there was no effect on self-expansion, in exploratory analyses assigned posture did affect one of the broaden-and-build measures: psychological flexibility. Specifically, expanded poses increased psychological flexibility. Results of Study 2 indicates that holding an expanded versus contracted bodily posture may not be enough to induce changes in self-concept size. Of course, a lack of main effects could be due to a range of unmeasured confounders, and/or the fragile and transient nature of the effect. Before ruling out the potential for an effect of power posing on self-concept expansion, future research should simplify the replication of our first study by solely focusing on the four self-concept size measures; the additional search for potential mediators may have inadvertently washed out an effect. As well, findings from Study 2 also suggest the need for further research on the possibility of a link between power posing and psychological flexibility.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Benita Jackson, Psychology
Perfectionism has been studied primarily in adults, and very little is known about how parenting characteristics, mental health concerns, and cognitive functioning relate to perfectionism in children. The purpose of this study is to learn more about the antecedents, correlates, and consequences of childhood perfectionism. The investigation was conducted among a community sample of primary caregivers and children (n=56 parent-child dyads). The study involved giving children (ages 8-12 years), and one primary caretaker, a set of questionnaires about these topics. Additionally, children were asked to engage in several problem solving and puzzle tasks related to cognitive and academic skills. Preliminary analysis provides evidence for the expected positive association between perfectionism and psychopathology in children. Self-oriented perfectionism, or setting high standards for oneself and being exceedingly self-critical, in children was found to be positively associated with measures of generalized anxiety (r=.309; p<.01), panic (r=.329; p<.01), and social phobia (r=.526; p<.05). Socially prescribed perfectionism, or the need to meet expectations of others, was found to be positively associated with measures of social phobia (r=.502; p<.05) and panic (r=.264; p<.01). Further, evaluative concerns perfectionism, or self-critical performance evaluation, was found to be positively correlated with all measures of child psychopathology except separation anxiety (p<.05). These findings are significant as targeting maladaptive perfectionism in preventive and treatment programs may help alleviate the psychological distress of children and further their emotional well-being.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Alexandra Burgess, Psychology
Operant Conditioning in Meadow Voles

Katrina Blandino/2017

Meadow voles (Microtus pennsylvanicus) provide an effective model for studying social behavior since they form a partner preference for a same sex partner, allowing the study of non-reproductive social behavior. Oxytocin affects their social behavior; decreased receptor density in the lateral septum has been linked to decreased partner preference. Less understood about social behavior is motivation: the reward value of social interaction. It has been measured using conditioned place preference, a test that determines if a rodent shows increased preference for an environment associated with social interaction. The purpose of this study was to complete pilot testing for a project that would ultimately quantify social motivation in meadow and prairie voles using operant conditioning. Since operant conditioning with a lever operated chamber has not been tested in meadow voles, this research would determine if a vole could learn to press a lever.

Operant conditioning chambers and clickers were purchased from Med Associates and meadow voles were trained to lever press for food. Voles were trained daily under a fixed ratio (FR) 1 schedule for 20 days and then under a progressive ratio (PR) 1 schedule. Non learners (voles that consistently did not press or show interest after 15 days) were dropped from the study. Two voles were tested with 1 hour testing sessions while two others with 30-minute testing session to maximize the numbers of voles in the study. The pair that were trained daily for an hour were tested under an FR1 and PR 1 schedule while the others were not due to time constraints. A summary of results for the voles under FR1 and PR1 are below as of Day 35 of testing (Table 1). While they had had a consistent pattern of 2-6 presses per day, a clicker was added to the training, which facilitated the learning process. As a result, on Day 35, they pressed at a higher rate. Since voles were able to be trained to consistently press the lever each testing session, it was determined that it is possible to train voles to press the lever. This sets the foundation of a thesis project for the academic year 2016-17. The project will compare the social motivation of female meadow and prairie voles. Operant conditioning in a modified chamber will be used to measure the motivation of a vole to huddle with a same-sex partner.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Annaliese Beery, Neuroscience

References:


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Table 1. Summary of results for two voles under PR1 training.

Figure 1. Experimental setup of lever pressing chamber and food dispenser
Characterizing Baseline Vitals in Meadow and Prairie Voles.

Chase Freschlin/2017

This study aims to characterize the baseline vitals of prairie voles (Microtus ochrogaster) and mountain voles (Microtus pennsylvanicus). Knowledge on what regular heart rate, temperature, and activity look like offers invaluable information on an organism’s biology. Such information allows one to research biological effects of behavior in response to the environment, which opens new methods for behavior quantification. Specifically, we hope to look at heart rate variability (HRV) and how this changes when the vole is exposed to different treatments. Along with chronic, fast heart rates, HRV is one of the premier characteristics that indicates poor health. Good health is marked by high heart rate variability, which indicates a fast recovery to baseline from stimuli. Decreased HRV can be an early indicator of any number of diseases, including cancer and poor recovery after surgery. Each beat of the heart consists of several different electrical stages. The pinnacle of the electrical response is the R-wave, which is characterized by the distinctive spike then dip in a cardiogram. The interval between these R-waves (R-R interval) is proportional to the heart rate. By analyzing the variability between the R-R interval, we can gain a better understanding of how the heart rate varies.

Prairie voles and mountain voles were surgically implanted with a small transmitter that records heart rate, temperature, and activity. Voles were allowed to recover for seven days before a baseline recording was taken over five days. During this time, ten minute recordings were collected every hour on the hour. After the baseline, each vole was subjected to four experimental trials with one day of recovery in between each treatment. The treatments were as follows: atenolol, atropine, saline, and a solution of atenolol and atropine. To collect the injection recordings, 30 minutes of baseline was collected pre-injection followed by 30 minutes of data collection post-injection. The resulting data was then analyzed to determine the heart rate, heart rate, temperature, and activity of the vole (Fig. 1).

While several voles have undergone the experiment, we regrettably do not have any results at this time. However, we expect that atropine and atropine plus atenolol will lower the HRV while atenolol and saline have no effect.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Annaliese Beery, Psychology

Fig 1. Baseline cardiogram of test subject pre-injection. This cardiogram depicts the heart rate recorded by the implanted device. The black lines denote where the program recognizes an R-wave. Blank spaces occur when there is data with different parameters from what the program regards as R-waves. These sections must either be manually marked or omitted from the data.
 Assessing the Processes Behind Language Learning

Halimat Ipesa-Balogun/2017

From infancy through preschool, children’s language abilities tend to grow over time. One of the many factors used to explain this growth is called fast mapping. Fast mapping is the process of connecting objects or actions to new words after very little exposure to the new words (Rice et al., 1990; Golinkoff et al, 1992; Golinkoff et. al, 1996). Yet, fast mapping is not typically evaluated in language assessments because they usually focus on “what a child knows” as opposed to “how a child learns” (Hirsh Pasek et al., 2005).

This current study involved the development of a fast mapping subtest that was situated within a larger touch-screen language assessment for two year-old children. Within this subtest, half of the items targeted the students’ ability to initially associate novel nouns with novel referents when they are placed amongst familiar objects. The other test items measured the ability to extend the initial association to contexts, where there are both familiar and novel objects. For example, in the former scenario students could be asked to locate the nonsense noun “bep”, amongst three options: a backpack, a marker, and a novel item. After responding, students would be given the opportunity to extend the association by locating the “bep” amongst three other options: a slightly modified version of the same novel item, another novel item and another familiar item. The objective of the current study was to compare the performance of twenty, two year-old children on the initial association and extension test items. It was predicted that students would perform better on the initial association items. Yet the results demonstrated that there was no significant difference in performance between the two forms of assessment, t (19) = 1.99, p < .05. This similarity in performance was further substantiated by the significantly moderate, positive correlation between the percentage of correct initial test items and the percentage of correct extension test items (r = .46, p < .01 ) (Figure 1). This suggests that initial association test items and the extension items give similar insight into students’ abilities to learn words.

Figure 1. Scatterplot of participants’ percentages of accurate performance on initial association and extension tasks

(Supported by the Mary Sweig Wilson Undergraduate Research Fellowship)

Advisor: Jill deVillers, Psychology
References:


Assessment during the early stages of language development is crucial for timely identification of language delays. While the importance of early identification is known, difficulty arises when testing 2-year-olds in distinguishing between those who simply have poor expressive language and those with a true language delay. Existing tests of language comprehension for 2-year-olds must be improved to meet demands of accessibility and ease of administration and interpretation of results.

Currently, a touchscreen language assessment has been developed for children between 3 and 5 and is undergoing publication. Working with one of the primary investigators on the original project, Jill de Villiers, I created a pilot touchscreen language comprehension test for 2-year-olds in preparation for a larger downward extension of the existing test. Using a program called Kids Interactive App Maker, I developed a tool targeted to test 2-year-olds’ comprehension of words and simple sentence structures. A point of interest is the comparison between touching and dragging as it will influence the final design of the app.

The test is narrated beginning with four training items to teach the child how to touch or move objects. Five language tasks targeting comprehension of nouns and prepositions, negation and transitive sentences and noun fast mapping followed (shown in Figure 1). The vocabulary level was age-appropriate while the tasks varied slightly in difficulty. Each task had six items, 3 of which were dragging, 3 touching. I made two versions of the app to counterbalance response type.

Initial piloting of the app (N=20; mean age = 33 months) shows touchscreen testing to be a suitable tool for 2-year-olds. Toddlers are able to listen to the prompts and complete the tasks, finishing the test above chance on 8/10 item types. Additionally, we found no significant differences in regard to dragging or touching. For this reason, dragging may act as an effective tool for testing linguistic contrasts that were more difficult to convey with static images.

As a continuation of this project, we hope to create a full test of more items that are discriminating and vary in linguistic form. The test design will incorporate many aspects learned from piloting and will continue to be age-appropriate and compelling for a 2-year-old to complete. Our hope that a simplistic, accessible test such as this will change the way language tests are administered for young children to aid in the identification of those in need of early intervention.

Figure 1: Sample items from the app:

Preposition: Put the dog under the chair
Negation: Find the boy with no shoes

(Supported by the Mary Sweig Wilson Undergraduate Research Fellowship)

Advisor: Jill de Villiers
Wh Adjunct Questions in Child Language Acquisition

Jessica Kotfila/2018

The syntax of the English language allows for Wh movement, wherein the Wh or question word of the sentence could apply to multiple clauses of a question. Some verb combinations produce more short distance (SD) answers than others, i.e. “tell” versus “say” or “think”. Parsing is preferential, and linguists like Akira Omaki, assert that children’s gap filling tendencies lead them to a SD parsing preference. But other research by Roeper & de Villiers suggests that children are drawn to the final verb. The present studies explore the effects of question, verb type, and recursion on answer preferences.

54 children (aged 3-5) and 56 adults received a carefully designed series of 9 stories accompanied by corresponding picture stimuli. At the end of each story, the subject was asked a wh adjunct question pairing “when”, “why”, or “where” with “say”, “think”, or “tell” to examine differences across verbs and questions. The stories themselves were constructed to include a false complement so that a second long distance answer would be available, integrating only the last verb for a reality answer. Each of the 9 questions had 3 possible answers (1=SD, 2=LD Reality, 3=LD), and the order that each answer appeared in the story was varied to rule out a recency effect (Fig 1). All children also received a touch screen linguistic assessment, the QUILS, developed by Jill de Villiers et al., which allowed for analysis based on linguistic skill and not exclusively on age.

The results revealed children’s parsing resembles adults in our study, with a preference for LD but a tendency for reality errors in children SD parsing was rare in both children and adults (Fig 2). Reality errors correlated more with linguistic skill than age. The study continued with a second test, which 28 of the children and a group of adults received. The second test included 6 story/question sequences designed with recursion. These questions allow integration of a third verb resulting in 6 possible answers (Fig 3). It appears that recursion reduces the occurrence of reality answers in children, while maintaining a bias for LD parsing in both children and adults. The implications of this study are valuable in the fields of both syntax and child language acquisition because they suggest that the syntax of children does not contrast sharply with that of adults, at least in the case of wh adjunct questions.

(Supported by the Mary Sweig Wilson Undergraduate Research Fellowship)

Advisor: Jill de Villiers, Psychology
This man came to clean out the chimney with his long brush. See? The boy was coming home from school and saw the man up on the roof. A cat was up there too. His mom heard noises from up above and asked the boy what it was. He said, "A man is up on the roof rescuing a cat".

Why did the boy tell his Mom that the man was on the roof?

**Answer choices**

1 (SD) first verb = because Mom asked him about the noise
2 (reality) = 2nd verb= *to clean the chimney
3)(LD) wh trace is in scope of both 1st and 2nd verb= to rescue a cat

Figure 1

Figure 2

**Adults (56)**

**Children (54)**

**Aged 3-5**
Story: Billy got a train set when he went to see his Grandma in the summer. One night, Dad said to Mom, “I really like that train Billy got on his first birthday.” Mom was a bit sleepy, so she didn’t listen very well. The next morning when she was taking a shower, she laughed about it. She thought Dad was talking in his sleep and said that Billy got the train when he was first born!

Question: When did Mom think Dad said Billy got his train?

Answers:
1 first verb (think)= in the shower
2 2nd verb (say)= in bed at night
3 3rd verb/reality (got)= in the summer
4 Integrate (think+say)= while sleeping
5 Integrate (say+ got)= at first birthday
6 Integrate (think+say+got)= when born

Figure 3.
Exploring Sex Differences in Circadian Disrupted Mice on a High Fat Diet

Caroline Labriola/2018

This research sought to uncover any potential differences in signs of metabolic syndrome due to sex in circadian disrupted mice on a high fat diet. It was inspired by a human study conducted on nurses who were more likely to experience symptoms of metabolic syndrome if they participated in shift work with constant rotating hours.1 We mimicked this environment with an altered light cycle, and hypothesized that female mice would experience fewer signs of metabolic syndrome and overall weight gain due to their high levels of estrogen. This prediction was inspired by a study that used ovarectomized female mice lacking the ability to produce estrogen who experienced far fewer cases of metabolic syndrome after circadian disruption than those who could produce estrogen, suggesting that estrogen is responsible for combating the negative metabolic effects of circadian disruption.2

Therefore, we predicted that male mice sans the estrogen-producing ability of females would experience both more weight gain and markers of metabolic syndrome under a circadian disrupted cycle than females.

Fourteen mice (7 male, 7 female) were randomly selected by paired starting weights to enter either a regular light cycle (12: 12 or T24) or a circadian disrupted cycle (10: 10 or T20). All subjects were fed high fat chow. Subjects were weighed once a week throughout the duration of the 10-week experiment. One glucose tolerance test (GTT) was conducted on each mouse the last week of the experiment, and heart, soleus, liver, pancreas, white adipose tissue, brown adipose tissue, blood, and suprachiasmatic nucleus samples were collected for further analysis of metabolic syndrome markers.

The initial findings of this experiment show that females under T20 have a higher normalized percent weight gain than females under T24, and that males in both categories have a higher normalized percent weight gain than both categories of females, but the difference between the effects of T20 versus T24 in the males appears to be smaller.

The data suggests that there is a difference due to sex on weight gain independent of light cycle, and a larger effect of T20 on weight gain in females versus T24 as opposed to the same comparison in males. The GTT results and the tissues collected are next to be analyzed during a special study this fall.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Mary Harrington, Psychology


2. L Zhu, F Zou, Y Yang, P Xu, K Saito, A Hinton, X Yan, H Ding, Q Wu, M Fukuda, Z Sun, Q Tong, Y Xu. Estrogens Prevent Metabolic Dysfunctions Induced by Circadian Disruptions in Female Mice. Endocrinology, March 2015; 156: 2114-2123
Image comparing females and males on a high fat diet in T24 vs T20. The blue and orange dots represent comparisons to a previous experiment using only females on high fat diet in T24 (blue) versus normal chow in T24 (orange).
Understanding Intergroup Relations Among Disadvantaged Groups: How Collaborative Collective Action can be Achieved?

Murong Li/2017

In 2014, the incident of Peter Liang, a Chinese American police officer, shooting an unarmed African American evoked wide protests across two racial communities where Chinese American protesters requested free trial for Liang while African Americans perceived Liang’s indictment as justice being served.

Though two racial groups had different appeals, there were some underlying similar interests such as appeal for judicial justice with regard to police shooting that could be achieved through the collaboration between two groups.

Having reviewed many related studies such as Social Identity Model of Collective Action (SIMCA), Common Ingroup Identity Model (CIIM) and Dual Identity Model (DIM), similarity with respect to disadvantaged identity stood out as a key moderator in smoothing intergroup relations. 1 It has been argued that similarity with respect to disadvantaged identity encourages groups to form coalition when a new disadvantaged categorization is created and the primary group identity is intact as opposed to CIIM at which primary group identity is submerged by the newly created superordinate identity.2

As illustrated in the preliminary model, two separate groups are presented at the bottom where it is proposed that each group can form its own collective action based on SIMCA theory, which suggests that the incorporation of perceived injustice, efficacy, and identification with the ingroup promotes individual’s participation in collective action.3 Moreover, I’m interested in whether priming subjects with similarity with respect to disadvantaged identity would increase their tendency to join collective action together as a bigger unity.

The research that I will conduct in my senior year is expected to enrich the literature of studying intergroup relations.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Fletcher Blanchard, Psychology

References:


The Use of Operant Conditioning to Assess the Presence of Affiliation Between Peer Meadow Voles: Lever-Pressing for Food Reward

Sarah Lopez/2017

Meadow voles are asocial, sexually promiscuous, and territorial rodents throughout the summer months, that exhibit group behavior in the fall an winter months. This shift in behavior is caused by day length and makes studying sociality and affiliation feasible in a lab setting. Day-length differences in female meadow voles cause them to perform same-sex huddling with their cage-mates over novel voles, which indicates that there is some motivation other than warmth causing voles to huddle. The purpose of this SURF project was to develop a method for assessing motivation to huddle between peer voles.

Operant conditioning was used to train 14 female meadow voles to press a lever to receive a food reward. Subjects were placed on a mild food restricted diet throughout testing in order to induce motivation for food. Voles underwent a maximum of 20 days of shaping to receive a food reward on a fixed ratio schedule. Subjects that learned to lever press progressed to 10 days minimum of a progressive ratio schedule (PR1). Sessions were either 30 minutes or 60 minutes long, after which voles were weighed and returned to their home cage. Five voles were removed from testing due to unhealthy weight loss or death of themselves or their partner. Chamber apparatus and accompanying programs were purchased from Med Associates Inc.

A main focus of this stage was to determine if voles could lever press. Two voles successfully made it to PR1, two voles were labeled non-learners, and the remaining four did not complete training due to time constraints. The greatest number of presses in a single session of PR1 was 53.

While there were many obstacles to face throughout setup and the duration of this summer research, many invaluable things were accomplished. The results are not overwhelming, but they do demonstrate that meadow voles are trainable and show potential for further study. The summer was essential for determining lever-pressing feasibility, which was shown. Future stages of this work will include the social aspect of this experiment. The progressive ratio demonstrates the level of motivation in a subject and thus will be used in determining social motivation in affiliative behavior in voles. Construction of the social apparatus occurred during SURF as well. Work will continue during the regular school year as part of these projects.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Annaliese Beery, Psychology


Figure 1. Image of side-by-side operant conditioning chambers from Med Associates Inc.
Endogeneous circadian rhythms, that span approximately 24h, regulate numerous cellular and systemic bodily processes. These rhythms are entrained by environmental cues such as light and temperature; and when these cues are disrupted the effects can be deleterious. Examples of ways in which people disrupt their circadian rhythms are through shift work and chronic jetlag.

One of the more recent pathologies to be attributed to circadian disruption is dysregulation of the immune system, which can lead to inflammation. Inflammation in peripheral systems can cause microglia in the brain to become activated. These glial cells are commonly thought to contribute to immunity in the brain and rapidly respond to protein markers that indicate the presence of infectious agents, trauma and inflammation. The effect of circadian disruption on neural inflammation is not yet clear. However, since neural inflammation is thought to play a role in neurodegenerative diseases, it is critical to understand how circadian disruption might contribute to these diseases.

In order to work towards achieving this understanding, I am in the process of creating an immunocytochemistry protocol for double fluorescent labeling of activated microglia in mouse brains. Microglia have two common morphologies: ramified and activated (Figure 1). The activated form is indicative of their immune response. The Iba1 protein is expressed in both morphologies; however, the CD68 protein is expressed only in the ramified form. Therefore, once the double staining protocol has been established, it will allow me to visualize and semi-quantify activated microglia.

This is a work in progress, and I have yet to interpret the stained cells. However, my working hypothesis is that there will be an increase in activated microglia in the brains of circadian disrupted mice. I intend to continue to perfect and implement the protocol during my special studies in the fall. Then, I intend to use the results to inform my thesis project, which will investigate the role of circadian disruption in neural inflammation.

Figure 1. (A) An illustration of ramified microglial cells labeled with a red immunofluorescent label specific to Iba1 (whole cells). (B) A representation of activated microglial cells double labeled with a red immunofluorescent label specific to Iba1 (whole cells) and a green immunofluorescent label specific to CD68 (small circular areas).

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Mary Harrington, Psychology

References:
Remarkable studies, such as the “thirty-million word-gap,” show that children’s later language and literacy development have been based on the quantity of the mothers’ language. However, recent studies suggest that the quality of the mother’s input and how rich the conversation is between the mother and child is more important than simply the amount of language spoken. The main purpose of this study examines mainstream English dialect (AAE) to young preschoolers on later literacy development. There were 60 African American mother-preschooler pairs from low-income communities who were in a larger longitudinal project studying the impact of curricular interventions on school readiness. The children varied in age from 3;10 to 5;0 (mean = 4;6) at the beginning of the study. The mothers and children were videotaped in a free play situation with a Fisher-Price castle, blocks, and playdough. Ten-minute samples of the caregiver’s language to the child were transcribed using FileMaker. The transcriptions were then analyzed for syntactic variation using the IPSyn Sentence Structure scale (Scarborough, 1990) and for depth of AAE dialect. A number of language measures were available on the children at the beginning of the preschool year, including: score on the dialect-neutral (risk) items of the DELV-ST screening test, expressive vocabulary (EOWPVT-R), and phonological awareness on the blending and elision subtests of the Pre-CTOPP. Reading scores on the Passage Comprehension subtests of the Woodcock-Johnson III were taken at the end of first grade, when the children’s average age was 7:1. The amount of the mothers’ talk was predicted by education level and the use of AAE dialect features. However, there was no relationship between the mothers’ IPSyn scores and either their use of AAE or their education levels. The amount of talk and the IPSyn scores were significantly related. (See Table 1). Linear hierarchical regression analysis revealed that the mothers’ IPSyn Sentence Structure scores on their utterances to the children were an independent positive predictor of the children’s reading comprehension scores at the end of first grade even when all of the other factors were controlled. Neither mothers’ depth of AAE dialect nor number of utterances was a significant independent predictor of the children’s later reading scores. This study confirms that in low-income African American mother-child pairs quality rather than quantity of the input of language matters most for later literacy development of children.

(Supported by the Mary Sweig Wilson Undergraduate Research Fellowship)

Advisor: Peter de Villiers, Psychology

References:


Affective disorders, such as depression, increase the risk of cardiac mortality, an association observed in patients with and without a history of cardiac disease. The mechanism by which depression impacts cardiac health is not exhaustively understood, but research with animal models has established that affective disorders are correlated with increased resting heart rate and reduced heart rate variation, characteristics also associated with cardiac mortality. This mechanism is indicative of autonomic nervous system (ANS) dysfunction and of abnormal vagal regulation of heart rate variability via the ANS. A connection between the ANS and cardiac mortality is well established in humans. This SURF research investigates the autonomic regulation of cardiac function in the socially monogamous prairie vole and promiscuous meadow vole species. The first project, led by Masters Student, Jennifer Christensen, characterizes the cardiac function of the female meadow vole surgically implanted with Data Science International (DSI) 10ETA-F20 radiotelemetry devices in the abdominal cavity. An undergraduate-led project investigates the effects of social isolation on the cardiac function of surgically implanted animals and on depression-relevant behavior of a separate cohort of animals. The aim of this project is to consider an autonomic/cardiac model using meadow voles, similar to the established paradigm in prairie voles. Work completed this summer includes contribution to Jennifer’s work, project development and proposal of an independent research project, and initial data collection from socially isolated voles with implanted transmitters. All projects are currently in the data collection stage.

Electrocardiogram analysis module from Data Science International (DSI). Ponemah software (DSI, New Brighton, MN) is used for data analysis to calculate average heart rate, heart rate variability, and other cardiac parameters in this research.
(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Annaliese Beery, Psychology

References:


Analyzing Shooter Bias Studies Using Signal Detection Theory

Ray Silveria/2016

Signal Detection Theory (SDT) is an analytic technique that is often used within Shooter Bias studies due to its ability to measure quick, implicit decisions. Shooter Bias refers to the bias that people will shoot unarmed Black men more than unarmed White men and is measured using a shooter video game task. This task involved showing a series of photos of Black and White men and participants were told to press a certain key indicating the decision ‘shoot’ when a person holding a gun and to press indicating ‘don’t shoot’ if a person was not holding a gun. Each photo was shown for less than a second so these decisions were quick and helped to show implicit biases.

My research this summer included literature review on Shooter Bias studies and learning about SDT and how to implement this technique with shooter bias data done by Smith students. I gathered data from three different shooter bias studies that were conducted within the last two years and I reanalyzed their data using SDT to replicate their findings. SDT measures the degree of which a participant, within a shooter task, is able to differentiate armed targets from unarmed targets, which is also known as the d' statistic. SDT also measures the degree to which participants favor a ‘shoot’ response over a ‘don’t shoot’ response, which is known as the c' statistic.

Using SPSS I calculated the d and c statistic by first calculating Hit Rates and False Alarm Rates. Hit Rate is the probability of correctly shooting at an armed target and a False Alarm Rate is the probability of participants shooting at unarmed targets. These rates were calculated by creating proportions variables for both Black and White Targets (men). The results I compiled compared with previous analysis with few differences. SDT is now a new analytical method for future research students within Professor Blanchard’s lab and a possible new topic in his course, Research Design and Analysis.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Fletcher Blanchard, Psychology

References:


The Effects of Ethanol Exposure on the Circadian Rhythm of Zebrafish Embryos

Margaret Anne Smith/2016

In October 2015 the American Academy of Pediatrics called for mothers to forego drinking any amount of alcohol during pregnancy.¹ When a pregnant mother consumes alcohol she can cause developmental changes in her offspring. These changes affect a variety of cells and organ systems throughout the human body. These effects can be short-lived, long-lasting, or permanent and become active within a short time or in later years. The negative effects of alcohol (ethanol) can be implicated in a variety of deleterious changes, including: in brain development and function, cancer development and progression, fetal alcohol spectrum disorders, immune system dysfunction, liver disease, gastrointestinal tract disorders, and circadian rhythm disruption. Circadian rhythms allow for the coordinated regulation of a wide-range of biological processes from individual cells up through whole organisms which synchronize their internal clocks to a 24-hour day. Because the disruption of the circadian system has been linked to a wide-range of animal and human disorders, the focus of my initial research was to investigate the effects of ethanol exposure on the circadian rhythms of zebrafish. The zebrafish model was chosen because of its quick development cycle and transparency. This project had its beginnings in Dr. Harrington’s Experimental Methods in Neuroscience course (NSC 230) in the spring 2015 semester. At the end of each trial we determined whether there were significant rhythmicity alterations in the zebrafish exposed to ethanol.

Based on the preliminary results during the spring of 2015, additional data was collected the following summer. Based on those results, I continued my work under the supervision of Dr. Harrington in a Special Studies Project during the fall 2015 semester.

The accrued results through the end of 2015 indicated that ethanol exposure increased the likelihood of significant rhythmicity alterations in the zebrafish embryos. Although the data appeared to have this trend, further trials were necessary to confirm the consistency of these results. Therefore, Dr. Harrington suggested that I complete the project within the 2016 Surf Program. The results conducted this summer confirm these findings. Based on the results of the study to date, I have decided to expand this research to shed light on additional factors and conditions associated with alcohol exposure and alterations in circadian rhythmicity. New avenues of research will include: the effect of alcohol on miRNAs, the role complex enzymes play during fetal development and the potential deployment of Nutraceuticals along with or shortly after the exposure of the zebrafish to ethanol to test for the possibility of ameliorating the negative effects of ethanol exposure. These and additional expansions of this ongoing study could lead to the identification of the underlying mechanisms and processes that are needed to facilitate breakthroughs in the recognition and treatment of alcohol-related disorders.

The goal of future studies will be to contribute information and data to more effectively address, treat or alleviate the wide range of alcohol-related disorders that infants develop prenatally.

(Supported by the Howard Hughes Medical Institute)

Advisor: Mary Harrington, Psychology

Reference:
Many psychological disorders and symptoms have been empirically linked to trauma history. On this basis, researchers advocate incorporating trauma into the standard curriculum for training and educating professional psychologists. We wanted to investigate whether current doctoral training is in line with these recommendations. Our SURF research was therefore aimed at understanding how future clinical practitioners are being trained to think about psychological trauma and its link to adult psychopathology.

We reviewed empirical articles assigned to doctoral students attending both clinical and counseling psychology programs accredited by the American Psychological Association (APA). Using public information from each program’s webpage, we contacted 116 doctoral programs requesting the most recent syllabi from professors teaching Adult Psychopathology courses. From the syllabi, we obtained supplementary articles assigned during weeks focusing on trauma and stressor related disorders. We categorized the articles in various ways to summarize their content. Furthermore, in order to gain a deeper understanding of what students learned from the courses, we also generated two surveys that will soon be administered to both professors and their students. The survey that will be distributed to professors will help us understand what they aimed to teach their students about trauma and psychopathology. The survey that will be administered to students will give us more information about what they took away from the class regarding the relationship between trauma and psychopathology.

After reviewing 21 syllabi, we found that the majority (71.4%) of courses assigned the Diagnostic and Statistical Manual for Mental Disorders (DSM) as a required reading. This implies that students were taught the American Psychiatric Association diagnostic criteria for trauma-related disorders. In addition, we noted that the two most popular textbooks were those written by Craighead et al. (2013) and Castonguay & Oltmanns (2013). Our review of the 57 supplemental readings showed that 22.8% focused on childhood maltreatment, 10.5% focused on somatization, 24.6% focused on dissociation, and 10.5% highlighted controversies in the trauma field.

In order to achieve our goal of investigating psychological trauma and how its link to adult psychopathology is taught to future clinical practitioners, more work needs to be done which will be conducted next semester.
(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Nnamdi Pole, Psychology

References:


Acquiring the Difference Between Absolute and Relative Gradable Adjectives: 4-Year olds Know Emotion Adjectives are Relative

Victoria Torres/2017

Gradable adjectives, also known as scalar, dimensional or spatial adjectives, are adjectives which describe properties of objects that hold to different degrees such as length, height, or intensity of emotion. Gradable adjectives can linguistically be classified into two types (absolute or relative) depending on the contexts of their use. Relative gradable adjectives like big, long, etc. are context dependent—what is “long” in one comparison set may change in another. Absolute gradable adjectives are not context dependent, however. An example of an absolute adjective is something like “striped” or “spotted”—your criterion for including something “striped” or “spotted” doesn't change depending on the context.

Previous research has demonstrated that children 3-5 are sensitive to the distinction between absolute and relative adjectives when using physical properties. However, no prior research has investigated whether children view gradable emotion adjectives as relative or absolute.

Graded picture sequences of people expressing base emotions (happy, sad, and angry) were created by morphing a neutral facial expression with an extreme facial expression. A group of adults rated each picture with the intensity of emotion, creating relatively linear 9-picture scales for each emotion. In a sorting experiment, 32 adults showed that they viewed the emotion adjectives as relative, because they changed their criterion of inclusion of that emotion adjective when more intense pictures of the emotion were added to the comparison set. However, adults viewed “striped” (umbrellas) and “spotted” (dogs) as absolute, as their criterion for inclusion didn't change when more striped or spotted items were added to the comparison set.

The same sorting game was played with 26 children. Experimenters laid out all cards for an emotion or physical property (either 9 or 13 depending on the condition, and orders/sequences varied across participants) and asked children to “Put all the X (happy, sad, angry faces/long pencils/striped or spotted items) into the box.”

Table 1 shows that children regarded emotion adjectives and long pencils as relative, while they regarded spotted dogs and striped umbrellas as absolute, and that the distinction between the two is clear.

Future research should investigate whether younger children, as well as children with autism, view emotion adjectives as relative and when that distinction is made.

Table 1:

<table>
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<tr>
<th>Items (Pictures or Objects)</th>
<th>Mean Shift (In # of Items)</th>
<th>F (1, 24)</th>
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<td><strong>ABSOLUTE ADJECTIVES</strong></td>
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<tr>
<td>Striped Umbrellas</td>
<td>-0.08</td>
<td>1.175</td>
<td>.289</td>
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<td><strong>RELATIVE ADJECTIVES</strong></td>
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<tr>
<td>Long Pencils</td>
<td>+1.11</td>
<td>5.285</td>
<td>.031*</td>
</tr>
<tr>
<td>Happy Man</td>
<td>+2.56</td>
<td>11.310</td>
<td>.004**</td>
</tr>
<tr>
<td>Happy Woman</td>
<td>+1.55</td>
<td>5.583</td>
<td>.027*</td>
</tr>
<tr>
<td>Sad Man</td>
<td>+1.25</td>
<td>4.260</td>
<td>.048*</td>
</tr>
<tr>
<td>Sad Woman</td>
<td>+1.61</td>
<td>4.659</td>
<td>.041*</td>
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<tr>
<td>Angry Man</td>
<td>+1.65</td>
<td>13.128</td>
<td>.001**</td>
</tr>
<tr>
<td>Angry Woman</td>
<td>+2.29</td>
<td>6.835</td>
<td>.015*</td>
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</table>

+ = shift towards more extended or more intense end of the dimension.
- = shift towards less extended or less intense end of the dimension.

(Supported by the Mary Sweig Wilson Undergraduate Research Fellows)

Advisor: Peter de Villiers, Psychology


The Effect of Circadian Disruptions in Fat Tissues

Ivana William/2018

The experiment we carried out during SURF was based on the results we obtained from a previous study that was carried out. This study which looked into the effect of circadian disruption (CD) on metabolic tissue in female mice, found no significant difference in the size of white adipose tissues (WAT) in the circadian disrupted group. We concluded that this was likely due to the effect of hormones such as estrogen which has an inhibitory action on metabolic syndrome which is caused by circadian disruption. Therefore, using these results, we carried out a new experiment where we looked into the joint effects of circadian disruption and high fat diet on male and female mice.

Our experiment included two groups of seven mice each, both on HFD. One group was under a normal light/dark cycle (12h light/12h dark, T24) while the other group was not (10h light/10h dark, T20). The two groups consisted of both males and females. The experiment was carried out for ten weeks. Weights were recorded weekly. Glucose tolerance tests were carried out and blood was collected during dissection to further carry out insulin tests. During dissection, heart, soleus, liver, pancreas, white adipose tissue, brown adipose tissue, blood, and suprachiasmatic nucleus samples were collected for further analysis.

The initial findings of the experiment showed that males in T20 and T24 have a higher percent weight gain than both categories of females. While females under T20 have a higher percent weight gain than females under T24. We concluded that there is a difference in weight gain due to sex. There is also a larger effect of T20 on weight gain in females versus T24, which is not the case in males. Future research involves analyzing the data collecting from the Glucose tolerance test.

(Supported by the Frances Baker Holmes Internship Fund)

Advisor: Mary Harrington, Psychology

Reference:

Historically, the fields of Human Computer Interaction (HCI) and Human Services have been disparate studies. Professor Crouser collaborated with mental health clinicians at the Justice Resource Institute (JRI) in order to integrate technology to optimize the works of JRI. One of the problems identified by the clinicians was a diagnostic bias stemming from a clinician's specialization in a field. To minimize this bias, my fellow lab members and I worked to create a tool that would suggest to the clinician alternative diagnoses via utilization of an interactive decision tree.

I created a Python program that uses BeautifulSoup, an open-source package used to parse HTML and XML documents, to extract information on each of the diagnoses from the online version of the Diagnostic and Statistical Manual of Mental Disorders (DSM-V). My fellow lab members and I manually grouped the data into new categories created by the clinicians. We created R and Python programs to wrangle the data and convert categorical data into quantitative terms.

I then created a decision tree of the data in Python using scikit-learn, an open-source package for data mining and analysis, and Graphviz, an open-source software for graph visualization (see Figure 1). I also conducted machine learning analysis using the random forest module available in scikit-learn, matplotlib (a Python plotting library), and Seaborn (a Python statistical data visualization library). The random forests helped determine whether the splits of the branches made by the decision tree were biased. The heat maps highlight significant patterns of the position and frequency within each category with respect to varying numbers of trees and features (see Figure 2). The decision tree was formatted into JSON which my fellow lab members used to create the front-end, interactive visualization of the decision tree.

Through our work, we learned manipulation of categorical data is a difficult process for two main reasons. First off, the DSM-V has many inconsistencies in its methods of defining and coding diagnoses. These discrepancies make it challenging to convert categorical to consistent quantitative data. In addition, situational factors such as the patient's background and the phrasing of the symptoms exert significant leverage on how clinicians categorize certain symptoms. This further solidified the need to classify the DSM-V to increase objectivity and decrease specialization bias. This research could improve objective, generalizable applicability by gathering data from various clinical institutes through crowd-sourcing.

Figure 1: Sample of Back-End Decision Tree

Figure 2: Heat Map of 100 trees of frequency of each category by position

(Supported by the Schultz Foundation)

Advisor: R. Jordan Crouser, Statistical and Data Sciences
When clinicians meet with mental health patients, they often determine a diagnosis for the patient based on personal knowledge and experience before checking the Diagnostic and Statistical Manual of Mental Disorders (a manual that lists all the mental health diagnoses and symptoms associated with them each) to verify that nothing disproves their original hypothesis. However, this leaves a large room for error and personal bias, which can lead to an incorrect diagnosis. This project aims to create a tool that applies a more scientific approach to diagnosing mental health patients using the symptoms outlined in the DSM-5, as well as implement a visualization of this process to aid clinicians.

While my research partners worked on scraping the DSM-5 for all the diagnoses and their corresponding symptoms and worked with a group of mental health clinicians to categorize all of the long-winded symptoms into clear cut groups, I worked on the front end of the application. This involved using the data my partners scraped and cleaned to create and traverse a decision tree that could ask simple questions, such as “Does the patient experience problems with activities of daily life?” which require a yes or no answer. As the clinician answers the questions, the application narrows down the possible diagnoses for the patients. To take our application one step further, I also designed a data visualization that would bring the data to life for the clinicians. Currently, the data visualization using a subset of the data can be seen in Figure 1. All the possibilities start in the middle column, and as symptoms are verified or falsified, the symptoms are bolded or crossed out accordingly. Furthermore, diagnoses that have a verified symptom, and thus are more likely to be correct, move to the left column, while the diagnoses that include a symptom that the patient does not have move to the right column. The left column containing the somewhat verified diagnoses is further organized by likelihood in descending order (based on how many symptoms have been verified by the clinician). Finally, when a certain diagnosis is clicked on, the user is brought to the page in the DSM-5 in which this diagnosis can be found.

Figure 1: An example of the project visualization.

This project has already begun to combine the fields of psychological mental health and data science into a productive application. In the future, we plan to improve the model by implementing it with the entire list of diagnoses, which likely will involve tweaking the data visualization technique used.

(Supported by the Department of Education)

Advisor: Jordan Crouser, Statistical and Data Sciences

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