20th Annual
Summer
Undergraduate
Research
Fest
Hosted by the College of Natural and Mathematical Sciences
Wednesday, August 9, 2017

STEM BUILD  STEM BUILD at UMBC—NIH/National Institute of General Medical Sciences
HHMI Scholars  Howard Hughes Medical Institute
HPC REU  Interdisciplinary Program in High Performance Computing—A National Science Foundation’s Research Experiences for Undergraduates Site
MARC U*STAR  Maximizing Access to Research Careers—Undergraduate Student Training in Academic Research Program—NIH/National Institute of General Medical Sciences
NSA Scholar  National Security Agency Scholar
NSF REM  National Science Foundation Research Experience and Mentoring
NSF REU  National Science Foundation Research Experience for Undergraduates Program in Chemical Sensing and Imaging at UMBC
SBTP  Summer Biomedical Training Program — CNMS and the UMBC Graduate School
SCI ART  Interface between Science and Art in the research laboratories of leading scientists, engineers and art conservators at the University of Maryland Baltimore County (UMBC), Johns Hopkins University, and the Walters Art Museum in Baltimore.
Wednesday, August 9, 2017

All events will be held in the Ballroom, University Center, 3rd Floor

8:30 am: Poster Set-up & Presenter Check-In Begins

Ballroom Lobby, University Center, 3rd Floor

Light Breakfast served

9:00 am – 10:30 am: Introductions & Oral Presentations

Bacteriophage in Freshwater Environments
Carl Bannerman, Kaaliyah McDowell, and Jonathan Poole
STEM BUILD

Detecting Bovine Lameness Using Spline Transformations of Three-Dimensional Limb Movement Variables in a Logistic Regression Model
William Dula and Ari Goldbloom-Helzner
HPC REU

Optimization of Gold Nanoparticles to be Used as a Multifunctional Drug Delivery System for Chemotherapy
Angelie Matar
NSF REU

A Computational Framework to Reverse-Engineer Intratumor Sub-Clonal Dynamic Models and Predict Optimal Treatment Targets
Caroline Larkin
MARC U*STAR, Meyerhoff Scholars

Structure and Function of the HIV-1 RNA Genome
Amara Ejikemeuwa and Brea Manuel
SBTP Trainees

10:30 am – 12:00 pm: Poster Presentations

Ballroom, University Center, 3rd Floor

10:30 am – 11:15 am – Poster Session 1
11:15 am – 12:00 noon – Poster Session 2

12:00 pm – 12:30 pm: Mentor Recognition and Closing

We request that there be no flash photography during the event to ensure the safety of all on stage.
Welcome to the 2017 UMBC Summer Undergraduate Research Fest, which is hosted annually by the College of Natural and Mathematical Sciences. This year marks the twentieth anniversary of the event and it is inspiring to see so many participants continue the tradition of students formally presenting the results of their summer research projects.

While some projects are the result of independent arrangements, many have been made possible by grants or other funds dedicated to encouraging undergraduate research. All projects are associated with UMBC and require the support of research mentors. The mentors’ passion for science and ongoing commitment to education continue to be exceptional. I commend the students on their extraordinary efforts this summer, and thank each of the faculty, graduate student, and peer mentors who have worked closely with them.

I also want to voice my appreciation of all the staff members who have supported this event over its 20-year history. I especially want to thank the CNMS staff members who are supporting this year’s event - Rebecca Dongarra, Data and Events Coordinator; Tim Ford, Manager of Research Graphics; Caitlin Kowalewski, Research Coordinator; Justine Johnson, Associate Director of the Meyerhoff Graduate Fellows Program; and Kathy Sutphin, Assistant Dean for Academic Affairs.

Congratulations and best wishes for a successful event,

William R. LaCourse, Ph.D.
Dean and Professor of Chemistry
College of Natural and Mathematical Sciences
University of Maryland, Baltimore County
20th Annual UMBC Summer Undergraduate Research Fest

Oral Presentations

Bacteriophage in Freshwater Environments
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Program Acronyms

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Viruses that infect bacteria, called bacteriophage, are the most abundant organisms on the planet. Bacteriophages are present wherever there are host bacteria, including aquatic environments. Although a number of studies have examined bacteriophages from marine waters, research on freshwater is more rare. The present study aimed to fill the gaps in our knowledge of freshwater microbial communities. We surveyed viral diversity from four locations on the University of Maryland, Baltimore County (UMBC) campus. Bacteriophages were identified to the family level by morphology.

Water samples were collected from the PigPen Pond, a nearby swamp, the Albin O. Kuhn (AOK) Library Pond and a north campus pond. Samples were purified and the filtrate was concentrated. Samples were negatively stained and imaged using a transmission electron microscope (TEM). Images were analyzed using IMAGEJ software for the following measurements (nm): capsid diameter, tail length and width, base plate length and width, and total body length.

We found representative species from all bacteriophage families known to inhabit aquatic environments. Across samples, Podoviridae was the most abundant, followed by Siphoviridae, Myoviridae, Tectiviridae and/or Corticoviridae (tailless bacteriophages with capsid spikes), and Microviridae. Between sample locations, we observed that the AOK Library Pond had the highest viral concentration and the most diversity. Unexpectedly, Myoviridae was only found in half the locations sampled, unlike other studies where they are typically the most abundant species. However, we suspect this may be a quality issue as we found evidence of Myoviridae broken tails throughout samples. Our observation of Tectiviridae/Corticoviridae in freshwater was a surprising result as they have previously only been recorded in marine waters and sewage. This suggests that local freshwater sources represent an untapped reservoir of bacteriophage diversity that can potentially be used to treat bacterial infections in the medical field or environmental control of contaminated water.

This work was supported by the STEM BUILD at UMBC initiative through the National Institute of General Medical Sciences (NIH Grants 8TL4GM118989, 8UL1GM118988, and 8RL5GM118987).
DETECTING BOVINE LAMENESS USING SPLINE TRANSFORMATIONS OF THREE-DIMENSIONAL LIMB MOVEMENT VARIABLES IN A LOGISTIC REGRESSION MODEL

REU Site: Interdisciplinary Program in High Performance Computing

William Dula¹, Jason Glover², Ari Goldbloom-Helzner³, Kayla Makela⁴,  
Graduate assistants: Qing Ji and Sai Kumar Popuri²  
Faculty mentors: Nagaraj Neerchal² and Andrew Raim⁶  
Client: Uri Tasch⁵

¹Department of Mathematics, Morehouse College  
²Department of Mathematics and Statistics, UMBC  
³Division of Applied Mathematics, Brown University  
⁴Department of Mathematics and Statistics, Michigan State University  
⁵Department of Mechanical Engineering, UMBC  
⁶United States Census Bureau

Bovine lameness is a common issue on commercial dairy farms, resulting in decreased productivity. Early detection is essential for effective treatment of lameness. Here and recent previous work, lameness status of a cow has been modeled using three-dimensional limb movement measurements related to its gait. A statistical model uses the software SAS® with its LOGISTIC and TRANSREG procedures. The model produces a binary classification, lame or not lame. Current implementation requires running several SAS® procedures manually and therefore is not amenable to a large scale application and model optimization. In this work, we implement an optimization algorithm in R to mirror the TRANSREG procedure, and thus speed up exploration of a large number of candidate models to optimize the widely used goodness of fit criteria such as the Receiver Operating Characteristic curve (AUC). The models are also evaluated using classification error rates (False Positives and False Negative or equivalently sensitivity and specificity). We also consider multinomial logistic models so that cows may be further classified into three categories: severely lame, mildly lame or sound. These results can be used in the commercial dairy industry for lameness detection.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-author Jason Glover was supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistants: Qing Ji and Sai Kumar Popuri were supported by UMBC.
OPTIMIZATION OF GOLD NANOPARTICLES TO BE USED AS A MULTIFUNCTIONAL DRUG DELIVERY SYSTEM FOR CHEMOTHERAPY

Angelie Matar¹, Arunendra Saha-Ray², Dr. Marie-Christine Daniel²
¹Department of Chemistry, Louisiana State University, 232 Choppin Hall, Baton Rouge, LA 70803
²Department of Chemistry and Biochemistry, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

The development of a gold nanoparticle (GNP) cored dendron as a drug deliver system can help people advance towards a better treatment of chemotherapy. The problem with the current application of the medicine is that it affects the whole human body, not just the tumor area. With the information collected in this research, we aim to eliminate the preexisting problem of excessive drug toxicity in the body and enhance targeting efficiency. By using these nanoparticles, we have found a way to get the drug into the body with enhanced permeability and retention (EPR) effect which provides passive targeting. Furthermore, through active targeting, the nanocarriers are able to attach to the cell and release the drug into the nucleus.

The design of this drug is a GNP core coupled with spacers and dendrons around the surface that have multifunctional uses on their termini. The dendron is a first generation poly(propylene imine) (PPI) dendron with a phenolic focal point, and the spacer is a tetraethylene glycol (TEG) molecule that is connected at that focal point of the PPI dendron. With this model, we have multiple branches around the core that are made in a very limited amount of steps, and we can organize the distribution of different functions on the molecule connected to each branch. This makes the targeting to the tumor more efficient, makes multimodal imaging easier, and increases the amount of drug molecules attached.

In this study, we have functionalized the first generation dendron with near infrared fluorescence marker. We have also attached Doxorubicin to a third generation dendron. With this ideal drug, we are able to have a multifunctional drug delivery system that releases into the body with decreased drug toxicity. Future studies include being able to attach molecules to all eight branches in the third generation PPI.

This research project is supported by the National Science Foundation Research Experience for Undergraduates (REU) Research award CHE-1460653 and National Science Foundation Research award CHE-1507462.
Cancer is a disease characterized by a variety of sub-clonal tumor cells dynamically interacting among themselves and the surrounding environment. This results in heterogeneous cancer phenotypes that can have increased tumor volumes and growth depending on their sub-clonal compositions. Sub-clonal tumor heterogeneity is a poorly understood aspect of cancer, which can grant new insights for the development of cancer therapeutics. It has been shown that different sub-clones have varying impacts on overall tumor growth and their interactions are essential to maintaining the proliferation of cancer cells. Perturbing certain sub-clones can significantly affect the development of the tumor, with results ranging from decreasing tumor volume to a massive rise in tumor growth rates. To better understand the dynamics of sub-clonal interactions and predict the effects of targeted therapeutic interventions, we developed a computational framework to construct non-spatial, dynamic mathematical models of tumor heterogeneity. Our method uses high-performance computing to automatically infer models from the data, simulate them through time, and evaluate the in silico results in comparison to the results obtained from the experiments at the bench. To test this approach, we developed three mathematical sub-clonal interaction equations and inferred complete models that can accurately recapitulate tumor volume and clonal frequency data from mice xenograft experiments with human cancer cells. Importantly, the reverse-engineered models can predict the results of novel experiments and perturbations, and hence determine the optimal clone or clones to target for therapeutic intervention to make the tumor stabilize or even collapse. This project will provide significant insight into the underlying complexity of tumor sub-clonal dynamics. Understanding the dynamics of tumor heterogeneity will provide essential information for cancer drug development and the potential for individualized medical treatments based on a patient’s unique tumor composition.

Acknowledgements: We thank all the members of the Lobo Lab for the support and advice. This project was funded in part by a grant from the PhRMA Foundation. We acknowledge the UMBC Meyerhoff Scholars Program and MARC U*STAR program for their support. Computations used the UMBC High Performance Computing Facility (HPCF) supported by the NSF MRI program (grants CNS-0821258 and CNS-1228778), the SCREMS program (grant DMS-0821311), and the University of Maryland, Baltimore County (UMBC).
The human immunodeficiency virus (HIV-1) infects CD4 cells, and causes the acquired immunodeficiency syndrome (AIDS). The 5′ leader of the RNA genome exists between monomer and dimer conformations. These conformations dictate the late phase of viral replication. The monomer conformation promotes translation, while the dimer RNA is selectively packaged into new virus particles. It was recently found that multiple viral RNA genomes exist in the cell differing by only one to two guanines on the 5′ end. This small difference significantly affects the monomer-dimer equilibrium. RNAs beginning with one guanine plus the native cap (Cap-1G) favors the dimer conformation, while RNAs beginning with two or three guanines plus the native cap (Cap-2G and Cap-3G) favor the monomeric conformation.

Previous studies found that the addition of a guanine to the dimer 5′L (Cap-2G) disrupts the polyA hairpin. This disruption causes the DIS to become sequestered, as polyA residues strengthen U5:DIS interactions, resulting in the monomer conformation. When exposed, the DIS contains a palindromic sequence for DIS-DIS base-pairing, causing dimerization. This results in more NC binding sites in the dimer conformation. We seek to determine the structure of the Cap-1G dimer conformation and characterize the Gag-RNA interactions essential to genome packaging.

We confirmed multiple structural elements in the full dimeric 5′L and hypothesize that the cap is involved in a unique coaxial stacking interacting with the adjacent hairpin. We are in the process of determining the specific NC binding sites for this dimeric 5′L. Our data shows that binding of Gag protein to these binding sites will promote the formation of the Gag hexamer, which potentially functions as the nucleation site for viral assembly. Ultimately, we want to solve the complex structure between Gag and the viral RNA, which will provide the detailed molecular mechanism for HIV-1 selective genome packaging.

This research was funded by NIH/NIGMS grants 1P50GM103297 and the Howard Hughes Medical Institute at UMBC, with support by the NIH-funded MARC U STAR program at UMBC under National Research Service Award T34 GM 008663.
POSTER SESSION

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STRUCTURAL ELUCIDATION OF THE HIV-1 REV RESPONSE ELEMENT

Sophia Abbott¹, Stanley Wang¹, Daniel Morris¹, Colin O’Hern¹, Julia Zaki², Dr. Jan Marchant¹, 
Dr. Michael F. Summers¹

¹Howard Hughes Medical Institute, Department of Chemistry and Biochemistry, University of 
Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD, 21250 
²Department of Undergraduate Studies, University of Maryland, College Park, 2110 Marie 
Mount Hall, College Park, MD 20740

Successful proliferation of HIV requires the reliable export of singly spliced and unspliced 
viral transcripts to create new pathogenic virions. The export of singly spliced and unspliced 
transcripts from the host nucleus is mediated by the protein-RNA Rev-RRE complex. Binding of 
HIV Rev proteins to the Rev-response element (RRE) in the HIV genome permits nuclear export 
of the complex with support of host cell proteins. In the absence of Rev binding, HIV-1 genomes 
are retained in the nucleus, preventing the complete assembly of new virions. 

The goal of this investigation is to elucidate a high-resolution structure for the RRE region 
of the HIV genome through NMR spectroscopy. Spectra from individual stem fragments, designed 
from computationally derived predictions of the RRE secondary structure, were compared to that 
of the complete RRE. This allowed for the assignment of outlying chemical signals, revealing 
integral characteristics of the RRE’s secondary structure, including two competing four and five 
stem loop conformations of the RRE.

Due to the presence of substantial signal overlap in the 2D NMR spectra of the full RRE, 
chemical shift assignments remained difficult. Utilizing deuteration and segmental labelling, the 
quantity of cross peaks can be reduced to alleviate signal crowding and better identify specific 
regions of the RRE. Targeted mutations to stabilize and select for the 4 stem and 5 stem 
conformations have also been made to improve current structure data from cryo-EM and NMR 
spectra.

The structure will reveal insights into the RRE’s ability to recruit multiple Rev proteins 
and form the export complex. Disruption of this process creates a promising target for future 
therapy developments.

We would like to acknowledge the Howard Hughes Medical Institute and National Institutes of 
Health for Grant #1P50 GM103297. We would also like to thank Dr. Michael Summers and Dr. 
Jan Marchant for their incredible mentorship.
IDENTIFYING NEW TARGETS OF THE LYSINE METHYLTRANSFERASE SET5 AND DETERMINING THE EFFECT OF PHOSPHORYLATION ON ITS METHYLATION ACTIVITY

Assefa Akinwole1, Sylvia Min1, Marlene Keisha Kontcho1, Rashi Turniansky1, James Moresco2, Julie Wolf1, John Yates III2 and Erin M. Green1

1Department of Biological Sciences, UMBC, Baltimore, MD 21230
2Scripps Research Institute, Department of Chemical Physiology, La Jolla, CA 92307
3Howard Hughes Medical Institute, Chevy Chase, MD 20815

Histones are a group of proteins that are associated with DNA, and they serve an important function to organize and regulate the accessibility of DNA. Histones play a key role during transcription and the post-translational modification of these histones regulates gene expression, allows for responses to environmental stresses, and promotes silencing of genomic regions that should not be expressed. Enzymes post-translationally modify histones, as well as other proteins, with chemical groups such as a in methylation, phosphorylation and acetylation. One such enzyme is Set5, the first discovered H4 methyltransferase in budding yeast that monomethylates lysines 5, 8, and 12. Set5 plays a role in regulating cell growth and stress responses, as well as promoting repression of genes at telomeres in conjunction with Set1, another methyltransferase. In order to understand more about Set5 function, we performed an immunoprecipitation of Set5 coupled to mass spectrometry. This allowed us to identify potential protein interacting partners, as well as post-translational modifications to Set5 itself. We determined that Set5 contains various phosphorylation sites which are likely key in its function within the cell. Using \textit{in vitro} methylation assays with versions of Set5 carrying mutations in the phosphorylation sites, we determined that these phosphorylation sites may affect the methylation by Set5 on histone H4. Additionally, we identified other interacting partners of Set5 that may be methylated by Set5, including Ssa1, Hxt6, Hem14, and Cos6. We will discuss our strategy for verifying whether these are true targets of Set5’s methyltransferase activity.

This research was supported in part by a grant to UMBC from the Howard Hughes Medical Institute through the Precollege and Undergraduate Science Education Program.
PROPERTIES OF THE 20 LARGEST SOLAR ENERGETIC PARTICLE EVENTS FROM THE RADIATION IMPACT PERSPECTIVE

Mary Aronne¹, Yihua Zheng²
¹Department of Geography and Environmental Systems, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
²Heliophysics Science Division, NASA Goddard Space Flight Center, 8800 Greenbelt Road, Greenbelt, MD 20771

Solar energetic particle (SEP) events are caused by solar flares and coronal mass ejections (CMEs) from the Sun. The term SEP event is typically referencing protons that are accelerated at relativistic speeds along magnetic field lines extending from the Sun into the solar system. SEP events can lead to disruption of satellites, communications blackouts, and radiation exposure for humans at high latitudes and in space. Historic SEP events have been detected by the Solar Maximum Mission (SMM), the Solar and Heliospheric Observatory (SOHO), and the Geostationary Operational Environmental Satellite (GOES).

The 20 largest SEP events from the radiation impact perspective from 1981 to 2006 were examined to discern commonalities such as source location, CME speed, and flare intensity. These events all had a 20 day period of very high SEP fluences. Many of these 20 main events had several solar flares, CMEs, and smaller SEP events associated with them.

Most of the events studied have a solar flare and CME origin in the southwest quadrant of the Sun. The CME velocity for each event was also recorded from various sources and compared to the other CME speeds based on longitude from east to west. The speeds varied from between the 300-500 km/s range all the way into the 2000 km/s range. Most of the events occur during the periods of solar maximum. Energy spectra properties were also studied to demonstrate the different acceleration mechanisms of these high energy particles. The aim of this project is to determine ways to better forecast SEP events based on their statistical properties.
A NOVEL REPORTER SYSTEM TO QUANTIFY MISSENSE ERRORS DURING TRANSLATION IN *SACHHAROMYCES CEREVISIAE*

Zain Abidin, Kartikeya Joshi and Philip J. Farabaugh
Department of Biological Sciences, University of Maryland, Baltimore County, MD-21250

Translation is the final step in central dogma, in which information is transferred from DNA to mRNA and finally to proteins. The key players during translation include mRNA, tRNA and ribosomes. The ribosomes have to maintain a fine balance between the rate of protein synthesis and the accuracy with which the proteins are synthesized. Even though the process is highly accurate, errors can occur. A common error that occurs during translation is when an incorrect tRNA comes and binds to the ribosome and adds an incorrect amino acid in the growing polypeptide chain, such an error is called a missense error. We have developed a reporter-based system that can be used to quantify missense errors made by a single tRNA in yeast. Such a system can be further employed to understand the mechanism and processes that regulate errors during translation. Errors made during protein synthesis, can lead to a non-functional or inactive protein and accumulation of such bad proteins, can be deleterious to cells, such as in the patients suffering from Alzheimers or Parkinsons disease. We have used this reporter system to quantify errors in yeast and found out that the eukaryotic ribosome is very efficient in discriminating against most incorrect errors, with accuracy as high as one in a million errors. However, certain codons can be misread by the ribosome with a 100-fold higher error rate than the background errors. These include Glycine codons, GGA and GGG and the Aspartic acid codons, GAU and GAC. My work is trying to establish whether the activity of the two Aspartic acid codons is due to misreading or functional replacement. I will be using hyperaccurate and error prone mutants of the selected gene to characterize this role.
Ivory is a material obtained from the tooth or tusk of an animal. It has been used for centuries for a wide range of applications including medicines, weapons, and art. Species-level identification of ivory works of art is necessary to meet government requirements in the traveling of exhibitions containing ivory, as well as learning origins of a piece, and discovering historical trade routes. One of the largest sources of ivory is elephants, of which there are three major species: African Bush Elephant (Loxodonta africana), African Forest Elephant (Loxodonta cyclotis), and Asian Elephant (Elephas maximus). While these elephants are distinct morphologically and genetically, there is no known means to visually distinguish the ivory. This effort is made more difficult for art pieces, as the ivory has been processed in the artifact’s construction and degraded over time. Our goal was to see if it was possible to differentiate between African and Asian elephant species using extraction, amplification, and sequencing of mitochondrial DNA (mtDNA) from ivory taking into account special needs of works of art. Results were compared to known single nucleotide polymorphisms (SNPs) that exist between elephant species in question. We employed a technique that has been used to distinguish between species in wildlife forensics. The sample size reported for this technique is too large for works of art and would disfigure the appearance of the object. Because of equipment limitations, we were unable to test the suitability of smaller sample sizes. In addition, there are portions of the tusk that may not contain an abundance of DNA, and often there is no means to determine from what part of the tusk the object was carved. Given the current technology, we do not believe that identification of species through genetic testing is a viable option for works of art made from ivory.

This research was funded by the Andrew W. Mellon Foundation. We would like to thank the Department of Chemistry and Dr. Zeev Rosenzweig for hosting the SCIART program, and the Department of Biological Sciences for use of their facilities. We would also like to thank Ms. Terry Weisser and the Walter’s Art Museum for their mentorship, guidance, and donation of ivory samples. Special thanks to The Elephant Sanctuary in Tennessee for generous donation of fecal and blood samples.
COMPARATIVE STRUCTURAL ANALYSIS OF UNMYRISTYLATED HIV-2 MATRIX PROTEIN USING NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY

Gabrielle Callwood-Jackson1, Milan Reed2, Hadeel Hamoud3, Nia Starr2, Janae Baptiste4, Dr. Holly Summers4, Dr. Michael Summers4

1Eleanor Roosevelt High School, 7601 Hanover Pkwy, Greenbelt, MD 20770
2Our Lady of Good Counsel High School, 17301 Old Vic Blvd, Olney, MD 20832
3Centennial High School, 4300 Centennial Ln, Ellicott City, MD 21042
4Department of Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle Baltimore, MD 21250

Human immunodeficiency virus type 2 (HIV-2) is a less virulent form of the human immunodeficiency viruses which produce AIDS. It is hypothesized that this is partially due to the behavior of its myristoyl group attached to the matrix protein at different pH levels. At a lower pH, the myristoyl group of the HIV-2 matrix protein is sequestered preventing membrane targeting which stops the virus from proliferating. This is in contrast to the myristoyl group of HIV-1 matrix protein which is exposed at the lower pH. Our main objective was to clone HIV2-MA into a new plasmid to produce the unmyristoylated form of the protein and compare it with the myristoylated HIV2-MA that was made and previously studied. We are hoping to gain a better understanding of the structure of the protein and gain insight as to why it acts differently compared to its close relative HIV1-MA. The HIV2-MA gene was acquired from the Resh plasmid and PCR amplified. It and the pET19b plasmid were double digested using the restriction enzymes Nco1 and BamH1, purified, and then ligated to form a new plasmid containing only the HIV2-MA gene. Previous plasmids contained Human or Yeast N-myristoyltransferase (NMT) which facilitated myristoylation in E. Coli. We wanted to prevent this with our new construct. After confirming the sequence of our new plasmid, we transformed it into BL21 competent cells for protein expression and grew up the protein in E. coli using minimal media to facilitate N15 labeling. Using nuclear magnetic resonance spectroscopy (NMR), we performed a heteronuclear single quantum coherence (HSQC) experiment to evaluate and compare the structure of this protein to the myristoylated HIV2-MA and to its relative HIV1-MA. These comparisons are ongoing.

Our research would not have been possible without the support of the Summer Biomedical Training Program led by Justine Johnson, Dr. Michael Summers, Dr. Holly Summers and Cindy Finch. We would also like to acknowledge The Howard Hughes Medical Institute and NIH/NIGMS (grant # R01 GM42561-28S1) for funding our research.
ARE YOU UP TO THE TUSK?: IDENTIFYING IVORY SPECIES USING RAMAN SPECTROSCOPY AND MASS SPECTROMETRY

Chloe Cao1, Hope La Farge2, Adam Ring3, Terry Drayman-Weisser4, Daniel Walter Rowlands2
1Department of Chemistry, Johns Hopkins University, 3400 N. Charles St., Baltimore, MD 21218
2Department of Chemistry and Biochemistry, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
3Department of Chemistry, Emory University, 1515 Dickey Drive, Atlanta, GA 30322
4The Walters Art Museum, 600 N. Charles St., Baltimore, MD 21201

Analytical chemistry research is needed to improve techniques for identifying elephant ivory by origin and species while minimizing sample size. Poaching of African elephants (Loxodonta africana and Loxodonta cyclotis) and Asian elephants (Elephas maximus) is an issue that ultimately threatens their survival; this makes identifying the origin of elephant ivory even more important for not only addressing ivory poaching and trading, but also assisting museums overseeing ivory artifacts.

To combat elephant poaching, enforcement of African elephant ivory regulations under CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) has intensified. These regulations greatly impact the movement of museum exhibitions, requiring them to identify the ivory’s originating species. While species identification is important for legal documentation, determining the origin of ivory artifacts is crucial for historical, curatorial, and art conservation purposes to achieve a better understanding of its unique context. Culturally significant ivory artifacts pose an additional challenge, as it is imperative to impose minimally invasive sampling techniques.

Bone and antler have a similar composition to ivory, and were included in this study as model systems due to greater availability. This study uses inductively coupled plasma-mass spectrometry (ICP-MS), Raman spectroscopy, and matrix assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF) to differentiate between various species using bone, antler, and ivory. Though results are still in progress, this analytical survey has strong potential to impact methods used to identify ivory and further the current understanding of cultural heritage.

This project is supported by the Baltimore SCIART research experience for undergraduate students, which is a collaborative program between UMBC, Johns Hopkins University, and the Walters Art Museum in Baltimore. The program is funded by the Andrew Mellon Foundation.
DEVELOPMENT OF FAST RECONSTRUCTION TECHNIQUES FOR PROMPT GAMMA IMAGING DURING PROTON RADIOTHERAPY

REU Site: Interdisciplinary Program in High Performance Computing

Johnlemuel Casilag¹, James Della-Giustina², Elizabeth Gregorio³, Aniebiet Jacob⁴,
Graduate assistant: Carlos Barajas⁴, Faculty mentor: Matthias K. Gobbert⁴,
Client: Jerimy Polf⁵

¹Departments of Computer Science and Electrical Engineering, UMBC
²School of Information Technology & Computer Science, Community College of Baltimore Co.
³Department of Physics, Hamline University
⁴Department of Mathematics and Statistics, UMBC
⁵Department of Radiation Oncology, University of Maryland - School of Medicine

Proton beam radiation treatment was first used by Robert Wilson in 1946. The advantage of proton beam radiation is that the lethal dose of radiation is delivered by a sharp increase toward the end of the beam range. This sharp increase is known as the Bragg peak and allows for the possibility of reducing the exposure of healthy tissue to radiation when comparing to x-ray radiation treatment. As the proton beam interacts with the molecules in the body, gamma rays are emitted. The origin of the gamma rays gives the location of the proton beam in the body, therefore, gamma ray imaging allows physicians to better take advantage of the benefits of proton beam radiation.

These gamma rays are detected using a Compton Camera (CC) and the SOE algorithm is used to reconstruct images of these gamma rays as they are emitted from the patient. This imaging occurs while the radiation dose is delivered, allowing physicians to make adjustments in the treatment room. This software is most effective with a resolution of about 1 mm, and a short run time. This project focuses on speeding up the image reconstruction software using a variety of parallel computing techniques involving OpenMP/MPI or hardware like 68-core Intel MIC based CPUs.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-authors Johnlemuel Casilag and Aniebiet Jacob were supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Carlos Barajas was supported by UMBC.
HOW HIV-1 SELECTIVELY PACKAGES ITS DIMERIC GENOME: MAPPING THE
NC BINDING SITES

Kennedy Chioma\textsuperscript{1}, Darin Gilchrist\textsuperscript{1}, Julia Harmon\textsuperscript{2}, Erik Vaughan\textsuperscript{3}, Canessa Swanson\textsuperscript{1}, Pengfei Ding, Ph.D\textsuperscript{1}, Michael Summers, Ph.D\textsuperscript{1}

\textsuperscript{1}Department of Chemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250
\textsuperscript{2}Towson High School, 69 Cedar Ave, Towson, Maryland 21286
\textsuperscript{3}Mt. Hebron High School, 9440 Old Frederick Rd, Ellicott City, Maryland 21042

Human immunodeficiency virus type-1 (HIV-1) is a retrovirus that is the causative agent of acquired immunodeficiency syndrome (AIDS). There are approximately 36.7 million people in the world infected with HIV. The viral genome is reverse transcribed which is a highly mutagenic process, however the 5'-Leader of the genome is the most conserved region. The 5'-Leader undergoes a dimerization process exposing more than a dozen nucleocapsid (NC) binding sites and is responsible for promoting packaging. Previous studies on the HIV-1 5'-Leader discovered that the minimal region required for viral genome selective packaging is the Core Encapsidation Signal (CES). Our research investigates the binding interactions between the NC domain of the Gag polyprotein and the CES of the dimeric viral genome. The high resolution nuclear magnetic resonance (NMR) structure of the CES revealed weakly base-paired and unpaired guanines, which are characteristic for NC binding sites. Utilizing techniques such as: electrophoretic mobility shift assays (EMSA), isothermal titration calorimetry (ITC), and mutagenesis, we elucidated the specific guanine residues within the CES responsible for NC binding. Ultimately, gaining a greater understanding of the mechanism for selective packaging of the viral genome could eventually translate into successful development of viral inhibitors.

Acknowledgement:
This research was funded by NIH/NIGMS grants 1P50GM103297 and the Howard Hughes Medical Institute at UMBC. Additional acknowledgment to the Summer Biomedical Training Program (SBTP).
USE OF TARGETED METABOLOMICS TO IDENTIFY BIOMARKERS FOR RADIATION-INDUCED INJURY

Zachary Clifford¹, Jace W. Jones², Raghavendhar Kotha², Maureen A. Kane²
¹Department of Chemistry and Biochemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
²Department of Pharmaceutical Sciences, UMB, 620 W. Lexington St., Baltimore, MD 21201

A biomarker is a measurable substance that can be used to indicate an organism’s biological state. In this study, high-throughput targeted metabolomics were used to identify potentially viable biomarkers of radiation-induced injury. The data collected in this study will be used to correlate biomarker response to tissue specific injury.

Non-human primate (NHP) plasma samples from a partial body irradiation model with 5% bone marrow sparing (PBI/BM5) were analyzed via targeted high-throughput quantitative metabolomics using the Biocrates Absolute IDQ p180 kit (Biocrates, Life Science AG, Innsbruck, Austria) and liquid chromatography-mass spectrometry. The concentrations of each of the metabolites in the kit were monitored at various time points post-irradiation. The time points to be analyzed were: Day: 0 ( naïve), 1, 3, 5, 7, 15, 21, day of euthanasia (dE, day 180 shown here) with n=4-8 NHP plasma samples per time point.

Several gastrointestinal injury, lung injury, and oxidative stress biomarkers were identified throughout the course of this study. In past studies, citrulline has been validated as an acute gastrointestinal injury biomarker. In this study, citrulline shows a similar response to previous reports. There has been some evidence that a decrease in glutamate levels can be correlated to pulmonary injuries. However, as of now, glutamate has yet to be verified as a chronic lung injury biomarker. It has also been reported that the concentration of lyso-phosphatidylcholine (Lyso PCs) is elevated under oxidative stress conditions such as radiation. However, in this study, the concentration of Lyso PCs transiently decreases and then increases over time.

The data collected in this study will serve as a starting point for defining the clinical endpoint relationship that is required for FDA biomarker qualification based upon the biomarker relationship with clinical endpoints and histopathological analysis.

The targeted metabolomics experimental data was collected by Jace Jones and Raghavendhar Kotha. This work was funded by NIH/NIAID Contract No. HHSN272201000046C and a sub-contract from SRI International. This work was also funded in part by the University of Maryland School of Pharmacy Mass Spectrometry Center (SOP1841-IQB2014). Zachary Clifford was supported by the Department of Pharmaceutical Sciences Summer Intern Program.
MICROWAVE-ASSISTED LYSING OF LISTERIA MONOCYTOGENES AND VIBRIO CHOLERAE

Amanda Harvey¹, Shannon Cole¹, Tonya M. Santaus¹,² and Chris D. Geddes¹,²
¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
²The Institute of Fluorescence, Columbus Center, University of Maryland Baltimore County, 701 E. Pratt Street, Baltimore, MD, USA 21202.

Listeria monocytogenes (Listeria) and Vibrio cholerae (Cholera) are two bacteria that cause detrimental health outcomes for infected individuals. Listeria is spread through contaminated food substances while Cholera is transmitted through contaminated water in areas of poor sanitation. Lysing is the process by which cell membranes are broken apart to release intracellular components such as DNA, which can be used for the subsequent identification of the infecting organism.

In this poster, we analyze microwave powers and times to lyse Listeria monocytogenes and Vibrio cholerae cells using a 900W microwave. We use Polymerase Chain Reaction (PCR) to determine which lysing parameters are optimal in yielding the highest concentration of isolated DNA from the cells.

Listeria and Cholera are mixed separately into deionized (DI) water making a $10^8$ colony forming unit (cfu) concentration. The solution is placed onto a blank slide or a slide with gold triangles and microwaved at various powers and times. The slides with the gold triangles increase the temperature of the solution compared to the blank slides. The solution is also placed onto filter paper with various concentrations. Once the filter paper is dry it is placed into 1mL of DI water and vortexed. The same tests are completed with the filter paper. After the experiments are complete and the microwaved solution has cooled, ethanol precipitation is performed. Next, the ethanol is extracted out and the pellet is left to dry. Lastly, the pellet is hydrated with DI water and sent for PCR.

A collaborator at the University of Maryland, Department of Epidemiology performs the quantitative PCR. The PCR cycle number is used to determine the optimum lysing parameters.

We would like to acknowledge Dr. Geddes, The Institute of Fluorescence, and UMBC for allowing us to conduct research in their facilities.
DISTRIBUTION OF CHEMOATTRACTANTS IN A HETEROGENEOUS TISSUE AND ITS IMPACT ON CELL MIGRATION

Jessica Cooley\(^1\), Aaron George\(^2\), Zachary Mekus\(^3\), Victoria Sabo\(^4\),
Graduate assistant: Morgan Strzegowski\(^1\), Faculty mentor: Bradford E. Peercy\(^1\),
Client: Michelle Starz-Gaiano\(^5\)

\(^1\)Department of Mathematics and Statistics, UMBC
\(^2\)Department of Mathematics, University of Maryland College Park
\(^3\)Department of Computer Science and Engineering, Washington University in St. Louis
\(^4\)Department of Mathematics and Statistics, Georgetown University
\(^5\)Department of Biological Sciences, UMBC

Cell migration is the process in living organisms by which the body heals and diseases spread. Comprehension of this mechanism will be beneficial to understanding its applications, yet little is known about this process. We study the cluster cell migration in the egg chamber of \textit{Drosophila melanogaster}, or fruit flies, because it is easy to observe and sample in that organism. A previous model simulated the cell cluster's migration using forces to determine movement of many individual cells; we improved and revised this system. We created a more geometrically accurate model of the egg chamber and mapped the diffusion of the chemoattractants through that domain using a reaction diffusion system. In addition, the base implementation is updated to more accurately simulate the cell migration process. This model will allow several questions to be investigated, such as identifying the source and quantity of the chemoattractants, the rate at which they are taken in by other cells in the egg chamber, if at all, and the time needed for them to reach the polar and border cells at the anterior of the chamber that gives the most faithful representation of experimental results.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-author Jessica Cooley was supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Morgan Strzegowski was supported by UMBC.
Human immunodeficiency virus (HIV) uses RNA as its source of genetic material. This RNA can exist as two different conformations, the monomer and the dimer, which ultimately determines the role of the RNA in later steps of the viral life cycle. As a monomer, the RNA will go on to be translated into viral proteins. The dimer, however, is packaged into new HIV virions. The key to understanding how these RNA structures determine function is in the start site heterogeneity (TSS) mechanism. This mechanism describes how manipulating the number of guanines on the viral RNA can cause the native dimer to change into the monomer conformation; therefore changing the final function of the RNA. Using nuclear magnetic resonance (NMR) spectroscopy and in vivo studies, the existence of TSS, as well as its nature to disrupt the regions around it, has been confirmed. In the monomer structure (Cap 2G, Cap 3G), additional guanosine residues cause remodeling of polyA and extended U5-DI5 Interaction. As for the dimer structure (Cap 1G), poly A is stable and DIS is no longer being sequestered thus allowing it to interact with a DIS of another RNA, forming a dimer. Our focus now includes assigning all the NOSEY signals for the Cap-1G dimer conformation to solve the secondary structure.

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TRANSCRIPTION AND CAPPING OF HIV-1 RNA: THE NEW PROTOCOL

Amara Ejikemeuwa¹, Joshua Brown², Michael Summers, PhD²
¹University of West Florida, 11000 University Parkway, Pensacola, FL 32514
²Department of Chemistry and Biochemistry, University of Maryland, Baltimore County,
Baltimore, MD 21250

The human immunodeficiency virus (HIV-1) is a retrovirus containing an RNA genome, rather than DNA. After fusing to a CD4 cell, the viral RNA is reverse transcribed and integrated into the host DNA. The infected cell then transcribes the DNA back to viral RNA. The RNA exists in equilibrium between monomer and dimer conformations, where monomers are translated into proteins, and dimers are packaged into genomic material for new virions. According to in vivo studies, there is a mixed population of capped viral mRNA, meaning that there are start sites beginning with one, two, or three guanosines plus the native cap residue (Cap1G, Cap2G, and Cap3G).

RNA purification is a tedious process, starting with an in vitro transcription reaction. The product is loaded into a denaturing gel, ran through an elutrap for RNA collection, and washed various times - taking up to four days. As viral RNA naturally exists in a capped conformation throughout the cell, the purified product can be capped for further studies. The capping protocol adds a guanosine to the 5’ Leader of the RNA, and follows through another round of the same purification process. This means that whole procedure can take up to eight days from start to finish. The new protocol utilizes filtration to combine the transcription and capping procedure into a single reaction. After in vitro transcription, the product is filtered to remove reagents and residues, and the capping procedure begins. This new protocol shortens the procedure by three to four days, increasing the pace of progress for the overall goal of understanding the three-dimensional structure of the monomer RNA through Nuclear Magnetic Resonance (NMR) spectroscopy.

This research was funded by the MARC U*STAR Program from the University of West Florida, and the NIH/NIGMS grant 1P50GM103297. This research was conducted at the Howard Hughes Medical Institute at UMBC with support in part by the Howard Hughes Medical Institute's Precollege and Undergraduate Science Education Program.
INVESTIGATION OF SMALL WORLDNESS OF PANCREATIC ISLETS

REU Site: Interdisciplinary Program in High Performance Computing

Elise Falgout¹, Destiny Frett², Lorenzo Neil³, Ryan Schumm⁴,
Graduate assistant: Janita Patwardhan⁵, Faculty mentor: Bradford Peercy⁵,
Client: Arthur Sherman⁶

¹Departments of Computer Science and Mathematics, Louisiana State University
²Departments of Computer Science and Mathematics, Elon University
³Department of Computer Science and Electrical Engineering, UMBC
⁴Departments of Physics and Mathematics, Marquette University
⁵Department of Mathematics and Statistics, UMBC
⁶Laboratory of Biological Modeling, National Institutes of Health

Diabetes occurs when the body's blood sugar levels are in a state of sustained elevation. The pancreatic beta cells, organized in the islets of Langerhans, secrete a hormone called insulin that is responsible for maintaining blood glucose at appropriate levels. Oscillations in insulin levels, which are thought to require synchronization in insulin secretion, are necessary for proper regulation of glucose. A loss of this periodic behavior has been observed in type 2 diabetic patients. We used the Single Slow Channel Model to compute the calcium and electrical dynamics during insulin secretion of a single beta cell. To replicate an islet, we coupled the cell cluster according to a hexagonal-close-packed lattice. The existence of small worldness in the islet and its effect on islet performance was investigated by using methods from graph theory. To quantify the performance, we developed a synchronization index from previously used indexes that reflects to what degree the electrical or calcium oscillations are in phase. The effect small worldness has on synchronization is indicative of the existence of hub cells, which have a larger influence on the rhythm of the islet. Thus, the destruction of hub cells within the islet would disrupt its synchronization.

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**tRNA<sub>Lys</sub> OUTCOMPETES PI(4,5)P<sub>2</sub> IN MODEL CELL MEMBRANES FOR HIV-1 MATRIX BINDING**

Matthew McDonough<sup>1</sup>, Noel Getachew<sup>2</sup>, Emre Tkacik<sup>2</sup>, Phoebe Somani<sup>2</sup>, Amalia Rivera-Oven<sup>2</sup>, Christy Gaines<sup>2</sup> and Dr. Michael Summers<sup>2</sup>

<sup>1</sup>Department Chemistry and Physics, Western Carolina University, 1 University Way, Cullowhee, NC 28723

<sup>2</sup>Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250

HIV-1 is a retrovirus that infects human immune T-cells. One of the proteins vital for HIV-1 replication is the Gag polyprotein. The N-terminal domain of Gag, matrix, is essential for Gag targeting and binding to the plasma membrane (PM) to start virion assembly. Matrix binds to phosphatidylinositol 4,5 bisphosphate [PI(4,5)P<sub>2</sub>] in the PM, and may also specifically target lipid rafts. Rafts are rigid, liquid order portions of the PM that are high in cholesterol and certain phospholipids. Matrix also binds to tRNAs in the cell, and many believe this regulates matrix-membrane interactions. According to the current model, tRNA binds to matrix in the cytosol but when this complex reaches the PM, PI(4,5)P<sub>2</sub> will outcompete the tRNA and matrix will bind to PI(4,5)P<sub>2</sub>. To test this theory, we created model membranes (liposomes), representing either raft or non-raft regions of the PM. We used the compositions consistent with earlier experiments in our lab, and also created liposomes using new ratios to mimic MT4 cell PMs and the HIV lipid envelope. For each liposome type we used membranes containing or lacking PI(4,5)P<sub>2</sub>. We performed 1D-<sup>1</sup>H NMR liposome competition assays in which we observed the interaction between the matrix-tRNA<sub>Lys</sub> complex and these different liposomes. With no tRNA present we saw more binding to rafts and liposomes containing PI(4,5)P<sub>2</sub>, than non-rafts and liposomes lacking PI(4,5)P<sub>2</sub>. This was expected based on the literature and previous experiments. Interestingly, in the competition assays which involved the tRNA-matrix complex, binding did not change regardless of the type of liposome, indicating that PI(4,5)P<sub>2</sub> does not outcompete tRNA<sub>Lys</sub> for matrix binding.

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DETECTING BOVINE LAMENESS USING SPLINE TRANSFORMATIONS OF THREE-DIMENSIONAL LIMB MOVEMENT VARIABLES IN A LOGISTIC REGRESSION MODEL

REU Site: Interdisciplinary Program in High Performance Computing

William Dula\textsuperscript{1}, Jason Glover\textsuperscript{2}, Ari Goldbloom-Helzner\textsuperscript{3}, Kayla Makela\textsuperscript{4},
Graduate assistants: Qing Ji and Sai Kumar Popuri\textsuperscript{2}
Faculty mentors: Nagaraj Neerchal\textsuperscript{2} and Andrew Raim\textsuperscript{6}
Client: Uri Tasch\textsuperscript{5}

\textsuperscript{1}Department of Mathematics, Morehouse College
\textsuperscript{2}Department of Mathematics and Statistics, UMBC
\textsuperscript{3}Division of Applied Mathematics, Brown University
\textsuperscript{4}Department of Mathematics and Statistics, Michigan State University
\textsuperscript{5}Department of Mechanical Engineering, UMBC
\textsuperscript{6}United States Census Bureau

Bovine lameness is a common issue on commercial dairy farms, resulting in decreased productivity. Early detection is essential for effective treatment of lameness. Here and recent previous work, lameness status of a cow has been modeled using three-dimensional limb movement measurements related to its gait. A statistical model uses the software SAS\textsuperscript{®} with its LOGISTIC and TRANSREG procedures. The model produces a binary classification, lame or not lame. Current implementation requires running several SAS\textsuperscript{®} procedures manually and therefore is not amenable to a large scale application and model optimization. In this work, we implement an optimization algorithm in R to mirror the TRANSREG procedure, and thus speed up exploration of a large number of candidate models to optimize the widely used goodness of fit criteria such as the Receiver Operating Characteristic curve (AUC). The models are also evaluated using classification error rates (False Positives and False Negative or equivalently sensitivity and specificity). We also consider multinomial logistic models so that cows may be further classified into three categories: severely lame, mildly lame or sound. These results can be used in the commercial dairy industry for lameness detection.

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DETERMINING THE STRUCTURE OF THE HIV-1 RNA WITH FRAGMENTATION USING NMR

Jana Hijji¹, Tatiana Rodriguez², Joshua Brown², Michael Summers, PhD²
¹University of Maryland-College Park, College Park, MD 20742
²Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, Baltimore, MD 21250

The structure of the 5'-Leader (5'-L) of the human immunodeficiency virus (HIV-1) viral RNA genome is under intense study, because of its role in determining the fate of the RNA. To help us determine the structure of the 5'-L, we use nuclear magnetic resonance (NMR) spectroscopy. The NMR spectrometer provides us with the chemical and structural environment of individual hydrogens within the RNA molecule. Using this information we can determine the three-dimensional structure of the HIV-1 5'-L. NMR spectra of large RNAs, such as the 5'-L are often crowded and contain overlapping signals, causing the data to be difficult to interpret. In order to counter this, we prepare smaller oligos that mimic the structural elements of the larger RNA, but whose spectra can be confidently assigned. This technique has allowed us to confirm multiple regions of the full-length leader by overlapping the data of the leader with that of the assigned smaller control oligos. With the help of NMR we have been able to determine the 2-D structure of multiple regions of the monomer and dimer conformations of the 5'-L.

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Transient protein-protein interactions (tPPIs) are essential for cellular processes, including the regulation of biological pathways. To overcome low signals of weak and transient interactions we developed a Vesicle Based Nano-Reaction Chamber system, which encloses a single protein within its nanoscale sack structure. These Nano-Reaction Chambers are designed to coalesce with separate vesicles, mix contents, and form larger vesicle chambers. This allows their single protein contents to all be contained in the same chamber and interact in order to study the transient interactions between the proteins. We utilize two independent fusion mechanisms to mix the single protein contents of chambers allowing multiple controlled single additions. Because this system operates at the single molecular level the Nano-Reaction chambers can provide us with details of kinetic data that is not provided by bulk studies.

To demonstrate the efficacy of using Vesicle Based Nano-Reaction Chambers to study transient protein-protein interactions, we designed a set of primers to alter the affinity of a well-studied ternary complex, the FKBP•Rapamycin•FRB complex. The resulting mutants with complexes with a range of different binding strengths that can be observed will be used in our Vesicle Based Nano-Reaction chambers to confirm the ability to distinguish varying levels of affinity within protein complexes. Here, we report results of Nano-Reaction Chambers produced by two independent methods of vesicle fusion completed sequentially as measured by single molecule microscopy.

This research project is supported by UMBC.
Transient protein-protein interactions (PPIs) occur in signaling and metabolic pathways. Studying PPIs will provide insight to these pathways, which is important for developing drugs that target PPIs and disrupt pathways related to disease. The binding affinities of transient PPIs will be quantified by utilizing two new techniques: nano-reaction chambers and super-spatiotemporal resolution microscopy. To validate these new techniques, a series of complexes with various binding affinities and interaction times will be developed.

Rapamycin binds to the FK506 binding protein (FKBP) and the mammalian target of rapamycin (mTOR) to form a ternary complex. The part of mTOR that participates in binding to rapamycin is called the FKBP-rapamycin binding domain (FRB). Binding affinities have previously been quantified for all components of the FKBP•rapamycin•FRB ternary complex, with the exception of the protein-protein interaction between FKBP and FRB, which may be too fast for previous techniques to characterize. The rapamycin binding pocket of FRB was modified by site-directed mutagenesis to form six FRB mutants that bind more weakly to and dissociate more quickly from the FKBP•rapamycin complex. The transient interactions between the FRB mutants and the FKBP•rapamycin complex will be characterized at the single-molecule level with new nano-reaction chambers. A consecutive fusion mechanism enables a single mutated ternary complex to form inside a single nano-reaction chamber. Interaction traces will be collected over time to detect the association and dissociation of this complex. The mutated ternary complexes will also be characterized by a new super-spatiotemporal resolution microscope that will provide sub-millisecond temporal resolution of this in vitro system. The characterized mutant complexes will be used as binding affinity markers to validate the kinetic properties of transient PPIs in various biological pathways.

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EFFECTS OF SLEEP ON THE IMMUNITY OF DROSOPHILLA MELANOGASTER

Ayodeji Adeleke¹, Safiya Ismail³, Nina Evans⁴, Decole Russell², Chetana Jadhav⁴, Jeff Leips⁴

¹ The Community College of Baltimore County, Baltimore, MD 21228
² Anne Arundel Community College, Arnold, MD 21012
³ Prince George’s Community College, Largo, MD 20774
⁴ Department of Biology, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

*Drosophila melanogaster,* also known as the fruit fly, is commonly used to study human diseases due to the genetic similarities between the two species. Previous studies have pointed to a connection between sleep and immunity, although the genetic basis of this relationship is unknown. Interestingly, there have been conflicting studies on the amount of sleep needed to promote immunity in *Drosophila.* The main goal of this project was to determine if flies genetically programmed for longer sleep patterns survived bacterial infection better than those programmed for shorter sleep. We inoculated 10 genetic lines (n=50 each) of short sleepers and 10 genetic lines (n=50 each) of longer sleepers with the bacterium *Enterococcus faecalis* and measured fly mortality at 24, 48, 72, and 96 hours post inoculation. Our results revealed that short sleepers had an increased mortality over time compared to long sleepers over the course of 72 and 96 hours. At the 72-hour time point, we found there to be a statistically increased mortality in the short sleepers (33.4 +/- 3.70%) compared to the long sleepers. At the 96-hour time point, we found there to be a statistically increased mortality in the short sleepers (51.4 +/- 2.65%) compared to the long sleepers. Our results provide evidence that there is a correlation between sleep duration and immunity. The future goal of this experiment is to identify genes that are responsible for both sleep and immunity that will allow us to understand the connection.

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The Bahama Oriole is a critically endangered songbird endemic to the Bahamas that is currently found only on the Andros island complex. The most recent published research concluded that fewer than 300 individuals remain in the shrinking population. Loss of coconut palms due to lethal yellowing disease, brood parasitism by the Shiny Cowbird, and the loss of foraging habitat are the most commonly cited threats to the species. Previous research and population estimates were influenced by the belief that the Bahama Oriole nests almost exclusively in non-native coconut palms within residential and agricultural habitats. However, our recent findings show conclusive evidence of pine forest nesting in both Caribbean Pine and native understory thatch palm. This new understanding of Bahama Oriole habitat usage represents a significant paradigm shift in our understanding of the breeding ecology and population size of the species. Since effective conservation is dependent on thorough and accurate knowledge of the species, the need for further study into the previously overlooked pine forest habitat became clear. Our research aimed to identify the potential predators in the pine forests of our North Andros study area using camera trapping. We documented the presence of feral cats in developed areas as well the most remote parts of the pine forest. These findings are of particular concern given that feral and domesticated cats are well-documented predators of birds and have led to the extinction of more than 30 island-endemic bird species. Moving forward, we will conduct more research on the Bahama Oriole and its pine forest habitat in order to accurately measure the population size, assess the threat of predation, and lead effective conservation efforts.

For funding we thank the American Bird Conservancy, Explorers Club Youth Activity Fund, MBZ Species Conservation Fund and University of Maryland, Baltimore County URA Scholarship.
36 million people are currently infected with human immunodeficiency virus (HIV), a retrovirus responsible for the onset of the acquired immunodeficiency syndrome (AIDS). Upon transmission, the virus invades CD4+ T cells and integrates its proviral genome into the host genome leading to a life-long infection. During the viral life cycle, interactions between the unspliced viral RNA and its translated product, the Gag polyprotein, initiate the packaging of two copies of the HIV genome. Gag contains three structured domains: Matrix (MA), Capsid (CA), and Nucleocapsid (NC). The NC domain of Gag binds to exposed or weakly base paired guanines of the 5’-leader (5’-L) to initiate genome packaging and viral assembly.

We seek to characterize the Gag-RNA interactions essential to genome packaging using a truncated 5’-L derivative and a hexameric NC protein. We mapped the multiple guanines responsible for NC binding and packaging through mutagenesis. NC binding promotes the formation of Gag hexamers that potentially function as the nucleation site to initiate the viral assembly. Due to hexamer-hexamer interactions in the C-terminal Domain (CTD) of CA, the protein aggregates and precipitates in the presence of RNA. To circumvent these problems, we have fused the NC domain of Gag to a hexameric protein scaffold, which mimics hexameric Gag, while forming isolated hexamers and preventing aggregation. The hexameric NC protein forms a 1:1 complex with the core region of dimeric 5’-L with a very low dissociation constant.

With our RNA and protein constructs, we aim to deduce structural information of the Gag-RNA interactions using X-ray crystallography and cryo-electron microscopy. These studies will further our understanding of the mechanism of HIV genome packaging, which can be helpful for target - based therapeutic developments.

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GRIPFAB: FACILITATING THE CREATION OF CUSTOMIZABLE GRIPS FOR INDIVIDUALS WITH LIMITED HAND MOBILITY

Maria Francine Lapid, Amy Hurst

1Department of Electrical and Computer Engineering, Texas A&M University, 400 Bizzell St, College Station, Texas 77843

2Information Systems Department, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250

The increasing prevalence and quality of desktop, consumer-friendly 3D printers has made it possible for more people who do not have a background in engineering or technology to create personalized items for everyday use. GripFab, a web-based computer-aided design (CAD) software, aims to empower physical therapists, who might not have a 3D modeling background, to make custom assistive technology for their patients who have limited hand mobility. The grips made via GripFab are 3D printed to be able to hold small utensils such as spoons and styluses.

In the first version of GripFab, the user chooses from four different grip bases and is asked for specific measurements based on the client. The program then creates a 3D model of the grip that is ready to be printed based on these measurements. This summer, I am exploring the possibility of making GripFab even more customizable by utilizing 3D scanning and image processing technology. Therapists make clay prototypes based on each patient’s hand shape and add small colored stickers to indicate the areas that need to be augmented. I created a 3D image processing program in C++ for GripFab that recognizes the different parts of the clay prototypes that need to be augmented. The program then writes an OpenSCAD script that edits the initial 3D scanned object based on the user’s new preferences. This allows the grips to be more customizable and leaves room for the grips to be able to hold multiple types of utensils.

By increasing the customizability and making GripFab more universal, we can introduce GripFab to other professionals besides therapists who also work with individuals who have limited hand mobility.

This project was made possible by the Computing Research Association -- Women (CRA-W) the Distributed Research Experiences for Undergraduates (DREU) under the mentorship of Dr. Amy Hurst in the Prototyping and Design Lab. The program is funded by the National Science Foundation (NSF).
Histone proteins are scaffolds for DNA within the nucleus. They organize the DNA and make it accessible to other proteins. In addition to providing the DNA structure, histones are also common sites for post translational modifications, such as lysine methylation. These modifications affect DNA templated processes, like chromosome segregation, transcription, and gene repair. Recent studies have suggested that abnormality in histones is linked to latent defects in chromatin structure, leading to cancer and other developmental complications. In this project, we used the model organism *Saccharomyces cerevisiae*, the budding yeast, to study an uncharacterized lysine methyltransferase: Set6. It contains a SET domain, a common signature of chromatin regulatory proteins that catalyzes lysine methylation on either histones or other non-histone targets. However, the function of Set6 in cells is entirely unknown. Therefore, the aim of our project was to investigate biological pathways in which Set6 is involved. We performed genetic interaction and protein-protein interaction studies of Set6. According to our preliminary data, cells without Set6 are sensitive to the drug benomyl, which indicates that these cells may have defective microtubule function or cell cycle regulation. Furthermore, since we know that lysine methyltransferases usually work in larger complexes, we performed immunoprecipitation followed by mass spectrometry to identify potential protein partners of Set6. We analyzed two candidate proteins that showed the strongest interaction with Set6: Gim3 and Yke2. These two proteins are part of the co-chaperone prefoldin complex that is important for microtubule assembly. They also localize to chromatin and participate in transcriptional elongation. We are using co-immunoprecipitation experiments to further test the interaction between Set6 and these factors. Overall, our data has opened new avenues of investigation for understanding the function of Set6, including determining its potential roles in microtubule assembly and transcription elongation.
OPTIMIZATION OF GOLD NANOPARTICLES TO BE USED AS A MULTIFUNCTIONAL DRUG DELIVERY SYSTEM FOR CHEMOTHERAPY

Angelie Matar¹, Arunendra Saha-Ray², Dr. Marie-Christine Daniel²
¹Department of Chemistry, Louisiana State University, 232 Choppin Hall, Baton Rouge, LA 70803
²Department of Chemistry and Biochemistry, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

The development of a gold nanoparticle (GNP) cored dendron as a drug deliver system can help people advance towards a better treatment of chemotherapy. The problem with the current application of the medicine is that it affects the whole human body, not just the tumor area. With the information collected in this research, we aim to eliminate the preexisting problem of excessive drug toxicity in the body and enhance targeting efficiency. By using these nanoparticles, we have found a way to get the drug into the body with enhanced permeability and retention (EPR) effect which provides passive targeting. Furthermore, through active targeting, the nanocarriers are able to attach to the cell and release the drug into the nucleus.

The design of this drug is a GNP core coupled with spacers and dendrons around the surface that have multifunctional uses on their termini. The dendron is a first generation poly(propylene imine) (PPI) dendron with a phenolic focal point, and the spacer is a tetraethylene glycol (TEG) molecule that is connected at that focal point of the PPI dendron. With this model, we have multiple branches around the core that are made in a very limited amount of steps, and we can organize the distribution of different functions on the molecule connected to each branch. This makes the targeting to the tumor more efficient, makes multimodal imaging easier, and increases the amount of drug molecules attached.

In this study, we have functionalized the first generation dendron with near infrared fluorescence marker. We have also attached Doxorubicin to a third generation dendron. With this ideal drug, we are able to have a multifunctional drug delivery system that releases into the body with decreased drug toxicity. Future studies include being able to attach molecules to all eight branches in the third generation PPI.

This research project is supported by the National Science Foundation Research Experience for Undergraduates (REU) Research award CHE-1460653 and National Science Foundation Research award CHE-1507462.
NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY CHARACTERIZATION OF
THE FELINE IMMUNODEFICIENCY VIRUS MATRIX PROTEIN MYRISTOYL
SWITCH

Simon Maxwell¹, Paige Canova¹, Constance Nyaunu², Janae Baptiste¹, Michael Summers¹
¹Howard Hughes Medical Institute and Department of Chemistry and Biochemistry, University
of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250, USA
²Department of Chemistry, Howard University, 2400 Sixth St NW, Washington, DC 20059,
USA

The feline immunodeficiency virus (FIV) is a lentivirus that infects roughly eight percent
of cats worldwide. FIV is of interest to humans because cats have potential as animal models
to develop HIV treatments. A key structural feature of both HIV and FIV is the myristoylated
matrix protein (myrMA), which is essential for viral assembly and, ultimately, the production
of new virus particles. The myristoyl group, a fourteen-carbon saturated fatty acid that is covalently
attached to the N-terminus of MA, adopts one of two conformations: either sequestered within a
hydrophobic cleft of MA or in an exposed conformation that serves to anchor MA to the plasma
membrane. These conformations exist in equilibrium and the conversion from one to the other is
referred to as the myristoyl switch. Previous research on HIV-1 MA suggests that this moiety
acts as a “pH sensor” such that, under high pH conditions, the myristoyl group is sequestered but
becomes exposed in an acidic environment. Gel electrophoresis data indicate that myrMA
solubility is decreased under acidic conditions, potentially as the result of myristoyl exposure and
subsequent protein aggregation. We hypothesize that the FIV myristoyl moiety adopts a similar
myristoyl switch mechanism to HIV-1 MA. This work describes efforts to evaluate whether pH
modulates myristoyl exposure by using nuclear magnetic resonance (NMR) spectroscopy to
obtain structural information as a function of pH and time. Ultimately, elucidating the structural
mechanisms of the FIV myristoyl switch may present a model system for the development of
therapeutic targets against HIV.

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THE ABSORBANCE CHARACTERISTICS OF FIVE UV-FILTERS USED IN COMMON SUNSCREEN PRODUCTS

Chelsea Mikal, Ethan Hain, Mamatha Hopanna, Lee Blaney
Department of Chemical, Biochemical and Environmental Engineering, 1000 Hilltop Cir, Baltimore, MD 21250

Sunscreen is a common personal care product that is used to absorb harmful ultraviolet (UV) rays from the sun. The absorption properties of the active ingredients (i.e., UV-filters) in sunscreen products protect skin from damage caused by UV light. However, previous authors have noted concerns about the ecotoxicity of UV-filters on aquatic and marine organisms. In this work, we studied five UV-filters, namely oxybenzone, octocrylene, octisalate, homosalate, and sulisobenzone, to determine their absorbance spectra and consider their environmental fate in natural and engineered systems. The absorbance spectra for the five UV-filters of concern were examined at 200-900 nm wavelengths and pH 4-10. The Beer-Lambert law was applied to calculate wavelength-dependent molar extinction coefficients for each compound. As expected based on their sunscreen properties, the UV-filters exhibited intense absorbance peaks below 400 nm. For instance, octocrylene showed an characteristic absorbance peak at 320 nm, with corresponding molar extinction coefficient of $2.0 \times 10^4 \, \text{M}^{-1}\text{cm}^{-1}$. On the contrary, octocrylene showed low molar extinction coefficient value of $5.0 \times 10^3 \, \text{M}^{-1}\text{cm}^{-1}$ at 254 nm. These results suggest that octocrylene will absorb more light in natural settings (i.e., 300-400 nm) compared to that used for disinfection processes in water/wastewater treatment (i.e., 254 nm). Findings for the other UV-filters will be presented to identify the ranges of high absorbance for each chemical compound and provide insight into their relative fate during natural and engineered processes.

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THE ASSOCIATIONS BETWEEN NON-NATIVE ACCENT, PERCEIVED DISCRIMINATION, AND DEPRESSION IN ASIAN AMERICANS

Tuong Vy C. Nguyen¹, Kathy T. T. Vu², Cheah S. L. Charissa²
¹Department of Psychology, Xavier University of Louisiana, 1 Drexel Drive, New Orleans, LA 70125
²Department of Psychology, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

Past research on language has focused primarily on language ability or proficiency rather than accents (Russo et al., 2016). However, the experience of Asian American individuals with non-native accents and the effect of the stigmatization of accents on these individuals are important to study because these individuals are at risk for experiencing discrimination (Lippi-Green, 1994) and consequent poor mental health (Stein, Supple, Huq, Dunbar, & Prinstein, 2016). One of the most pervasive stereotypes for Asian Americans is the Perpetual Foreigner Stereotype, which is the perception that Asian Americans are not Americans despite having American citizenship or having lived in the U.S. for an extended period of time. The presence of non-native accents may exacerbate this stereotype. As a result, Asian Americans with non-native accents may experience greater racial discrimination, which has been associated with more depressive symptoms (Chakraborty & MacKenzie, 2002). Past research has also found that ethnic identity can buffer against the negative effect of perceived discrimination on mental health among Asian Americans (Rogers-Sirin & Gupta, 2012). However, these associations have been understudied in Asian Americans with non-native accents despite the rapid growth of this particular group (U.S. Census Bureau, 2012). Therefore, this proposal aimed to examine the associations between having non-native accents, perceived discrimination, and depressive symptoms among Asian Americans. In addition, the moderating role of ethnic identity in the associations between discrimination and depressive symptoms will be explored.

This research experience would not have been possible without the financial support of the McNair Program from Xavier University of Louisiana. I am grateful for Charissa S. L. Cheah and Kathy T. T. Vu, from the Department of Psychology at the University of Maryland, Baltimore County, who both guided me in this project. I am also thankful for Justine Johnson, the Associate Director of the Meyerhoff Graduate Fellows Program, and the Summer Biomedical Training program for providing me this research opportunity. I have had an insightful learning experience thanks to these programs and individuals.
The human immunodeficiency virus (HIV) is a lentivirus that has infected millions of people since the beginning of the epidemic in 1981. HIV is still prevalent in today’s society, and one step that can be taken in order to improve HIV treatment is through the use of an animal model. Cats infected with the feline immunodeficiency virus (FIV) have potential as models because the feline immune response to FIV and progression to cat AIDS is similar to humans with HIV. FIV and HIV viral assembly occurs in a similar fashion to, ultimately, prepare new virions. During these assembly processes, the Gag polyprotein (Gag) targets a lipid raft on the cellular membrane by means of the matrix domain (MA). The myristoyl moiety, a fourteen-carbon saturated fatty acid, must be linked to MA and is critical for MA targeting to the plasma membrane. N-myristoyltransferase (NMT) recognizes the myristoylation signal of the substrate MA and binds the myristoyl group to the N-terminal residue. Previous studies identified that FIV MA has a unique myristoylation signal in comparison to the signals of other host and mammalian proteins. The goal of this project is to understand if there is a structural reason for this distinct sequence through nuclear magnetic resonance (NMR) spectroscopy. One obstacle to overcome in the process of solving the protein structure was the inability to collect data corresponding to the N-terminus of the protein under the established conditions. In order to retrieve this data, the impact of pH on NMR data was investigated. The NMR results indicated that, upon lowering the pH from 8 to 5, additional data were observed which will allow for determination of the protein structure. By knowing the structure of MA, inhibitors can be developed to prevent assembly in FIV infected cats which may be useful in the advancement of HIV therapies.
The human immunodeficiency virus type one (HIV-1) is a retrovirus that suppresses activity of the human immune system. During the late phase of HIV-1 replication, transcripts of the viral genome are produced, and alternative splicing events occur. For viral replication to proceed, unspliced and singly spliced RNAs must be exported out of the nucleus. This requires some spliced transcripts to be translated into Rev, a viral protein. Rev is imported back into the nucleus, where it binds and oligomerizes along a landmark within unspliced and singly spliced RNAs, called the Rev Response Element (RRE). RRE-Rev complexes are then recognized by nuclear export machinery, which leads to export of unspliced and singly spliced RNAs. A recent publication suggests the RRE also has an affinity for Gag, a polyprotein that recognizes and packages the HIV-1 genome during replication. This research will attempt to understand the binding of the RRE with HIV-1 proteins, specifically Rev and a variety of Gag constructs.

Electrophoretic mobility shift assays (EMSAs) are used to study RNA-protein interactions. As RNA-protein complexes form, gel bands migrate slower than the RNA alone, due to increased molecular size. RRE-Rev complexes are challenging to study with this method due to Rev oligomerization at the minimum concentrations required for ethidium bromide (EtBr) staining. In order to effectively visualize RRE-Rev interactions, a more sensitive dye is necessary, such as SYBR Green II. A side-by-side analysis of SYBR Green II and EtBr staining is currently in process, which will help determine the best dye for EMSA use. This work will aid in accomplishing one of the goals of the project, which is to characterize the interactions between the RRE and HIV-1 proteins. Accomplishing this goal will be significant for HIV-1 therapeutic development.

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DEVELOPING AN ADAPTATION AND OTHER FEATURES FOR PINATA, A CHROME-BASED ADAPTIVE USER INTERFACE TOOL

Christian Ortega1, Tejas Bhalerao2, Amy Hurst2, Foad Hamidi2

1Department of Computer Science and Engineering, Texas A&M University, 400 Bizzell St, College Station, Texas 77843
2Information Systems Department, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250

The Internet has become an essential tool for communication, entertainment, and employment. Unfortunately, web-based interfaces can be inaccessible or difficult to navigate for users who experience difficulty in accurately controlling pointing devices such as computer mice due to limited hand motion. We have developed PINATA (Pointing Interaction Notifications and AdapTAtions), a Chrome-based plugin that adapts a browser’s content to assist users with pointing problems and helps them become more aware of their pointing difficulties by providing feedback via customizable notifications.

PINATA was first developed in 2015 and initial work focused on assessment and visualization. Over the summer I extended these features, but focused on supporting a new adaptation. Oftentimes hyperlinks to websites can be difficult to target and click due its -text’s size, color, and/or boldness. To combat this issue, I developed a new adaptation that detects when the user is experiencing difficulty in clicking on a hyperlink, manipulates its text to make it stand out, then offers a hotkey on the keyboard that can be pressed to take the user to the desired link.

PINATA is a work in progress that helps users who face pointing problems by allowing them to navigate the Internet with ease. In the future, the new adaptation should be evaluated to learn of its usefulness to users by conducting user studies. PINATA could also be extended in several ways: customization of the newest adaptation, redesign of the notification manager so that all clickable elements can be easily pressed, and added touch-screen functionality.

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CONDUCTIVE YARN-LIKE THREAD FOR WIRELESS WOUND SENSING

Taylor Patrick¹, Meera Punjiya², Dr. Sameer Sonkusale²
¹Department of Computer Science and Electrical Engineering, Univ. of Maryland - Baltimore County, 1000 Hilltop Cir, Baltimore, MD, 21250
²Department of Electrical and Computer Engineering, Tufts Univ., 419 Boston Ave, Medford, MA 02155

Chronic wounds affect up to 6.5 million individuals in the United States, and are the source of a growing economic burden, estimated to cost $2 billion annually in lost wages and work days. Thread-based sensors have recently been demonstrated for continuous, non-invasive chronic wound monitoring. When combined with wireless technologies, these sensors enable chronic wound healing to be monitored continuously and remotely by a health care professions which may help to improve wound health management practices. Here, thread-based sensors are used in conjunction with a microcontroller and Bluetooth module for demonstration of a system capable of remotely monitoring wound area and closure. Thick yarn-like threads were coated in a carbon polymer and were readout using an Ohmmeter to determine wound area. Use of an alginate/carbon hydrogel coating was explored, but deemed too brittle for long-term use. Base resistance measurements of individual threads were found to be between 100 - 200 Ω. For initial characterization, thread resistance measurements were taken as various volumes of DI water were pipetted onto the thread sensors to mimic wound exudate. These sensitivity measurements were used as a basis for the readout circuitry design. For demonstration of the final system, threads are sewn in a grid-like arrangement into a commercial bandage and placed over an alginate hydrogel as a wound model. Data is acquired using the designed circuitry and is transmitted wirelessly to a mobile device.

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OPTIMIZING AND AUTOMATING THE PHOSPHORUS EXTRACTION AND RECOVERY SYSTEM (PEARS)

Samina Musa¹, Charles Portner¹, Utsav Shahsvatt¹, Josh Benoit¹, Lee Blaney¹

¹Department Chemical, Biochemical & Environmental Engineering, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

The high phosphorus content of agricultural runoff, such as that generated on poultry farms, contributes to eutrophication of surface waters, disrupting environmental processes and decreasing biodiversity in affected ecosystems. By reframing “pollution prevention” as “resource recovery”, we can reduce or reverse eutrophication, while also fostering a closed-loop nutrient management system for agriculture. In this project, we reduced nutrient concentrations in poultry litter by developing a fully automated Phosphorus Extraction and Recovery System (PEARS), which was designed to extract phosphorus, nitrogen, and magnesium from poultry litter and to recover a high-value product, struvite. The laboratory-scale PEARS reactor consists of three tanks in series, and the system was fully automated to allow mixing, fluid transfer, and acid/base addition. A Matlab-based electronics interface was used for feedback control and adjustment of fluid levels, solids/struvite removal, reactant loading, and pH adjustment. The overall run-time of the PEARS operation was approximately 60 minutes. During operation, a 30-L tank was loaded with a 40-80 g/L poultry litter slurry and vigorously mixed. Then, the slurry was transferred to the extraction reactor, where phosphorus was extracted from the slurry at pH 4.5-5.5 after bubbling CO₂(g) and adding 1 M HCl. Solid-liquid separation was achieved through gravity settling, and the supernatant was transferred to the precipitation reactor. The nutrient-rich extract was aerated and dosed with 1 M NaOH to increase the pH to 8.0-9.0. These conditions resulted in production of struvite. In preliminary trials, we were able to extract 66% of the phosphorus from the poultry litter, and we recovered 54% of the extracted phosphorus in the form of struvite. Based on these findings, the PEARS reactor can be deployed on Maryland poultry farms to help ameliorate nutrient pollution.

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Ribosomes are essential to life, because they synthesize the proteins found in all biological cells. Past studies have advanced the understanding of the structure and function of the ribosome, but the biogenesis of the ribosome is not fully understood. Ribosomal ribonucleic acid (rRNA) and ribosomal proteins (rps) join to form the ribosome, but the order and position of these interactions is yet to be completely understood. Studies of bacterial ribosomes have shown that rp binding to rRNA is hierarchical and requires ribosomal assembly factors. Studies done mainly with the yeast Saccharomyces cerevisiae (S. cerevisiae) have shown that this hierarchical binding phenomenon also holds true for eukaryotes, but the assembly process in eukaryotes is far more complex. Previous investigations of ribosomal assembly in eukaryotes were performed under biological stress—conditions of depleted rps or ribosomal assembly factors. This study seeks to develop a method for the isolation of precursor ribosomal particles at different stages of stress-free assembly, without any disruption in or to the process. By inducing the synthesis of tagged rps, then immune-precipitating ribosomal complexes (harboring the tagged proteins using antibodies against the protein tag), the analysis of the rRNA and proteins found in the immune-precipitated particles show great promise for analyzing the assembly of eukaryotic rps into ribosomal precursor particle under stress-free conditions. In addition, current quantitative optimizations of these results will ensure the certainty of these results.

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CHARACTERIZATION OF THE MOLECULAR MECHANISM FOR MATURATION INHIBITORS AGAINST THE HIV-1 CAPSID-SP1 DOMAIN

Carly A. Sciandra\textsuperscript{1}, Emily Cannistraci\textsuperscript{1}, Pengfei Ding\textsuperscript{1}, Eric O. Freed\textsuperscript{2}, Michael F. Summers\textsuperscript{1}

\textsuperscript{1}Department of Chemistry & Biochemistry, Howard Hughes Medical Institute, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21052
\textsuperscript{2}Virus-Cell Interaction Section, HIV Dynamics and Replication Program, Center for Cancer Research, National Cancer Institute, Frederick, MD 21702

HIV/AIDS is a global pandemic and infects roughly 40 million people worldwide. Current therapies target fusion, reverse transcription, and integration processes, but serve only to minimize the effects of the disease. Only the maturation process is targeted in the late phase of the HIV replication cycle, which involves proteolytic cleavage of the Gag polyprotein. During viral assembly, thousands of Gag molecules form a hexagonal lattice, while the CA-SP1 junction region transitions from a random coil to a helical structure in order to assemble into a 6-helix bundle. This CA-SP1 junction is cleaved last during viral maturation, and previous work has suggested that maturation inhibitors prevent the cleavage of the CA-SP1 junction by targeting and stabilizing the 6-helix bundle structure. The 6-helix bundle is only formed in assembled Gag molecules, hindering the solution study of its interaction with maturation inhibitors. Meanwhile, efforts to crystallize the assembled CA-SP1/inhibitor complex have not been successful. Thus, the molecular mechanism for the inhibitory function of these compounds still remains elusive. This work aims to provide insight to the mechanism of maturation inhibition, by utilizing a hexameric scaffolding protein to construct an isolated CA-SP1 junction hexamer. This protein will facilitate probing into the binding stoichiometry and binding affinity of maturation inhibitors, exemplified by Bevirimat, to the hexameric protein by isothermal titration calorimetry (ITC) and solution nuclear magnetic resonance (NMR). Once the binding ratio is better understood, NMR structural studies can be performed to probe the binding mechanism of maturation inhibitors to the CA-SP1 junction helix.

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MODELING THE AUTONOMIC NERVOUS SYSTEM AND CARDIOVASCULAR SYSTEM TO CLASSIFY BEHAVIORAL PERFORMANCE

Theresa Sheets\(^1\), Kathleen Hoffman, Ph.D.\(^1\), Justin Brooks, M.D., Ph.D.\(^2\)

\(^1\)Department of Mathematics, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250

\(^2\)Army Research Laboratory, Human Research and Engineering Directorate, 459 Mulberry Point Road, Aberdeen Proving Ground, Maryland, 21005

Distinguishing features of individuals who are higher performers in fatigue states is important for many situations, such as long distance driving and cybersecurity monitoring. Many methods have been proposed to identify individuals who are likely to maintain high performance despite mental fatigue. These methods have largely focused on quantifying physiologic measures of autonomic nervous system (ANS) activity as determined by heart rate variability (HRV) to identify high and low performers. Success with this approach has been limited, principally because HRV measures are non-specific and limited in their treatment of the cardiovascular system. In this project, we model the cardiovascular system as a dynamical system, then extract ANS activations from this system and use those activation patterns to classify high and low performers.

In our analysis, we examined data from eight participants who performed a cognitive depletion task for 16-20 minutes followed by a Stroop task. Participants were categorized into two groups, high and low performers, based on their performance in the Stroop task. Heart rate (HR) and blood pressure (BP) were collected during the experiment and an established system of equations modeling the cardiovascular system was fit to this data to gather parameters which correspond to parasympathetic control of HR, sympathetic control of HR and sympathetic effect on arterial resistance. Windowing methods were used to develop a time series of these parameters and subsequently utilized to classify participants into two groups. These groups were then compared to the high and low performer classifications.

Results indicate classification accuracy greater than either chance or previously proposed methods. Further analysis reveals that the sympathetic effect on arterial resistance may be more significant for distinguishing performers than previously predicted. These equations may serve as valid ways to estimate parameters of significance to autonomic regulation of the cardiovascular system and psychological function.
OPTIMIZATION OF PHOSPHOPEPTIDE ENRICHMENT OF β-CASEIN

Grace Smith, Lan Ly, Joshua Wilhide, William LaCourse
Molecular Characterization and Analysis Complex, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore MD 21250

Phosphorylation is a post-translational modification (PTM) to a protein by which phosphates are added to the structure of the polypeptide. Phosphorylation regulates the activation of existing proteins and subsequently is fundamental for ideal physiological performance. The identification of the sites where phosphates bind is difficult without the presence of an enrichment protocol to increase the abundance of phosphoproteins. In this study, titanium dioxide (TiO₂) is used as an enrichment tool to identify all the known phosphate sites located on Beta Casein (β-Casein). This protein is used for transporting materials such as calcium, phosphorus, and essential minerals that are crucial for building bodily tissues, muscles, and bones. It is also chosen as a model protein because it has only two sites of phosphorylation and its molecular weight of 24kDa makes it a midpoint for protein analysis. The goal of this research is to optimize the capture step of an existing protocol. Ultimately, this work will be combined with concurrent research with the goal of developing an optimized protocols for the capture of proteins of different lengths.

The enrichment is first performed through the use of a TiO₂ ziptip (ThermoFisher, A32993). Parameters of the capture step are varied to find optimal conditions. After capture, the sample is digested according to the protocol. The sample is then analyzed using high performance liquid chromatography (HPLC) coupled to a high resolution mass spectrometer (HRMS), allowing for high mass accuracy detection. The data is analyzed using PEAKS 7 software, providing sequence coverage and allowing for identification of the phosphorylated sites. The sequence coverage of each sample is analyzed to determine which procedure produces the highest sequence coverage and thus is determined to be optimal. Future research includes continued optimization of established protein analysis protocols to create a set of robust techniques for protein analysis.

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THE ROLE OF Mg2+ IN STABILIZING tRNA:MA INTERACTIONS

Emre Tkacik1, Phoebe Somani1, Amalia Rivera-Oven1, Matthew McDonough1,2, Noel Getachew1, Christy Gaines1, Dr. Michael Summers1

1Department of Chemistry and Biochemistry, Howard Hughes Medical Institute at the University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
2Department of Chemistry and Physics, Western Carolina University, 1 University Way, Cullowhee, NC 28723

Human immunodeficiency virus-1’s (HIV-1) matrix domain (MA) of the Gag polyprotein targets Gag to the cell membrane through interactions between MA’s highly basic region and phosphatidylinositol 4,5-bisphosphate [PI(4,5)P2] on the cell membrane. Targeting to the plasma membrane is critical for the assembly of the virus. In addition to interacting with the plasma membrane, other studies have shown that MA specifically binds to certain tRNAs, including tRNA\textsubscript{Lys} and tRNA\textsubscript{GlycGCC}, \textit{in vivo}. Host tRNA\textsubscript{Lys} acts as a primer for reverse transcription and is packaged into budding virions. Mg\textsuperscript{2+} is crucial for stabilizing the tertiary structure of tRNA, which will allow for the molecule to bind to matrix in vitro. However, when using NMR, increasing the Mg\textsuperscript{2+} concentration in our sample causes peaks to broaden and disappear, making the spectra difficult to interpret. By optimizing conditions that maximize the tightness of binding while minimizing the amount of Mg\textsuperscript{2+} needed for successful folding using the ITC, we will be able to determine the best conditions for further structural studies with the NMR. The studies confirmed that both tRNA\textsubscript{GlycGCC} and tRNA\textsubscript{Lys} form a complex \textit{in vitro} and that increasing the amount of Mg\textsuperscript{2+} strengthens the tRNA:MA interaction. Additionally, we discovered that at a given Mg\textsuperscript{2+} concentration, tRNA\textsubscript{Lys} binds more tightly than tRNA\textsubscript{GlycGCC}. If we can characterize the structure of the MA: tRNA complex using NMR, it would allow for better understanding of the mechanism by which HIV specifically targets the cell membrane and how viral assembly and budding occur.

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VAN-GOGH-LIKE 2, FRIZZLED, KNYPEK, and N-CADHERIN CONTROL DISTINCT ASPECTS OF POLARIZED CELLULAR MIGRATION DURING NEURAL CONVERGENCE

Karndeep Singh, Sharlene Brown, Jonathan Werner, Eudorah Vital, Stephanie Sanchez, Rebecca McFarland, PhD, and Rachel Brewster, PhD.
Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

The neural tube, the precursor of the central nervous system, arises from morphogenetic movements that shape the neural plate into a tubular structure. Narrowing of the neural plate or neural convergence (NC) is mediated in part by the planar cell polarity (PCP) pathway, a branched pathway that includes the glypican Knypek (Knyfr6), Van gogh-like 2 (Vangl2vu67), an integral membrane protein, and the cell surface receptor Frizzled 7a/7b (Fzd7a-e3; Fzd7bhu3495). Evidence from the literature and our laboratory suggests that NC in zebrafish requires elongation and midline-directed polarized migration of neural plate cells. Delayed NC can result in severe neural tube defects (NTDs), which have been observed in all vertebrates studied. Although the PCP pathway is associated with NTDs in several model organisms and humans, the underlying neural cell behaviors remain elusive. To investigate the cellular function of the PCP pathway in NC, we analyzed zebrafish lines carrying null mutations in Knyfr6, Vangl2vu67, and Fzd7a-e3; Fzd7bhu3495. We confirmed that all PCP mutations delay NC by measuring the width of neural plate labeled via in situ hybridization with lateral neural ectoderm markers. Next, we performed a comparative analysis of cell elongation and membrane dynamics in these embryos. Using time-lapse confocal imaging, we showed that wild type neural plate cells elongate and medially restrict membrane protrusions, thereby migrating efficiently towards the midline. In contrast, cells in all mutants failed to elongate initially and extended randomized protrusions, resulting in delayed NC. To address whether these abnormal cells behaviors are caused by defective cell adhesion, we are currently performing several assays that will indicate whether PCP components genetically interact with the neural cell-cell adhesion molecule N-cadherin (N-Cad), reveal whether the levels of N-Cad are reduced in PCP mutants and measure the level of adhesiveness of PCP mutant cells relative to controls.

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OVEREXPRESSION OF CARBON CONCENTRATION MECHANISM GENES CAH1 AND LCI-1 IN CHLAMYDOMONAS REINHARDTII FOR INCREASED ALGAL GROWTH AND OIL PRODUCTION

Newton Mayaka1, Kimuel Villanova1, Lian Jackson1, Amrita Madabushi1, Stephen M. Miller2
1 Biotechnology Program, Baltimore City Community College, 2901 Liberty Heights Ave, Baltimore, MD 21215
2 Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

High fossil fuel consumption and global warming concerns have encouraged exploration of renewable resources, including biofuels produced by algae. A major problem with algae as a source of biofuels is the relatively low amount of oil that algae can produce under optimal conditions. Most efforts towards improving algae as a fuel source are focused on increasing the output and decreasing the generation time of algae growth. Here we attempt to increase the growth rate of the model green alga Chlamydomonas Reinhardti for overexpression of two genes related to the Carbon Concentrating Mechanism (CCM). The focus here is to use molecular cloning and genetic transformation methods to overexpress LCI-1 (Low Carbon Inducible Membrane Protein) and CAH1 (Carbonic Anhydrase 1), which are responsible for carbon dioxide uptake from the environment into the cell. LCI-1 is located within the plasma membrane and CAH1 is located in the periplasmic space between the cell wall and plasma membrane. Accumulation of both LCI1 and CAH1 is induced by low carbon dioxide concentrations. Our cloning strategy involves ligating into existing vectors containing LCI1 and CAH1 coding sequence, a bleomycin selectable marker gene that is linked to a viral 2A peptide sequence, to ensure production of our CCM proteins. These plasmids will be transformed into Chlamydomonas reinhardtii and transformants will be tested by western blot for CCM protein accumulation. Transformant growth rates will be analyzed in a multi-cultivator that determines culture OD at regular intervals. If our strategy for improving growth is successful, it could be implemented in related algae better suited for large-scale industrial production of biofuels.

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Human immunodeficiency virus type 1 (HIV-1) has become an epidemic where millions of people are infected worldwide. During the HIV life cycle, the viral assembly is initiated by interactions between the unspliced viral genomic RNA and its translated product, the Gag polyprotein. Although there is a large excess of non-viral RNA, Gag protein is able to efficiently package the viral genomic RNA. We seek to understand the molecular mechanism underlying the selective genome packaging for HIV-1. Gag contains three structured domains: Matrix (MA), Capsid (CA), and Nucleocapsid (NC). The NC domain of Gag binds to the 5’-leader (5’-L) within the dimeric HIV-1 genome for selective packaging, while the CA domain mediates Gag-Gag interaction resulting in the hexagonal Gag shell that encapsulates the viral RNA. The basic unit of the immature viral shell is Gag hexamer. Mutations at the hexameric interface cause a significant reduction in Gag’s selectivity towards dimeric viral RNA.

We hypothesize that the hexameric structure of the CA domain contributes to the RNA genome selection. Using Nuclear Magnetic Resonance (NMR) and Isothermal Titration Calorimetry (ITC), about sixteen binding sites for multiple Gag proteins on the 5’-L have been identified and mapped, which promote the formation of the Gag hexamer. This functions as the nucleation site to recruit more Gag proteins. With RNA and protein constructs, to obtain structural information needed, chemical crosslinking, engineered disulfide bonds and cryo-electron microscopy are utilized to probe the formation of Gag hexamer. Based on results, RNA is needed to promote the formation of the Gag hexamer. We aim to solve this protein-RNA initiation complex which will provide the detailed molecular mechanism for selective genome packaging. These studies will further our understanding of the mechanism of HIV genome selection, a stage in the life cycle that can be targeted with therapeutics.

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CHARACTERIZING THE DYNAMIC CAPSID-SP1 JUNCTION HELIX OF THE HIV-1 GAG POLYPROTEIN AND THE MECHANISMS OF MATURATION INHIBITORS

Emily Cannistraci¹, Carly Sciandra¹, Pengfei Ding¹, Eric O. Freed², Michael F. Summers¹
¹Department of Chemistry & Biochemistry, Howard Hughes Medical Institute, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore MD 21250
²Virus-Cell Interaction Section, HIV Dynamics and Replication Program, Center for Cancer Research, National Cancer Institute, Frederick, MD 21702

The Human Immunodeficiency Virus (HIV) is a global pandemic that has affected an estimated 36.7 million people worldwide. After HIV integrates its genome into the host cell’s genome, viral RNA and the subsequent Gag polyprotein are produced. The Gag polyprotein is composed of multiple domains including the capsid linked to the SP1 peptide (CA-SP1). Preceding assembly, immature CA-SP1 exhibits a random coil conformation. During assembly, CA-SP1 forms a 6-helix bundle facilitating the formation of Gag hexamers, which rapidly oligomerizes into a highly ordered hexagonal lattice. In order to become an infectious virion, the virus must undergo a maturation process to proteolytically cleave the individual domains of Gag. The rate limiting step of the maturation process is the final cleavage between CA-SP1, for which the 6-helix bundle must be disassembled.

A new class of antiretroviral HIV-1 drugs known as Maturation Inhibitors (MIs), such as Bevirimat, are known to interact with the CA-SP1 junction helix region and prevent cleavage; however, the working mechanism of Bevirimat’s inhibitory function remains elusive. Previous studies hypothesized that MIs stabilize the 6-helix bundle and consequently prevent access to the proteolytic cleavage site by viral protease. This hypothesis remains unconfirmed because the dynamic CA-SP1 junction helix region has yet to be characterized in solution due to the rapid formation of the hexagonal lattice. In this study, a single hexamer will be isolated using a scaffolding protein in order to probe the dynamic properties of the CA-SP1 junction helix by Nuclear Magnetic Resonance (NMR). A NMR solution titration experiment with Bevirimat will provide a better understanding of the interactions between this compound and the CA-SP1 region. Characterizing the CA-SP1 junction helix in solution will assist in elucidating the mechanisms of Bevirimat, thus allowing the development of more potent MIs against multiple strains of HIV-1.

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OVEREXPRESSION OF TWO CARBONIC ANHYDRASE ENZYMES TO AMPLIFY ALGAL CELL GROWTH

Charity Ensor¹, Sarah Carpe¹, Lian Jackson¹, Amrita Madabushi¹, Stephen M. Miller²
¹Arts and Sciences, Biotechnology Program, Baltimore City Community College, 2901 Liberty Heights Ave, Baltimore, MD 21215
²Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

Green algae are photosynthetic plant-like organisms that have great promise as a source of sustainable biofuels. Production of biofuels from algae is a sustainable alternative to fossil fuels and is potentially more economical than ethanol produced from corn, another alternative fuel source. Chlamydomonas reinhardtii, a single celled microalga, is the most widely used model organism for algal biofuel production research. Many tools are available for molecular genetic manipulation of C. reinhardtii, and it is easy to culture, making it an excellent platform for biotechnology research. The limiting factor for algal growth, carbon dioxide (CO), is the focus of much algal biofuels research. It is believed that certain enzymes involved in carbon dioxide uptake into cells for conversion to carbohydrates and eventually lipids, can be manipulated to improve photosynthesis and growth. The focus of this study are two genes that encode enzymes involved in CO₂ uptake, CAH6 and CAH3. Carbonic anhydrase 6 (CAH6) converts CO₂ into carbonate in the chloroplast stroma, increasing carbon flow into the pyrenoid, where CO₂ fixation into carbohydrate takes place. Carbonic anhydrase 3 (CAH3) converts carbonate back into CO₂ in the pyrenoid. Our prediction is that over expression of these enzymes should improve photosynthesis and thus cell growth. Utilizing recombinant DNA technology, we are generating vectors with CAH6 or CAH3 coding regions connected to ble gene sequence (encodes a selectable marker protein), with the hope of increasing expression of these proteins. We will electroporate these vectors into C. reinhardtii then use western-blot analysis to determine transgenic protein expression. Finally, we will use an algal multicstellator to observe and compare growth of our transformants to a control strain. If overexpression of CAH3 and CAH6 improves growth in C. reinhardtii, then our methods can be applied to other algal species, such as Chlorella vulagris, a biotechnology production organism.

Acknowledgement(s): These results were obtained as part of the Research Experience and Mentoring (REM) program in the Department of Biological Sciences at the University of Maryland Baltimore County. This program is funded by a grant (REM supplement to NSF-EFRI-1332344) from the National Foundation (NSF) Directorate for Engineering (ENG) Office of Emerging Frontiers in Research and Innovation (EFRI). We also thank Jose Ortega, Rudolph Park, Erin Pueblo, Jacqueline Rivera, Holly Asbury, Jihye Yeon, Newton Bosire, and Kimuel Villanova for technical assistance and helpful discussions.
FOOD INFECTIONS WITH *LISTERIA MONOCYTOGENES*

Loran Margevich\(^1\), Kavneet Chahil\(^1\), Tonya M. Santaus\(^1,2\), Dr. Chris D. Geddes\(^1,2\)

\(^1\)Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

\(^2\)The Institute of Fluorescence, Columbus Center, University of Maryland, Baltimore County, 701 E. Pratt Street, Baltimore, MD, USA 21202.

This experiment aims to analyze DNA extraction from various foods infected with *Listeria monocytogenes* (Listeria) such as shredded cheese, spinach, cantaloupe, ground beef, and hot dog, through conventional heating and microwave irradiation. Listeria DNA and proteins will be extracted and detected through Ethidium Bromide gel electrophoresis and Ruby stained sodium dodecylsulfate gel electrophoresis (SDS PAGE).

To carry out the experiment, ground foods were infected with 10\(^8\) colony forming units (cfu) concentration of *Listeria monocytogenes*, and incubated for 24, 48, and 72 hours respectively. After each period, the foods were either conventionally heated (boiled) or microwave irradiated with and without the use of microscope slides containing gold bow-ties at varying powers and times. Conventionally heated foods were analyzed from 40-80\(^\circ\)C, and microwaved foods from 10\% power to 50\% power for 60 seconds. The gold bow-tie triangle lysing slides are designed to increase bacterial cellular lysing, DNA fragmentation, and protein degradation. After ethanol precipitation and DNA rehydration, ethidium bromide gels were run to analyze DNA fragmentation. A collaborator at the University of Maryland, Medical Center performed Polymerase Chain Reaction (PCR) to determine if Listeria can be detected from infected foods. SDS PAGE was also run to analyze protein degradation.

The ethidium bromide gels display smearing and banding from combined food and Listeria DNA demonstrating the ability of DNA to be extracted from conventionally heated and microwave irradiated samples. The protein gels show smearing as the temperature and power increases with and without the gold bow-tie structures. It can be analyzed that the use of gold bow-ties aid in the extraction of DNA and proteins from Listeria contaminated food products and can be detected on the PCR platform.

We would like to acknowledge Dr. Chris D. Geddes, the Institute of Fluorescence, UMBC, and collaborators for their support.
EXAMINING THE ELECTRICAL EXCITATION, CALCIUM SIGNALING, AND MECHANICAL CONTRACTION CYCLE OF A HEART CELL

Wesley Collins¹, Kristen Deetz², Nygel Foster³, Darius Leftwich³, Chad Meyer⁴, Shalin Patel⁵
Graduate assistant: Carlos Barajas¹, Faculty mentor: Matthias K. Gobbert¹,
Client: Zana Coulibaly⁶

¹Department of Mathematics and Statistics, UMBC
²Department of Mathematics, Eastern University,
³Departments of Computer Science and Electrical Engineering, UMBC
⁴Department of Mathematics, University of Pittsburgh,
⁵Department of Computer Science and Mathematical Science, Kean University
⁶Department of Pharmacology, University of California, Davis

As the leading cause of death in the United States, heart disease has become a principal concern in modern society. Cardiac arrhythmias can be caused by a dysregulation of calcium dynamics in cardiomyocytes. Calcium dysregulation, however, is not yet fully understood and is not easily predicted; this provides motivation for the subsequent research. Excitation-contraction coupling (ECC) is the process through which cardiomyocytes undergo contraction from an action potential. Calcium induced calcium release (CICR) is the mechanism through which electrical excitation, is coupled with mechanical contraction through calcium signaling. The study of the interplay between electrical excitation, calcium signaling, and mechanical contraction has the potential to better our understanding of the regular functioning of the cardiomyocytes and help us understand how any dysregulation can lead to potential cardiac arrhythmias.

ECC, of which CICR is an important part, can be modeled using a system of partial differential equations that link the electrical excitation, calcium signaling, and mechanical contraction components of a cardiomyocyte. We extend a previous model to implement a seven variable model that includes the mechanical component of the ECC. We conduct a parameter study to determine how the interaction of electrical and calcium systems can impact the cardiomyocyte’s levels of contraction.

Acknowledgements: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-authors Wesley Collins, Nygel Foster, and Darius Leftwich were supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Carlos Barajas was supported by UMBC.
THE EFFECT OF YAP PROTEIN EXPRESSION ON CELL PROLIFERATION

Eliana Crentsil134, Nicolas Perez13, Nash Rochman23, Sean Sun23, Denis Wirtz13
1 Department of Chemical and Biomolecular Engineering, Johns Hopkins University, 3400 North Charles Street, Baltimore, MD 21218
2 Department of Mechanical Engineering, Johns Hopkins University, 3400 North Charles Street, Baltimore, MD 21218
3 Johns Hopkins Institute for NanoBioTechnology, 3400 North Charles Street, Baltimore, MD 21218
4 Department of Chemical, Biochemical, and Environmental Engineering, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

The Yes-Associated Protein (YAP), a proto-oncogene in the Hippo Pathway, has been shown to increase cell proliferation and promote tumorigenesis; however, the mechanism by which YAP increases cell proliferation is unclear. To examine a potential mechanism by which YAP expression is associated with shortening of the phases of the cell cycle. To evaluate whether YAP expression is associated with shortening of the phases of the cell cycle, Cycler experiments were performed to construct cell cycle trajectories. In the Cycler experiments, 3T3 cells and Mesenchymal stem cells were stained via an immunofluorescence assay and imaged with epifluorescence microscopy. The relationship between YAP expression and the DNA distribution, and, in turn, the distribution of the phases of the cell cycle was used to evaluate whether YAP expression increases cell proliferation. Our preliminary data indicate that while there was more uniform YAP expression in the 3T3 cells, YAP expression in the Mesenchymal stem cells was variable. Additionally, trends in the YAP expression within the phases of the cell cycle in 3T3 cells suggest a higher cell cycle dependency on YAP expression. With respect to the distribution of the phases of the cell cycle, the cell cycle trajectories showed that the 3T3 cells had a longer S phase than G1 and G2 phases and a longer G1 to S transition than S to G2 transition. MSC cells showed a longer G1 and G2 phase than S phase. In conclusion, our preliminary data suggest YAP expression may be associated with the distribution of the G1, S, and G2 phases of the cell cycle and the transition between these phases.

References:

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QUANTIFYING VARIABILITY OF BASEFLOW OF WATERSHEDS FOR THE CHESAPEAKE BAY

REU Site: Interdisciplinary Program in High Performance Computing

Christian Dixon\textsuperscript{1}, Gabriel Martinez\textsuperscript{2}, Hwan Hee Park\textsuperscript{3}, Maddie Rainey\textsuperscript{4},

Graduate assistants: Nadeesri Wijekoon\textsuperscript{1} and Sai Popuri\textsuperscript{1},

Faculty mentor: Kofi Adragni\textsuperscript{1},

Client: Jeff P. Raffensperger.\textsuperscript{5}

\textsuperscript{1}Department of Mathematics and Statistics, UMBC
\textsuperscript{2}Department of Mathematics, California State University, Fullerton
\textsuperscript{3}Department of Computational & Applied Math, Rice University
\textsuperscript{4}Department of Mathematics, Grand Valley State University
\textsuperscript{5}United States Geological Survey (USGS)

The U.S. Geological Survey National Water-Quality Assessment Project conducted a study of 225 sites in the Chesapeake Bay watershed to estimate baseflow. Baseflow is the estimated volumetric discharge of water, primarily from groundwater sources, that is relayed to the measurement sites. The study is necessary to address the Nation's water supply for changes in the environment. Baseflow is estimated using a recursive digital filter. Calculating the variability of baseflow water discharge is important to make informed decisions about water regulation. We explore the estimation of variability of the baseflow using two methods: the bootstrap method and the delta method.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Author Christian Dixon was supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistants Nadeesri Wijekoon and Sai Popuri were supported by UMBC.
MODELING THE MELANOPSIN PHOTOTRANSDUCTION CASCADE

Sam Giannakoulias1, Dr. Phyllis Robinson2, Dr. Hye Won Kang1, Dr. Kathleen Hoffman1

1 Department of Mathematics, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
2 Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

The mammalian retina contains three photoreceptors, rods, cones, and intrinsically photosensitive retinal ganglion cells (IPRGC’s). IPRGC’s, unlike rods and cones, function primarily in non-image forming vision such as circadian photoentrainment and the pupillary light response. There are five subtypes of IPRGC’s, all of which express melanopsin, a G-protein coupled receptor encoded by the OPN4 gene. It is currently unknown which GPCR pathway each IPRGC subtype functions through. The goal of our study is to use current and electrophysiology data obtained from M1 IPRGC cells, as well as calcium imaging data obtained from melanopsin transfected HEK293 cells, to create mathematical models based on a Gq pathway. First, we wrote a series of chemical reactions describing the Gq activated biochemical cascade. Using the Law of Mass Action, we generated a set of differential equations that were numerically solved using MATLAB. After solving the differential equations, parameter fitting was used to match the numerical solutions to the experimental data. Parameter sensitivity analysis determined the most important parameters within the pathway. The simulation of the model of the Gq pathway in IPRGC’s matched remarkably well with data generated from IPRGC’s in vivo, thus supporting M1 IPRGC’s functioning through the Gq pathway. Similar results were found for the simulations of the Gq pathway in HEK293 cells when compared with the in vitro calcium assay data produced in melanopsin transfected HEK293 cells. Further, the parameters of the model fit to the wild-type HEK293 cell data, also predicted the increased inactivation rate in the presence of over-expressed beta-arrestin, by only increasing the initial conditions in the model to simulate comparable experimental conditions. In the future, we will investigate the signaling pathway found in M2 IPRGC’s as well as incorporate calcium dynamics to further match melanopsin adaptation in M1 IPRGC’s from multi-flash experiments and low-levels of background light.
Carbon nanodots (CND) are spherical carbon nanostructures that have been extensively studied for various fluorescence-based applications, primarily because they have been shown to be non-toxic, photostable, thermostable, and somewhat easily collected. This study seeks to achieve a novel phosphorescent signal through the addition of bromine and iodine to the carbon nanodot structure, therefore employing the heavy atom effect. This emission is sought after because phosphorescence decays on a longer timescale than fluorescence, which opens an alternative non-radiative decay pathway via transfer to ambient, dissolved oxygen. The product of this effect is the generation of highly reactive singlet oxygen, which has been shown in literature to induce cell death for bacterial cells.

Consequently, the basis of this study is to optimize reaction conditions to generate phosphorescent signal through bromination of the carbon nanodots (Br-CND) or iodinated carbon nanodots (I-CND). This study explores the effects of reaction time, reaction temperature, heavy atom addition strategies, and varying pH on the intensity of phosphorescence. Once an optimal method for producing high intensity phosphorescent signals is established, we hope to achieve a structure that will produce singlet oxygen for antibacterial applications.

We would like to acknowledge the Institute of Fluorescence, Dr. Chris Geddes, Rachael Knoblauch, and lab members for all of their support and guidance.
INTEGRATING 3D PRINTING IN PHYSICAL THERAPY PRACTICE

Pavan Konanur¹, Mark Minnis², William Easley² Amy Hurst²
¹Department of Biological Sciences, UMBC,
²Department of Information Systems, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

3D printing is a revolutionary technology that can be leveraged to create customizable and affordable objects. One potential application for 3D printing is to support the creation of customized assistive devices in physical therapy (PT) practice. In collaboration with the University of Maryland, Baltimore’s (UMB) School of Physical Therapy, we are studying how PT students can integrate 3D objects into physical therapy. Traditionally, in PT classrooms, students work with simulated case scenarios to build custom assistive devices, but for this study they experience working with a real patient.

This summer, in cooperation with physical therapy professors and faculty from UMB, physical therapy graduate students were paired with real patients in need of assistive devices. Groups of students were tasked with designing assistive devices to help the patients with their everyday obstacles. Twelve groups (four to five students each) worked clay into a prototype, which was then 3D scanned and printed. Students were then given a survey to gather feedback on designing assistive devices and working with real patients. To analyze the feedback from the students, survey data was first fully transcribed, and then thematically coded and organized into high-level categories. Questions about cost estimates were studied by viewing the central tendencies.

Inspection of the survey data shows that therapists may hold different perspectives (that of the designer vs. patient) when considering the cost of custom 3D printed assistive devices. Findings from this study shed light on how therapists may integrate the use of 3D scanning and printing in a clinical setting. Future studies can explore the implementation of 3D printing into the healthcare system. Implications of this study can be impactful for physical therapy schools, the healthcare system, and patients in need of assistive devices.

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EXPLORING THE RELATIONSHIP BETWEEN NANOPARTICLE SURFACE CHARGE AND COMPLEMENT ACTIVATION AS INDICATED BY PHYSIOLOGICAL DATA

Chimdiya Onwukwe, Erin Lavik
Department of Chemical, Biochemical and Electrical Engineering, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 20720

A part of the innate immune system, the complement system poses an obstacle to the efficacy and safety of intravenously administered hemostatic nanoparticles. The adsorption of proteins onto foreign materials triggers the alternative pathway of the complement system, resulting in complement-activation related pseudo-allergy (CARPA). CARPA is marked by rapid exsanguination and cardiopulmonary malfunction in animal injury models. Previous research in mice and rat models has shown that surface modification with Poly (ethylene-glycol) (PEG), nanoparticle size, and surface charge influence complement activation. However, fundamental differences in the innate immune system of murine and rat models impede the translation of these findings to clinical applications in humans. The CARPA response occurs in humans and pigs at much lower doses than in rat models, highlighting pigs as a better model for nanoparticle-mediated complement activation in humans. This work focuses on analyzing the relationship between molecular properties of polyethylene glycol-polylactic acid (PEG-PLA) nanoparticles and complement activation in a porcine liver injury model. Specifically, the relationship between surface charge and complement indicators such as cardiopulmonary and blood loss data is explored. These data are gathered from a database of pig surgeries performed by the Lavik lab and analyzed using correlation tests.

This investigation was supported by NIH/NIGMS MARC U*STAR T34 GM 008663 National Research Award to UMBC.
Organometallic chemicals, which consist of a metal atom covalently bound to at least one carbon, have unique physicochemical properties that make them widely applicable to many fields. This project focused on the photolysis of organoselenium compounds. Such compounds are commonly introduced into the environment from use in municipal/industrial wastewater (Se), among other sources. Selenomethionine (SEM) is found in wheat, soybeans, and selenium-rich yeast. Ebselen (EBS) is utilized in the medical industry and has potential toxicity in mice and rats. While this information is known, the photolytic transformation of SEM and EBS in the environment are not well understood. To investigate this mechanism, we first mapped the molar extinction coefficients of the compounds. We deconvoluted the apparent molar extinction coefficients to determine their specific extinction coefficients for protonated and deprotonated species. We fit these data to Gaussian models. Then, we measured the time-based photodegradation kinetics using a Rayonet RPR 600 Reactor. The results suggested that indirect photolysis may be more important than direct photolysis for these compounds. For example, following 24 hours of irradiation, SEM showed no transformation, whereas the addition of 5 mg C/L of DOM resulted in 99.9% degradation in 3 hours. Furthermore, EBS exhibited low transformation after 2.5 hours of irradiation with variable pH and initial concentrations. These data will be evaluated to calculate fluence-based rate constants and quantum yields. This information will be used in ongoing studies to construct the fate and toxicity profiles of organometallics in environmental systems.

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Successful proliferation of HIV requires the reliable export of singly spliced and unspliced viral transcripts to create new pathogenic virions. The export of singly spliced and unspliced transcripts from the host nucleus is mediated by the protein-RNA Rev-RRE complex. Binding of HIV Rev proteins to the Rev-response element (RRE) in the HIV genome permits nuclear export of the complex with support of host cell proteins. In the absence of Rev binding, HIV-1 genomes are retained in the nucleus, preventing the complete assembly of new virions.

The goal of this investigation is to elucidate a high-resolution structure for the RRE region of the HIV genome primarily through NMR spectroscopy. Spectra from individual stem fragments, designed from computationally derived predictions of the RRE secondary structure, were compared to that of the complete RRE. This allowed for the assignment of outlying chemical signals, revealing integral characteristics of the RRE’s secondary structure, including two competing four and five stem loop conformations.

Due to the presence of substantial signal overlap in the 2D NMR spectra of the full RRE, chemical shift assignments remained difficult for high density signal regions. Utilizing deuteration and segmental labelling, the quantity of cross peaks can be reduced to alleviate signal crowding and better identify specific regions of the RRE. Targeted mutations to stabilize and select for the 4 stem and 5 stem conformations have also been made to improve current structure data from cryo-EM and NMR spectra.

The structure will reveal insights into the RRE’s ability to recruit multiple Rev proteins and form the export complex. Disruption of this process creates a promising target for future therapy developments.

We would like to acknowledge the Howard Hughes Medical Institute, the Meyerhoff Scholars Program and National Institutes of Health for Grant #1P50 GM103297.
Abdominal fat is stored in subcutaneous (superficial) and peritoneal (around visceral organ) compartments. Excess abdominal adiposity is a risk factor for healthy aging. We used MRI imaging, to classify adipose tissue into subcutaneous and peritoneal compartments and studied their differential effects on other markers of body. We performed these analyses on members of the Old Order Amish (OOA) population isolate who are characterized by uniform living arrangements and high environmental homogeneity. Analyses were performed in 82 participants from the Amish Connectome Project (ACP). ACP uses the Human Connectome project-derived MRI protocol that was expanded with 3D abdominal imaging, liver spectroscopy and multi-shell DWI imaging. We calculated the correlation between subcutaneous and peritoneal fat volumes and the following physiological measurements: age, very low-density lipoproteins, low-density lipoproteins, triglycerides, abdominal muscle, and total cholesterol. Both volumes significantly correlated with age, abdominal muscle volume, and waist circumference. The subcutaneous fat volumes did not correlate with the total cholesterol, very low density lipoproteins (VLDL), low density lipoproteins (LDL), or triglyceride levels (all p>.05). The peritoneal fat volumes significantly correlated with the total cholesterol ($r^2=0.1579$ p= 0.00001), VLDL ($r^2=0.1676$ p= 0.0007), LDL ($r^2=0.1222$ p=0.0006), and triglycerides ($r^2=0.1815$ p=0.00003) (all p<0.05). The correlation for total cholesterol, very low-density lipoproteins, low-density lipoproteins, and triglycerides were all positive. In conclusion, increased fat in the peritoneal compartment is hazardous for healthy aging. Age related increases in the subcutaneous fat compartment did not show significant correlation with other measurements of body health.

Support was received from NIH grants U01MH108148, R01EB015611, P50MH103222, R01DA027680, R01MH085646, T32MH067533, and U54 EB020403, and NSF grants IIS-1302755 and MRI-1531491.
High internal resistance acts as a major barrier in the development of safe Lithium-ion batteries which offer both high capacity and voltage from their battery cathodes. Linking of cathode nanoparticles with single wall carbon nanotubes through bifunctional peptide nanobridges may allow for faster charge/discharge speeds, decreased internal resistance and higher cyclability. Phage display is a combinatorial approach that utilizes M13 bacteriophage that have been engineered to express a random 12 amino acid sequence on one end of each virus. These random sequences can be exposed to inorganic materials like lithium ion battery cathode materials. If there is some specific interaction between the phage and the inorganic material, then the polypeptides responsible for that interaction can be identified through a process of artificial selection. Polypeptides isolated from M13 bacteriophage Phage Display serve as a useful component in a ‘biological toolbox’, functioning to bio-tethering electrodes to other components inside a battery cathode such as conducting carbon nanotubes. In this project, M13 bacteriophages which bind to various battery components will be identified by Phage Display bio-panning and their performance will be investigated in Li-ion coin cells.

This research project is supported by a National Science Foundation Research Experience for Undergraduate (REU) Research award CHE-1460653
Carbon nanodots are fluorescent particles with luminescence in the visible range. These structures are photostable, thermostable, and are inexpensive to produce, providing a competitive alternative to traditional organic and inorganic fluorophores for future applications.

Commonly used synthetic methods for carbon nanodots produce relative low quantum yields, including our unique combustion strategy. To overcome this limitation, metal enhanced fluorescence (MEF) is investigated. It has been shown that carbon dots in close proximity to a metal nanoparticle film demonstrate an increase in fluorescence intensity and a decrease in the radiative decay of the emitted fluorescence. We report here a strategy to deposit elemental silver directly onto carbon nanodots using the light deposition to produce a MEF effect that becomes intrinsic to the nanostructure. We investigated the effects of different reducing agents, varied silver concentrations, different solvents, and reflux effect to deduce the optimal strategy for producing colloidal MEF.

I would like to acknowledge the Institute of Fluorescence, Dr. Chris Geddes, Rachael Knoblauch, and lab members for all of their support and guidance.
ELEPHANT SPECIES IDENTIFICATION FROM IVORY THROUGH POLYMERASE CHAIN REACTION AND SEQUENCING ANALYSIS FOR APPLICATION IN WORKS OF ART

Thomas Burnett¹, Stacy Davis², Claire Scott¹
Tracy Smith¹, Cynthia Wagner¹

¹Department of Biological Sciences, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
²Department of Chemistry, Towson University, 8000 York Road, Towson, MD, 21252

Ivory is a material obtained from the tooth or tusk of an animal. It has been used for centuries for a wide range of applications including medicines, weapons, and art. Species-level identification of ivory works of art is necessary to meet government requirements in the traveling of exhibitions containing ivory, as well as learning origins of a piece, and discovering historical trade routes. One of the largest sources of ivory is elephants, of which there are three major species: African Bush Elephant (*Loxodonta africana*), African Forest Elephant (*Loxodonta cyclotis*), and Asian Elephant (*Elephas maximas*). While these elephants are distinct morphologically and genetically, there is no known means to visually distinguish the ivory. This effort is made more difficult for art pieces, as the ivory has been processed in the artifact’s construction and degraded over time. Our goal was to see if it was possible to differentiate between African and Asian elephant species using extraction, amplification, and sequencing of mitochondrial DNA (mtDNA) from ivory taking into account special needs of works of art. Results were compared to known single nucleotide polymorphisms (SNPs) that exist between elephant species in question. We employed a technique that has been used to distinguish between species in wildlife forensics. The sample size reported for this technique is too large for works of art and would disfigure the appearance of the object. Because of equipment limitations, we were unable to test the suitability of smaller sample sizes. In addition, there are portions of the tusk that may not contain an abundance of DNA, and often there is no means to determine from what part of the tusk the object was carved. Given the current technology, we do not believe that identification of species through genetic testing is a viable option for works of art made from ivory.

This research was funded by the Andrew W. Mellon Foundation. We would like to thank the Department of Chemistry and Dr. Zeev Rosenzweig for hosting the SCIART program, and the Department of Biological Sciences for use of their facilities. We would also like to thank Ms. Terry Weisser and the Walter’s Art Museum for their mentorship, guidance, and donation of ivory samples. Special thanks to The Elephant Sanctuary in Tennessee for generous donation of fecal and blood samples.
COMPARATIVE STRUCTURAL ANALYSIS OF UNMYRISTYLATED HIV-2 MATRIX PROTEIN USING NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY

Hadeel Hamoud1, Nia Starr2, Gabrielle Callwood-Jackson3, Milan Reed2, Janae Baptiste4, Dr. Holly Summers4, Dr. Michael Summers4
1Centennial High School, 4300 Centennial Ln, Ellicott City, MD 21042
2Our Lady of Good Counsel High School, 17301 Old Vic Blvd, Olney, MD 20832
3Eleanor Roosevelt High School, 7601 Hanover Pkwy, Greenbelt, MD 20770
4Department of Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle Baltimore, MD 21250

Human immunodeficiency virus type 2 (HIV-2) is a less virulent form of the human immunodeficiency viruses which produce AIDS. It is hypothesized that this is partially due to the behavior of its myristoyl group attached to the matrix protein at different pH levels. At a lower pH, the myristoyl group of the HIV-2 matrix protein is sequestered preventing membrane targeting which stops the virus from proliferating. This is in contrast to the myristoyl group of HIV-1 matrix protein which is exposed at the lower pH. Our main objective was to clone HIV2-MA into a new plasmid to produce the unmyristoylated form of the protein and compare it with the myristoylated HIV2-MA that was made and previously studied. We are hoping to gain a better understanding of the structure of the protein and gain insight as to why it acts differently compared to its close relative HIV1-MA. The HIV2-MA gene was acquired from the Resh plasmid and PCR amplified. It and the pET19b plasmid where double digested using the restriction enzymes Nco1 and BamH1, purified, and then ligated to form a new plasmid containing only the HIV2-MA gene. Previous plasmids contained Human or Yeast N-myristoyltransferase (NMT) which facilitated myristoylation in E. Coli. We wanted to prevent this with our new construct. After confirming the sequence of our new plasmid, we transformed it into BL21 competent cells for protein expression and grew up the protein in E. coli using minimal media to facilitate N15 labeling. Using nuclear magnetic resonance spectroscopy (NMR), we performed a heteronuclear single quantum coherence (HSQC) experiment to evaluate and compare the structure of this protein to the myristoylated HIV2-MA and to its relative HIV1-MA. These comparisons are ongoing.

Our research would not have been possible without the support of the Summer Biomedical Training Program led by Justine Johnson, Dr. Michael Summers, Dr. Holly Summers and Cindy Finch. We would also like to acknowledge Howard Hughes Medical Institute and NIH/NIGMS (grant # R01 GM42561-28S1) for funding our research.
ARE YOU UP TO THE TUSK?: IDENTIFYING IVORY SPECIES USING RAMAN SPECTROSCOPY AND MASS SPECTROMETRY

Chloe Cao¹, Hope La Farge², Adam Ring³, Terry Drayman-Weisser⁴, Daniel Walter Rowlands²
¹Department of Chemistry, Johns Hopkins University, 3400 N. Charles St., Baltimore, MD 21218
²Department of Chemistry and Biochemistry, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
³Department of Chemistry, Emory University, 1515 Dickey Drive, Atlanta, GA 30322
⁴The Walters Art Museum, 600 N. Charles St., Baltimore, MD 21201

Analytical chemistry research is needed to improve techniques for identifying elephant ivory by origin and species while minimizing sample size. Poaching of African elephants (Loxodonta africana and Loxodonta cyclotis) and Asian elephants (Elephas maximus) is an issue that ultimately threatens their survival; this makes identifying the origin of elephant ivory even more important for not only addressing ivory poaching and trading, but also assisting museums overseeing ivory artifacts.

To combat elephant poaching, enforcement of African elephant ivory regulations under CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora) has intensified. These regulations greatly impact the movement of museum exhibitions, requiring them to identify the ivory’s originating species. While species identification is important for legal documentation, determining the origin of ivory artifacts is crucial for historical, curatorial, and art conservation purposes to achieve a better understanding of its unique context. Culturally significant ivory artifacts pose an additional challenge, as it is imperative to impose minimally invasive sampling techniques.

Bone and antler have a similar composition to ivory, and were included in this study as model systems due to greater availability. This study uses inductively coupled plasma-mass spectrometry (ICP-MS), Raman spectroscopy, and matrix assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF) to differentiate between various species using bone, antler, and ivory. Though results are still in progress, this analytical survey has strong potential to impact methods used to identify ivory and further the current understanding of cultural heritage.

This project is supported by the Baltimore SCIART research experience for undergraduate students, which is a collaborative program between UMBC, Johns Hopkins University, and the Walters Art Museum in Baltimore. The program is funded by the Andrew Mellon Foundation.
DEVELOPMENT OF FAST RECONSTRUCTION TECHNIQUES FOR PROMPT GAMMA IMAGING DURING PROTON RADIOTHERAPY

REU Site: Interdisciplinary Program in High Performance Computing

Johnlemuel Casilag\textsuperscript{1}, James Della-Giustina\textsuperscript{2}, Elizabeth Gregorio\textsuperscript{3}, Aniebiet Jacob\textsuperscript{1},
Graduate assistant: Carlos Barajas\textsuperscript{4}, Faculty mentor: Matthias K. Gobbert\textsuperscript{4},
Client: Jerimy Polf\textsuperscript{5}

\textsuperscript{1}Departments of Computer Science and Electrical Engineering, UMBC
\textsuperscript{2}School of Information Technology & Computer Science, Community College of Baltimore Co.
\textsuperscript{3}Department of Physics, Hamline University
\textsuperscript{4}Department of Mathematics and Statistics, UMBC
\textsuperscript{5}Department of Radiation Oncology, University of Maryland - School of Medicine

Proton beam radiation treatment was first used by Robert Wilson in 1946. The advantage of proton beam radiation is that the lethal dose of radiation is delivered by a sharp increase toward the end of the beam range. This sharp increase is known as the Bragg peak and allows for the possibility of reducing the exposure of healthy tissue to radiation when comparing to x-ray radiation treatment. As the proton beam interacts with the molecules in the body, gamma rays are emitted. The origin of the gamma rays gives the location of the proton beam in the body, therefore, gamma ray imaging allows physicians to better take advantage of the benefits of proton beam radiation.

These gamma rays are detected using a Compton Camera (CC) and the SOE algorithm is used to reconstruct images of these gamma rays as they are emitted from the patient. This imaging occurs while the radiation dose is delivered, allowing physicians to make adjustments in the treatment room. This software is most effective with a resolution of about 1 mm, and a short run time. This project focuses on speeding up the image reconstruction software using a variety of parallel computing techniques involving OpenMP/MPI or hardware like 68-core Intel MIC based CPUs.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-authors Johnlemuel Casilag and Aniebiet Jacob were supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Carlos Barajas was supported by UMBC.
Human immunodeficiency virus type-1 (HIV-1) is a retrovirus that is the causative agent of acquired immunodeficiency syndrome (AIDS). There are approximately 36.7 million people in the world infected with HIV. The viral genome is reverse transcribed which is a highly mutagenic process, however the 5’-Leader of the genome is the most conserved region. The 5’-Leader undergoes a dimerization process exposing more than a dozen nucleocapsid (NC) binding sites and is responsible for promoting packaging. Previous studies on the HIV-1 5’-Leader discovered that the minimal region required for viral genome selective packaging is the Core Encapsidation Signal (CES). Our research investigates the binding interactions between the NC domain of the Gag polyprotein and the CES of the dimeric viral genome. The high resolution nuclear magnetic resonance (NMR) structure of the CES revealed weakly base-paired and unpaired guanines, which are characteristic for NC binding sites. Utilizing techniques such as: electrophoretic mobility shift assays (EMSA), isothermal titration calorimetry (ITC), and mutagenesis, we elucidated the specific guanine residues within the CES responsible for NC binding. Ultimately, gaining a greater understanding of the mechanism for selective packaging of the viral genome could eventually translate into successful development of viral inhibitors.

Acknowledgement:

This research was funded by NIH/NIGMS grants 1P50GM103297 and the Howard Hughes Medical Institute at UMBC. Additional acknowledgment to the Summer Biomedical Training Program (SBTP).
MICROWAVE-ASSISTED LYSING OF LISTERIA MONOCYTGENES AND VIBRIO CHOLERAE

Shannon Cole¹, Amanda Harvey¹, Tonya M. Santaus¹,² and Chris D. Geddes¹,²
¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
²The Institute of Fluorescence, Columbus Center, University of Maryland Baltimore County, 701 E. Pratt Street, Baltimore, MD, USA 21202.

Listeria monocytogenes (Listeria) and Vibrio cholerae (Cholera) are two bacteria that cause detrimental health outcomes for infected individuals. Listeria is spread through contaminated food substances while Cholera is transmitted through contaminated water in areas of poor sanitation. Lysing is the process by which cell membranes are broken apart to release intracellular components such as DNA, which can be used for the subsequent identification of the infecting organism.

In this poster, we analyze microwave powers and times to lyse Listeria monocytogenes and Vibrio cholerae cells using a 900W microwave. We use Polymerase Chain Reaction (PCR) to determine which lysing parameters are optimal in yielding the highest concentration of isolated DNA from the cells.

Listeria and Cholera are mixed separately into deionized (DI) water making a $10^8$ colony forming unit (cfu) concentration. The solution is placed onto a blank slide or a slide with gold triangles and microwaved at various powers and times. The slides with the gold triangles increase the temperature of the solution compared to the blank slides. The solution is also placed onto filter paper with various concentrations. Once the filter paper is dry it is placed into 1mL of DI water and vortexed. The same tests are completed with the filter paper. After the experiments are complete and the microwaved solution has cooled, ethanol precipitation is performed. Next, the ethanol is extracted out and the pellet is left to dry. Lastly, the pellet is hydrated with DI water and sent for PCR.

A collaborator at the University of Maryland, Department of Epidemiology performs the quantitative PCR. The PCR cycle number is used to determine the optimum lysing parameters.

We would like to acknowledge Dr. Geddes, The Institute of Fluorescence, and UMBC for allowing us to conduct research in their facilities.
DISTRIBUTION OF CHEMOATTRACTANTS IN A HETEROGENEOUS TISSUE AND ITS IMPACT ON CELL MIGRATION

REU Site: Interdisciplinary Program in High Performance Computing

Jessica Cooley1, Aaron George2, Zachary Mekus3, Victoria Sabo4,
Graduate assistant: Morgan Strzegowski1, Faculty mentor: Bradford E. Peercy1,
Client: Michelle Starz-Gaiano5

1Department of Mathematics and Statistics, UMBC
2Department of Mathematics, University of Maryland College Park
3Department of Computer Science and Engineering, Washington University in St. Louis
4Department of Mathematics and Statistics, Georgetown University
5Department of Biological Sciences, UMBC

Cell migration is the process in living organisms by which the body heals and diseases spread. Comprehension of this mechanism will be beneficial to understanding its applications, yet little is known about this process. We study the cluster cell migration in the egg chamber of Drosophila melanogaster, or fruit flies, because it is easy to observe and sample in that organism. A previous model simulated the cell cluster's migration using forces to determine movement of many individual cells; we improved and revised this system. We created a more geometrically accurate model of the egg chamber and mapped the diffusion of the chemoattractants through that domain using a reaction diffusion system. In addition, the base implementation is updated to more accurately simulate the cell migration process. This model will allow several questions to be investigated, such as identifying the source and quantity of the chemoattractants, the rate at which they are taken in by other cells in the egg chamber, if at all, and the time needed for them to reach the polar and border cells at the anterior of the chamber that gives the most faithful representation of experimental results.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-author Jessica Cooley was supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Morgan Strzegowski was supported by UMBC.
Water-soluble cadmium telluride quantum dots (CdTe QDs) are utilized in various optoelectronic and biological applications due to their tunable size and optical properties. CdTe QD cores obtained from an aqueous synthesis route produces QDs with excellent water solubility, biological compatibility, and stability. However, aqueous synthesis using the traditional reflux method requires long reaction times that can generate many surface defects, resulting in a low quantum yield. Microwave irradiation methods can be used instead of the traditional reflux methods to rapidly synthesize high quality CdTe QDs. Microwave synthesis uses cheap precursor materials, has fast reactions times, does not require an inert atmosphere, and is environmentally friendly. In our study, CdTe QDs were synthesized by combining cadmium chloride and tellurium dioxide in the presence of 3-mercapropionic acid (MPA) in Millipore water. MPA was used as both a capping ligand and a reducing agent of tellurium dioxide. Reaction time, temperature, pH and the molar ratio of MPA stabilizer to Cd²⁺ were found to affect QD particle size and emission quantum yield. As expected, coating the CdTe QDs with CdS and ZnS shells induced significant red shifts in the absorption and emission maxima and increased the QD emission quantum yield. In the near future, experiments will be conducted to assess the toxicity of CdTe, CdTe/CdS, CdTe/ZnS, and CdTe/CdS/ZnS QDs which are prepared using our microwave-based technique in comparison to the same QDs when prepared using traditional synthesis routes in aqueous solutions and organic solvents, against bacteria and aquatic organisms which are ubiquitous in the environment.

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TRANSCRIPTIONAL REGULATION OF THE HIV-1 RNA GENOME

Frances Grace Ghinger¹, Yash Desai¹, Joshua Brown¹, Michael Summers. PhD¹
¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County,
Baltimore, MD 21250

Human immunodeficiency virus (HIV) uses RNA as its source of genetic material. This RNA can exist as two different conformations, the monomer and the dimer, which ultimately determines the role of the RNA in later steps of the viral life cycle. As a monomer, the RNA will go on to be translated into viral proteins. The dimer, however, is packaged into new HIV virions. The key to understanding how these RNA structures determine function is in the start site heterogeneity (TSS) mechanism. This mechanism describes how manipulating the number of guanines on the viral RNA can cause the native dimer to change into the monomer conformation; therefore, changing the final function of the RNA. Using nuclear magnetic resonance (NMR) spectroscopy and in vivo studies, the existence of TSS, as well as its nature to disrupt the regions around it, has been confirmed. In the monomer structure (Cap 2G, Cap 3G), additional guanosine residues cause remodeling of polyA and extended U5-DIS Interaction. As for the dimer structure (Cap 1G), poly A is stable and DIS is no longer being sequestered thus allowing it to interact with a DIS of another RNA, forming a dimer. Our focus now includes assigning all the NOSEY signals for the Cap-1G dimer conformation to solve the secondary structure.

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Diabetes occurs when the body's blood sugar levels are in a state of sustained elevation. The pancreatic beta cells, organized in the islets of Langerhans, secrete a hormone called insulin that is responsible for maintaining blood glucose at appropriate levels. Oscillations in insulin levels, which are thought to require synchronization in insulin secretion, are necessary for proper regulation of glucose. A loss of this periodic behavior has been observed in type 2 diabetic patients. We used the Single Slow Channel Model to compute the calcium and electrical dynamics during insulin secretion of a single beta cell. To replicate an islet, we coupled the cell cluster according to a hexagonal-close-packed lattice. The existence of small worldness in the islet and its effect on islet performance was investigated by using methods from graph theory. To quantify the performance, we developed a synchronization index from previously used indexes that reflects to what degree the electrical or calcium oscillations are in phase. The effect small worldness has on synchronization is indicative of the existence of hub cells, which have a larger influence on the rhythm of the islet. Thus, the destruction of hub cells within the islet would disrupt its synchronization.
HIV-1 is a retrovirus that infects human immune T-cells. One of the proteins vital for HIV-1 replication is the Gag polyprotein. The N-terminal domain of Gag, matrix, is essential for Gag targeting and binding to the plasma membrane (PM) to start virion assembly. Matrix binds to phosphatidylinositol 4,5 bisphosphate \([\text{PI}(4,5)\text{P}_2]\) in the PM, and may also specifically target lipid rafts. Rafts are rigid, liquid order portions of the PM that are high in cholesterol and certain phospholipids. Matrix also binds to tRNAs in the cell, and many believe this regulates matrix-membrane interactions. According to the current model, tRNA binds to matrix in the cytosol but when this complex reaches the PM, \text{PI}(4,5)\text{P}_2 will outcompete the tRNA and matrix will bind to \text{PI}(4,5)\text{P}_2. To test this theory, we created model membranes (liposomes), representing either raft or non-raft regions of the PM. We used the compositions consistent with earlier experiments in our lab, and also created liposomes using new ratios to mimic MT4 cell PMs and the HIV lipid envelope. For each liposome type we used membranes containing or lacking \text{PI}(4,5)\text{P}_2. We performed 1D-^1\text{H} NMR liposome competition assays in which we observed the interaction between the matrix-tRNA\text{Lys}^3 complex and these different liposomes. With no tRNA present we saw more binding to rafts and liposomes containing \text{PI}(4,5)\text{P}_2, than non-rafts and liposomes lacking \text{PI}(4,5)\text{P}_2. This was expected based on the literature and previous experiments. Interestingly, in the competition assays which involved the tRNA-matrix complex, binding did not change regardless of the type of liposome, indicating that \text{PI}(4,5)\text{P}_2 does not outcompete tRNA\text{Lys}^3 for matrix binding.

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Bovine lameness is a common issue on commercial dairy farms, resulting in decreased productivity. Early detection is essential for effective treatment of lameness. Here and recent previous work, lameness status of a cow has been modeled using three-dimensional limb movement measurements related to its gait. A statistical model uses the software SAS® with its LOGISTIC and TRANSREG procedures. The model produces a binary classification, lame or not lame. Current implementation requires running several SAS® procedures manually and therefore is not amenable to a large scale application and model optimization. In this work, we implement an optimization algorithm in R to mirror the TRANSREG procedure, and thus speed up exploration of a large number of candidate models to optimize the widely used goodness of fit criteria such as the Receiver Operating Characteristic curve (AUC). The models are also evaluated using classification error rates (False Positives and False Negative or equivalently sensitivity and specificity). We also consider multinomial logistic models so that cows may be further classified into three categories: severely lame, mildly lame or sound. These results can be used in the commercial dairy industry for lameness detection.

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INVESTIGATING MICRORNAS IN SIGNALING PATHWAYS THAT CONTROL
BORDER CELL MIGRATION IN DROSOPHILA MELANOGASTER

Joel Hayford\textsuperscript{1,2}, Urooba Ali\textsuperscript{1,3} and Tyra Sellers\textsuperscript{1,4}
Michelle Starz-Gaiano\textsuperscript{1}

\textsuperscript{1}Department of Biological Sciences, University of Maryland, Baltimore County, 1000 Hilltop Circle Road, Baltimore, MD, 21250, USA
\textsuperscript{2}Montgomery College, 51 Mannakee Street, Rockville MD, 20850
\textsuperscript{3}Howard Community College, 10901 Little Patuxent Parkway, Columbia MD, 21044
\textsuperscript{4}Morgan State University, 1700 E. Coldspring Lane, Baltimore MD, 21251

\textit{Drosophila melanogaster} requires Janus kinase (JAK) and Signal transducers and activators of transcription (STAT) signaling to specify migratory cells called border cells and promote their migration in the female ovary. The JAK/STAT pathway is similar to signaling pathways in the human body that are specific to biological processes involved in cell migration and immune function. Mutations in these signaling pathways can cause health problems such as birth defects and metastatic cancers in the human body, and as such we expect our research to be of wide interest. To understand the mechanisms by which signals are converted through the JAK/STAT pathway and regulate transcription factors involved in the specification and migration of border cells, we studied the role of microRNAs (miR-8, miR-252 and miR-284) that are predicted to disrupt the JAK/STAT signaling pathway. MicroRNAs (miRs) regulate gene expression through translational inhibition or cleavage of mRNA molecules. This study focuses on how miR-8, miR-252 and miR-284 may change the phenotype of border cell migration in egg chambers. In order to conduct this study we used transgenic fly lines to determine if overexpression or depletion of the selected miRNAs impacts border cell migration.

We examined egg chambers from our miRNA engineered flies during stages 8-10 of development. We compared an experimental group of flies with its sibling control for each miRNA to identify the differences among border cell phenotypes. For abnormal border cell migration, the percentage of egg chambers affected by the miRNA for each genotype was counted. Some egg chambers with miR-8 overexpression displayed delayed cell migration. From this we can conclude that miR-8 may alter the levels of STAT activity. miR-252 and miR-284 may not have effects on border cell migration. Further experimentation is needed to confirm these conclusions.

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DETERMINING THE STRUCTURE OF THE HIV-1 RNA WITH FRAGMENTATION USING NMR

Tatiana Rodriguez¹, Jana Hijji², Joshua Brown¹, Michael Summers, PhD¹
¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, Baltimore, MD 21250
²University of Maryland-College Park, College Park, MD 20742

The structure of the 5′-Leader (5′-L) of the human immunodeficiency virus (HIV-1) viral RNA genome is under intense study, because of its role in determining the fate of the RNA. To help us determine the structure of the 5′-L, we use nuclear magnetic resonance (NMR) spectroscopy. The NMR spectrometer provides us with the chemical and structural environment of individual hydrogens within the RNA molecule. Using this information we can determine the three-dimensional structure of the HIV-1 5′-L. NMR spectra of large RNAs, such as the 5′-L are often crowded and contain overlapping signals, causing the data to be difficult to interpret. In order to counter this, we prepare smaller oligos that mimic the structural elements of the larger RNA, but whose spectra can be confidently assigned. This technique has allowed us to confirm multiple regions of the full-length leader by overlapping the data of the leader with that of the assigned smaller control oligos. With the help of NMR we have been able to determine the 2-D structure of multiple regions of the monomer and dimer conformations of the 5′-L.

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3D SCREEN BIOPRINTING THE EXTRACELLULAR MATRIX OF THE OUTER RETINA

Bryan Ibarra1, Adam R. Day2, Erin B. Lavik2

1 Biomedical Engineering, University of Miami, 1251 Memorial Dr. Coral Gables, FL 33146
2 Chemical, Biological, and Environmental Engineering, University of Maryland Baltimore County, 1000 Hilltop Rd., Baltimore, MD 21250

3D bioprinting has allowed for more control in therapeutic applications such as: growing tissues in vitro, testing them with treatments for induced diseases and injuries, and surgically implanting them into hosts. Current 3D bioprinting techniques are expensive and incompatible with sensitive cell types such as neural and stem cells, so an alternative technology is needed. A 3D bioprinting technique using screen printing technology is inexpensive can be widely used in essentially any lab to model diseases found in layered tissues. Age-related macular degeneration (AMD) is one such disease that can be effectively modeled thanks to this technique, which can print the retinal pigment epithelium cell layer found in the outer retina. We are prototyping a 3D screen bioprinter that can effectively produce a high throughput model of the outer retina. The scaffold on which these cells grow was made from VS-PEG-PLL copolymer cross-linked with SH-PEG-SH via Michael-type addition. The extracellular matrix of the outer retina was mimicked by loading the gel with laminin and fibronectin to improve cell adhesion and survivability.

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EFFECTS OF SLEEP ON THE IMMUNITY OF *DROSOPHILLA MELANOGASTER*

Decole Russell¹, Chetana Jadhav², Ayodeji Adeleke³, Safiya Ismail⁴, Nina Evans², Jeff Leips²

¹ Anne Arundel Community College, Arnold, MD 21012
² Department of Biology, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
³ The Community College of Baltimore County, Baltimore, MD 21228
⁴ Prince George’s Community College, Largo, MD 20774

*Drosophila melanogaster*, also known as the fruit fly, is commonly used to study human diseases due to the genetic similarities between the two species. Previous studies have pointed to a connection between sleep and immunity, although the genetic basis of this relationship is unknown. Interestingly, there have been conflicting studies on the amount of sleep needed to promote immunity in *Drosophila*. The main goal of this project was to determine if flies genetically programmed for longer sleep patterns survived bacterial infection better than those programmed for shorter sleep. We inoculated 10 genetic lines (n=50 each) of short sleepers and 10 genetic lines (n=50 each) of longer sleepers with the bacterium *Enterococcus faecalis* and measured fly mortality at 24, 48, 72, and 96 hours post inoculation. Our results revealed that short sleepers had an increased mortality over time compared to long sleepers over the course of 72 and 96 hours. At the 72-hour time point, we found there to be a statistically increased mortality in the short sleepers (33.4 +/- 3.70%) compared to the long sleepers. At the 96-hour time point, we found there to be a statistically increased mortality in the short sleepers (51.4 +/- 2.65%) compared to the long sleepers. Our results provide evidence that there is a correlation between sleep duration and immunity. The future goal of this experiment is to identify genes that are responsible for both sleep and immunity that will allow us to understand the connection.

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SCRATCHING THE SURFACE OF BAHAMA ORIOLE PINE FOREST NESTING:
NEW DOCUMENTATION OF CATS AS POTENTIAL PREDATORS OF A
CRITICALLY ENDANGERED SONGBIRD

Mathew Kane1, Daniel Stonko1, Kevin Omland1
1Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

The Bahama Oriole is a critically endangered songbird endemic to the Bahamas that is currently found only on the Andros island complex. The most recent published research concluded that fewer than 300 individuals remain in the shrinking population. Loss of coconut palms due to lethal yellowing disease, brood parasitism by the Shiny Cowbird, and the loss of foraging habitat are the most commonly cited threats to the species. Previous research and population estimates were influenced by the belief that the Bahama Oriole nests almost exclusively in non-native coconut palms within residential and agricultural habitats. However, our recent findings show conclusive evidence of pine forest nesting in both Caribbean Pine and native understory thatch palm. This new understanding of Bahama Oriole habitat usage represents a significant paradigm shift in our understanding of the breeding ecology and population size of the species. Since effective conservation is dependent on thorough and accurate knowledge of the species, the need for further study into the previously overlooked pine forest habitat became clear. Our research aimed to identify the potential predators in the pine forests of our North Andros study area using camera trapping. We documented the presence of feral cats in developed areas as well the most remote parts of the pine forest. These findings are of particular concern given that feral and domesticated cats are well-documented predators of birds and have led to the extinction of more than 30 island-endemic bird species. Moving forward, we will conduct more research on the Bahama Oriole and its pine forest habitat in order to accurately measure the population size, assess the threat of predation, and lead effective conservation efforts.

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A CUSTOMIZABLE OPEN-SOURCE ASSISTIVE TECHNOLOGY FOR COMMUNICATION

Megha Kottapalli\textsuperscript{1}, Sanjay Kumar\textsuperscript{2},
Foad Hamidi\textsuperscript{2}

\textsuperscript{1}Department of Computer Science and Electrical Engineering, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
\textsuperscript{2}Department of Information Systems, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

According to the National Institute on Deafness and Other Communication Disorders, approximately 7.5 million individuals have a speech and voice disorder in the USA. The Assistive Technology Act defines Assistive Technology (AT) as any product, device, or equipment that is used to maintain, increase, or improve the functional capabilities of individuals with disabilities. People with speech disorders can benefit from ATs because they can help them to efficiently communicate and easily customize what they want to say. TalkBox is an open-source communication-board for individuals with communication or speech disorders that can be built and customized by the users. This device incorporates the principles of the do-it-yourself (DIY) movement, such as using open-source technology and sharing information, to create a participatory and accessible approach to AT design.

TalkBox is being developed using a community-engaged design approach where input from stakeholders is incorporated into the design of the interface. Collaboration with local community partners, therapists working with people with disabilities, allowed us to make informed decisions on different aspects of the design from altering the materials to testing the changed prototype. In this phase, we contributed to the project in two ways: (1) by changing the form and functionality of the device (specifically, adding a volume change option and improving the 3D printed casing) and (2) by developing a concise form of documentation for changes that occurred within the design.

We have explored the potential of DIY ATs. Specifically, we have customized TalkBox based on input from users. We improved TalkBox by making it more user-friendly, by altering its form and functionality, and adding easy-to-use documentation. Through this process, we examined how an open-source technology contributes to the development of more inclusive and affordable systems. In the future, similar systems could help create a diverse set of assistive technologies for this population.
36 million people are currently infected with human immunodeficiency virus (HIV), a retrovirus responsible for the onset of the acquired immunodeficiency syndrome (AIDS). Upon transmission, the virus invades CD4+ T cells and integrates its proviral genome into the host genome leading to a life-long infection. During the viral life cycle, interactions between the unspliced viral RNA and its translated product, the Gag polyprotein, initiate the packaging of two copies of the HIV genome. Gag contains three structured domains: Matrix (MA), Capsid (CA), and Nucleocapsid (NC). The NC domain of Gag binds to exposed or weakly base paired guanines of the 5’-leader (5’-L) to initiate genome packaging and viral assembly.

We seek to characterize the Gag-RNA interactions essential to genome packaging using a truncated 5’-L derivative and a hexameric NC protein. We mapped the multiple guanines responsible for NC binding and packaging through mutagenesis. NC binding promotes the formation of Gag hexamers that potentially function as the nucleation site to initiate the viral assembly. Due to hexamer-hexamer interactions in the C-terminal Domain (CTD) of CA, the protein aggregates and precipitates in the presence of RNA. To circumvent these problems, we have fused the NC domain of Gag to a hexameric protein scaffold, which mimics hexameric Gag, while forming isolated hexamers and preventing aggregation. The hexameric NC protein forms a 1:1 complex with the core region of dimeric 5’-L with a very low dissociation constant. With our RNA and protein constructs, we aim to deduce structural information of the Gag-RNA interactions using X-ray crystallography and cryo-electron microscopy. These studies will further our understanding of the mechanism of HIV genome packaging, which can be helpful for target-based therapeutic developments.

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A COMPUTATIONAL FRAMEWORK TO REVERSE-ENGINEER INTRATUMOR SUB-CLONAL DYNAMIC MODELS AND PREDICT OPTIMAL TREATMENT TARGETS

Caroline Larkin, Kyle Pickford, Daniel Lobo
Department of Biological Sciences, University of Maryland, Baltimore County
1000 Hilltop Circle, Baltimore, MD 21250

Cancer is a disease characterized by a variety of sub-clonal tumor cells dynamically interacting among themselves and the surrounding environment. This results in heterogeneous cancer phenotypes that can have increased tumor volumes and growth depending on their sub-clonal compositions. Sub-clonal tumor heterogeneity is a poorly understood aspect of cancer, which can grant new insights for the development of cancer therapeutics. It has been shown that different sub-clones have varying impacts on overall tumor growth and their interactions are essential to maintaining the proliferation of cancer cells. Perturbing certain sub clones can significantly affect the development of the tumor, with results ranging from decreasing tumor volume to a massive rise in tumor growth rates. To better understand the dynamics of sub-clonal interactions and predict the effects of targeted therapeutic interventions, we developed a computational framework to construct non-spatial, dynamic mathematical models of tumor heterogeneity. Our method uses high-performance computing to automatically infer models from the data, simulate them through time, and evaluate the in silico results in comparison to the results obtained from the experiments at the bench. To test this approach, we developed three mathematical sub-clonal interaction equations and inferred complete models that can accurately recapitulate tumor volume and clonal frequency data from mice xenograft experiments with human cancer cells. Importantly, the reverse-engineered models can predict the results of novel experiments and perturbations, and hence determine the optimal clone or clones to target for therapeutic intervention to make the tumor stabilize or even collapse. This project will provide significant insight into the underlying complexity of tumor sub-clonal dynamics. Understanding the dynamics of tumor heterogeneity will provide essential information for cancer drug development and the potential for individualized medical treatments based on a patient’s unique tumor composition.

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NEST SITE CHARACTERISTICS OF THE BAHAMA ORIOLE: HABITAT REQUIREMENTS OF A CRITICALLY ENDANGERED SPECIES

Briana Yancy1, Jennifer Christhilf1, Scott Johnson2, Kevin Omland1
1Department of Biological Sciences, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, Maryland 21250
2 Bahamas National Trust, Bay St. Business Centre, Bay St. East Nassau, Bahamas

The Omland Lab at the University of Maryland, Baltimore County (UMBC) has been conducting research on Bahama Orioles (Icterus northopi) since 2015. This is a critically endangered species, restricted to just to Andros Island. The Bahama Orioles were extirpated from Abaco Island in the 1990’s for reasons unknown. Previous research suggested that the only species of tree the Bahama Oriole used for nesting was the Coconut Palm (Cocos nucifera) in developed habitats. However, in May 2016, UMBC undergraduates found the birds nesting in a native understory palm (Leucothrinax morrisii) in remote pine forests for the first time. In May 2017, our team found several more nests in pine forests. For each nest tree we measured the tree height, diameter and nest height. At each nest tree we took measurements of characteristics for the habitat in a 10 meter radius plot around the nest. These characteristics include 1) number and height of thatch palms 2) number and height of pines, 3) average height of the pine understory and 4) the burn history. We then measured two randomly selected control plots 50 meters from the nest. Then we measured the same characteristics for the control plots as well in order to statistically compare the actual nest site to the control points. My goal is to determine first if the birds are choosing habitats at random or not. Then if the orioles are not choosing habitats at random, I will determine which specific habitats the Bahama Oriole needs.

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Raman spectroscopy has emerged as a leading analytical technique for rapid and selective detection. The implementation of high intensity laser sources, fiber optic coupled spectrographs and sensitive Intensified Charged-Coupled Device detection systems have helped to overcome sensitivity issues in Raman experiments. Yet, the selection of the optimal wavelength of the excitation source or the ideal resolution of the collected scatter remains a problem to be overcome. An ultraviolet excitation source offers larger scattering cross-sections when compared to light in the visible spectrum. Ultraviolet excitation also offers potential resonance enhancements, but simultaneous phenomena such as absorption and photodegradation of the analyte often distort, or completely hinder the collection of Raman scatter. Conversely, the use of a visible excitation source may avoid absorption phenomena, but the scatter is inherently weaker and background emissions are more readily observed. This work explores the capabilities of a novel spectrograph that differentiates between Raman spectra when both 266 and 532nm-light are used for excitation. To further test the capabilities of the spectrograph, it was adapted to allow multiple spectra of differing resolution to be collected. This adaptation proved to marginalize the current trade-off of high resolution for low spectral width. The preliminary findings indicate these methods may be extrapolated to a more effective apparatus; eliminating the need for a priori deduction of the optimal excitation wavelengths or resolutions for Raman experiments.

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Oxygen, the last electron acceptor in the electron transport chain, is a critical component of oxidative phosphorylation that is required to generate cellular energy (adenosine triphosphate or ATP). Consequently, in complete absence of oxygen or anoxia, most organisms experience an imbalance in ATP production and consumption, which leads to cell death. However, some organisms such as the zebrafish have evolved adaptive mechanisms to survive oxygen deprivation. Under anoxia, zebrafish embryos can reversibly arrest their development to reduce oxygen demand and ATP depletion. In particular, we have found that mid-blastula (“dome stage”) embryos undergo the most clear and rapid arrest in response to anoxia. We have further shown that cell cycle progression in dome stage embryos appears to arrest in S (DNA Synthesis) and G2 (Gap 2), consistent with previously published work. Based on these observations, we hypothesize that anoxia activates a cell cycle checkpoint in S/G2, leading to accumulation of cells in these phases of the cell cycle. The purpose of my research project is to test candidate molecules that may function as such anoxia-induced cell cycle checkpoints, including the Retinoblastoma protein (a tumor suppressor that restricts the cell’s ability to replicate DNA), p21 and p27 (cyclin-dependent kinase inhibitors), and p53 (a tumor suppressor activated in response to stress signals). I will analyze the levels and distribution of these proteins using wholemount immunolabeling of dome stage embryos exposed to different durations of anoxia. Thus far, my preliminary data indicates a reduction in the levels of hyper-phosphorylated Retinoblastoma, which may correlate with arrest in S phase. Identification of an anoxia-induced cell cycle checkpoint would represent a significant advance in this field, as this molecule is likely to be the target of a more proximal signal that mediates the adaptive response to low oxygen.
SUPPORTING KNOWLEDGE TRANSFER IN AFTER SCHOOL EMPLOYMENT AT LOCAL 3D PRINT SHOP

Mark Minnis¹, Pavan Konanur², William Easley¹, Dr. Amy Hurst¹
¹Department of Information Systems, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
²Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

3D print shops are unique and interesting spaces because of the limited number in existence, and the complex technologies utilized in these spaces. Furthermore, these shops provide their local communities more access to this advanced technology. However, the print shop located at the Digital Harbor Foundation (DHF), in Baltimore, Maryland stands out, because the DHF print shop staff is comprised of high school students working an after school job. We conducted an exploratory study to better understand the work processes at the DHF print shop, and to identify possible areas for technical intervention.

Over a period of ten weeks we conducted daily observations at the print shop identify problem areas in work processes. These observations helped us to identify recurring breakdowns in communication (between print shop staff, management, and clients) and troubles with job documentation. We then conducted interviews with print shop staff to contextualize our observations, and better understand how communication could be improved in the work places. One problem area identified was the “jobs dashboard”; a tool used to track job progress. However, major flaws prevented it from being an effective tool. We found that the dashboard failed to adequately support knowledge transfer between shifts. We designed and implemented an updated jobs dashboard, which included multiple new areas where details about a job could be recorded. This increase in detail documentation made the process of transferring information between shifts much simpler.

After iterative prototyping and design with print shop management, we deployed the dashboard into the print shop work process. From the limited interaction the staff had with the new dashboard we saw an increase in communication and improvements in job documentation. Further observations will be needed to see if this new jobs dashboard was truly successful.

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NMR STUDIES OF HIV-1 REV RESPONSE ELEMENT CONFORMATIONAL HETEROGENEITY

Daniel Morris¹, Sophia Abbott¹, Colin O‘Hern¹, Stanley Wang¹, Julia Zaki², Jan Marchant¹, and Michael F. Summers¹

¹Department of Chemistry and Biochemistry, Howard Hughes Medical Institute, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
²Department of Undergraduate Studies, University of Maryland, 7950 Baltimore Avenue, College Park, MD 20742

Human immunodeficiency virus (HIV) needs to export its large RNA genome out of the host cell nucleus and traffic it toward the plasma membrane for packaging into new virions. Unspliced and singly spliced viral RNAs are retained within the nucleus and contain a noncoding region of the HIV genome known as the Rev response element (RRE). The viral accessory protein Rev binds to RNAs containing the RRE, an essential process for nuclear export. This RNA-Rev complex uses the CRM1 nuclear export system to transport the complex into the cytoplasm for translation or packaging.

We are characterizing the structure of the RRE by Nuclear Magnetic Resonance (NMR). First, we identified structural elements by comparing spectra of the full-length RRE to those of an array of smaller fragments based on computational secondary structure predictions. Spectra from mutually incompatible fragments were found to have good agreement with the full-length spectra leading us to predict the existence of two alternative conformations. These folds are highly similar but contain one variable region that can adopt different base pairings to produce an overall RRE structure of either four or five stem-loops originating from a central bulge. Full-length RRE was analyzed by electron microscopy and resulted in an envelope lacking electron density where we predict variable folding to occur. This loss of signal could be due to an averaging of data from the two folds. To test this, we have introduced mutations to stabilize the four or five stem-loop structures of the RRE and analyzed them by NMR. These mutants will also be investigated by electron microscopy for analysis of the isolated conformers.

We hope to utilize these techniques to further characterize the RRE, in both conformations, and RRE-Rev complex. By understanding the three-dimensional structure, further progress can be made toward inhibiting viral genome export.

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The retinal pigment epithelium (RPE) of the eye acts as a protective barrier and transports nutrients to the retina. Within the retina, visual pigments in the outer segment discs of photoreceptor cells are responsible for absorbing light and initiating the visual response. Every day outer segment discs are shed and removed by RPE phagocytosis to get rid of the toxic accumulation of build-up. RPE cells can be grown in culture but they do not perfectly maintain in vivo properties. The aim of this study was to compare marker expression for RPE cells grown on plastic and transwell substrates. In order to test this, three different cell lines (RPEJ, ARPE19, and D407) were plated and grown on transwells and plastic over the course of multiple weeks to determine which surface is best for differentiation. After the cells reached confluency, they were harvested, sonicated, and a protein assay was conducted to quantify the amount of protein present in each cell line. Dilutions were made from these calculations and the protein samples were separated by SDS-PAGE for analysis by Western blotting. Beta-actin, MERTK, and OTX2 were visualized by alkaline phosphatase detection to evaluate protein loading and expression. The results showed that there was more protein per surface area from cells grown on transwells than on plastic, the D407 cell line had the most protein present. The RPEJ cell line showed the biggest difference in protein whereas ARPE19 showed the biggest difference in marker expression between cells grown on transwells and plastic. From these data, I can conclude that the culture conditions affect RPE cell growth and marker expression and further experimentation is needed to optimize culture conditions to achieve the phenotype most like in vivo.

I would like to thank the University of Michigan SMART Program for the funding support of this research project.
The human immunodeficiency virus type one (HIV-1) is a retrovirus that suppresses activity of the human immune system. During the late phase of HIV-1 replication, transcripts of the viral genome are produced, and alternative splicing events occur. For viral replication to proceed, unspliced and singly spliced RNAs must be exported out of the nucleus. This requires some spliced transcripts to be translated into Rev, a viral protein. Rev is imported back into the nucleus, where it binds and oligomerizes along a landmark within unspliced and singly spliced RNAs, called the Rev Response Element (RRE). RRE-Rev complexes are then recognized by nuclear export machinery, which leads to export of unspliced and singly spliced RNAs. A recent publication suggests the RRE also has an affinity for Gag, a polyprotein that recognizes and packages the HIV-1 genome during replication. This research will attempt to understand the binding of the RRE with HIV-1 proteins, specifically Rev and a variety of Gag constructs.

Electrophoretic mobility shift assays (EMSAs) are used to study RNA-protein interactions. As RNA-protein complexes form, gel bands migrate slower than the RNA alone, due to increased molecular size. RRE-Rev complexes are challenging to study with this method due to Rev oligomerization at the minimum concentrations required for ethidium bromide (EtBr) staining. In order to effectively visualize RRE-Rev interactions, a more sensitive dye is necessary, such as SYBR Green II. A side-by-side analysis of SYBR Green II and EtBr staining is currently in process, which will help determine the best dye for EMSA use. This work will aid in accomplishing one of the goals of the project, which is to characterize the interactions between the RRE and HIV-1 proteins. Accomplishing this goal will be significant for HIV-1 therapeutic development.

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USING NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY TO ASSIGN THE MONOMERIC CONFORMATION OF THE HIV-1 5'-LEADER RNA

Matthew Orellana¹, Joshua Brown¹, Michael Summers¹, Ph.D.
¹Department of Chemistry and Biochemistry, Howard Hughes Medical Institute, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

The 5' Leader (5'-L) of the human immunodeficiency virus-1 (HIV-1) RNA genome is a highly conserved region that is constantly in equilibrium between a monomeric and dimeric conformation, which has not been exploited as a therapeutic target. HIV-1 is currently treated with a medication cocktail where each drug targets a specific viral protein in the replication cycle. These non-conserved proteins undergo high rates of mutations that leads to the frequent emergence of resistant strains. In the monomeric conformation, the RNA is translated into proteins necessary for viral replication while the dimeric conformation is packaged as the genomic material for the new virion. While the structure of the dimeric conformation has already been determined, the monomeric conformation has remained elusive. We have used nuclear magnetic resonance (NMR) spectroscopy to extensively probe the secondary structure of the monomeric conformation. NMR studies of large RNAs is challenging due to the large amount of overlapping signals in the NMR spectrum. In order to confidently assign signals from the 5'-L, we have constructed various fragment controls and used nucleotide-specific labeling schemes to collect data that can then be overlapped with the full length 5'-L data. We have found that the dimer-promoting stem-loop is sequestered in an extended intramolecular interaction with the U5 region. Additionally, the polyA region is completely unstructured possibly for a binding site for translation machinery. We plan to continue studying the 5'-L in order to determine its three-dimensional structure.

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ROLE OF N-MYC DOWNSTREAM REGULATED 1 (NDRG1) IN ADAPTATION OF THE KIDNEY TO LOW OXYGEN

Afia Osei-Ntansah, Jong Park, Rachel Brewster, PhD
1Department of Biological Sciences, University of Maryland, Baltimore County
1000 Hilltop Circle, Baltimore MD, 21250

Oxygen deprivation, which occurs in pathological conditions such as stroke and congenital heart disease results in irreparable cellular damage and even death in humans. However, a number of organisms, including zebrafish, seemingly defy such odds. Under anoxia (0% oxygen), zebrafish embryos enter a hypometabolic state characterized by reversible developmental arrest that enables them to conserve cellular energy (ATP) and survive for up to 50 hours. Developmental arrest is manifested by cessation of most ATP-demanding processes. The molecules that sense low oxygen and orchestrate arrest are for the most part unknown, yet knowledge of such signals would be tremendously beneficial for therapeutic purposes. In an effort to identify molecules that promote arrest, the Brewster laboratory performed metabolic profiling and found that lactate is one of several metabolites that are up-regulated in embryos exposed to anoxia. Lactate has previously been shown to bind to NDRG3 in hypoxic cancer cells and to promote cell survival identifying lactate/NDRG as a candidate signal for adaptation to low oxygen. We have further found that NDRGs are expressed in tissues with high metabolic demand in the zebrafish embryo. My research project focuses on NDRG1 that is expressed in the embryonic kidney and ionocytes (which maintain ionic homeostasis). Preliminary data indicate that NDRG1 is localized to the cytosol of ionocytes under normoxia and shifts to the cell cortex under anoxia, where it downregulates the ATP-demanding Na+K+ATPase pump. The goal of my project is to determine whether this change in cellular distribution also occurs in the kidney and if so, to assess whether it is dependent on lactate binding. To address this, I am employing immunolabeling using NDRG1 antibody and a marker for the plasma membrane to examine NDRG1 localization in presence/absence of oxygen and lactate. If correct, this model represents a novel, rapid response mechanism to low oxygen.

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OPTIMIZING AND AUTOMATING THE PHOSPHORUS EXTRACTION AND RECOVERY SYSTEM (PEARS)

Charles Portner¹, Samina Musa¹, Utsav Shahsvatt¹, Josh Benoit¹, Lee Blaney¹

¹Department Chemical, Biochemical & Environmental Engineering, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

The high phosphorus content of agricultural runoff, such as that generated on poultry farms, contributes to eutrophication of surface waters, disrupting environmental processes and decreasing biodiversity in affected ecosystems. By reframing “pollution prevention” as “resource recovery”, we can reduce or reverse eutrophication, while also fostering a closed-loop nutrient management system for agriculture. In this project, we reduced nutrient concentrations in poultry litter by developing a fully automated Phosphorus Extraction and Recovery System (PEARS), which was designed to extract phosphorus, nitrogen, and magnesium from poultry litter and to recover a high-value product, struvite. The laboratory-scale PEARS reactor consists of three tanks in series, and the system was fully automated to allow mixing, fluid transfer, and acid/base addition. A Matlab-based electronics interface was used for feedback control and adjustment of fluid levels, solids/struvite removal, reactant loading, and pH adjustment. The overall run-time of the PEARs operation was approximately 60 minutes. During operation, a 30-L tank was loaded with a 40-80 g/L poultry litter slurry and vigorously mixed. Then, the slurry was transferred to the extraction reactor, where phosphorus was extracted from the slurry at pH 4.5-5.5 after bubbling CO₂(g) and adding 1 M HCl. Solid-liquid separation was achieved through gravity settling, and the supernatant was transferred to the precipitation reactor. The nutrient-rich extract was aerated and dosed with 1 M NaOH to increase the pH to 8.0-9.0. These conditions resulted in production of struvite. In preliminary trials, we were able to extract 66% of the phosphorus from the poultry litter, and we recovered 54% of the extracted phosphorus in the form of struvite. Based on these findings, the PEARS reactor can be deployed on Maryland poultry farms to help ameliorate nutrient pollution.

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Human Immunodeficiency Virus (HIV) is a disease that interferes with the immune system of the human body. It spreads through the exchange of bodily fluids and once in the body, it attacks CD4 cells. The lower the CD4 cell count, the more susceptible a person is to obtaining Acquired Immunodeficiency Syndrome (AIDS). Since there is no cure to HIV/AIDS thus far, research is being done to see if there is a way to stop HIV replication.

HIV has an RNA genome that consists of a monomer and a dimer conformation. The functions of a monomer include translating and splicing while the dimer packaged. In order to study the RNA genome, an efficient method must be used to synthesize and purify the RNA. PCR amplification is required to create more copies of a specific DNA template. After making copies of DNA, a trial transcription reaction is required to find a method to get the best yield of DNA. *in vitro* RNA transcription is used to produce the RNA. The transcribed RNA then goes through the first step of the purification process, the denaturing gel. The denaturing gel helps disrupt and separate the proteins from the transcribed RNA in order to solely have the RNA strand. When the RNA gets extracted from the gel, it is put into an Elutrap in order to separate the RNA from the gel. The RNA then goes through a series of washes; initial, salt, and water, in order for it to fully purify.

Once the RNA is fully purified, it goes through the capping process and then gets examined further in the nuclear magnetic resonance (NMR) spectrometer. The NMR is used to analyze structures and properties of certain parts of RNA.

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Stimuli-responsive polymers have recently been investigated for their wide range of applications in drug delivery, optical systems, and biological engineering, among other uses. This project will focus specifically on temperature-sensitive polymers, which can change conformation in response to a change in temperature. The polymer forms a hydrophilic coil at lower temperatures and then collapses to form a hydrophobic globule when heated above its lower critical solution temperature (LCST). Similarly, environment-sensitive fluorophores are also able to respond to changes in their local environment. Solvatochromic fluorophores exhibit unique stimuli-induced emission properties that are caused by changes in solvent polarity in the local environment. By attaching a solvatochromic dye to a temperature-sensitive polymer, it may be possible to monitor any conformational changes in the polymer by observing a fluorescence change in the dye. In this research, a novel solvatochromic dye has been synthesized and characterized through absorption, fluorescence, and \textsuperscript{1}H-NMR spectroscopy. The dye, a 4-substituted 1,8-naphthalimide, was purified with a chromatotron and its solvatochromism was investigated in solvents of varying polarity. A temperature-sensitive polymer has also been synthesized, containing an amine-reactive group to which the dye will be attached. Synthetic strategies are still being explored to attach the dye to the polymer. The dye-labeled polymer will then be characterized above and below its LCST using fluorescence spectroscopy to validate that the dye can be used to monitor real-time “coil-to-globular” phase changes in the polymer.

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INVESTIGATION OF SLIDING HYDROGELS FOR USE AS A HEMOSTATIC AGENT

Eve Schodowski1, Chris Heckert2, Erin Lavik2, Jennie Leach2
1Department of Biological Sciences, Cleveland State University, 2121 Euclid Avenue, Cleveland, OH 44115
2Department of Chemical, Biochemical, and Environmental Engineering, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

Traumatic injury is the leading cause of death for the 1-44 year old age group, and while many products have been developed to treat the external injuries involved with trauma, few options currently exist for first responders to treat internal bleeding. To illustrate the magnitude of this problem, fatal severe blood loss can occur in 5-10 minutes and is associated with over 30% of civilian deaths before the patient reaches the hospital, and 50% of mortalities in military settings. Surprisingly, these statistics have not improved in almost 40 years. Addressing this issue, the collaborative labs of Dr. Jennie Leach and Dr. Erin Lavik work to engineer hemostatic nanoparticles that would be delivered intravenously to quickly treat widespread, uncontrolled, internal hemorrhages. More specifically, the focus of this project is to investigate whether polyrotaxanes, a sliding hydrogel, could be used for hemostatic applications due to their unprecedented range of platelet like properties such as high elasticity, strength, and capacity for self-healing. The research approach aims to synthesize polyrotaxanes using literature published by Okumura & Ito at the University of Tokyo and Tong & Yang at Stanford University, then characterize the polymer using nuclear magnetic resonance (NMR) and mass spectroscopy. Once a successful synthesis and molecular characterization are achieved, the next aim would be to characterize the physical properties of the hydrogel through swelling and rheology experiments. Using Okumura & Ito’s method, the hydrogel has not yet been successfully synthesized. However the Tong & Yang method, which remains to be conducted, appears to be a promising and biocompatible alternative.

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STUDYING CELL POLARIZATION USING A STOCHASTIC MODEL

Jamshaid Shahir¹, Hye-Won Kang¹
¹Department of Mathematics and Statistics, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21163

Under chemical stimulation, motile eukaryotic cells polarize in response to external stimulus. This polarization involves the recruitment of various proteins to the plasma membrane, where they segregate to a front or back end of the cell. Previous studies have shown that the mechanisms of cell polarization can be explained using a simplified partial differential equation (PDE) model consisting of a single active/inactive protein pair with positive feedback. In this study, we focus on creating a continuous-time Markov chain model, which can be simulated by the Gillespie’s algorithm, and investigate how inherent noise affects the dynamics of cell movement especially when there is low quantity of the proteins. In the Markov chain model, we use discrete compartments to describe the diffusion of molecules in the domain. We apply this methodology to simulate a travelling wave model of cell polarization and compare the results with a PDE model by taking into account the volume of the cell. Finally, we examine how changing related parameters affects the wave propagation in the model.

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Comparison of ground based observations for detecting falling snow

Bailey Swick\textsuperscript{1}, Ali Tokay\textsuperscript{2}

\textsuperscript{1}Department of Atmospheric and Hydrologic Science, St. Cloud State University, 720 4\textsuperscript{th} Ave S, St. Cloud, MN 56301
\textsuperscript{2}Joint-Center for Earth Systems Technology, University of Maryland Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

One of the four level one requirements of the National Aeronautics and Space Administration Global Precipitation Measurement (GPM) program is to detect falling snow at an effective resolution of five km of dual frequency precipitation radar (DPR) and 15 km of GPM microwave imager. Two operational weather radar products, Hydrometeor Identification (HID) and Multi Radar Multi Sensor (MRMS), are considered a validation tool for GPM DPR precipitation type algorithms over the continental United States. The former product classifies the precipitation type at nine different classes for each radar pixel at a given range and azimuth. The latter product outputs percent snow at 12.5\% increments at 0.01x0.01 degree pixel. The temporal resolution ranged between 5-10 minutes, depending on the volume coverage pattern (VCP), for HID and 30 minutes for MRMS.

This study uses the Automated Surface Observing System (ASOS) present weather to evaluate the performance of HID and MRMS products. ASOS reports present weather, surface temperature, and several other observables at one-minute resolution. The database consisted of 11 ASOS stations covered by ten radar sites from the winter of 2014-2015. A total of 143 days were identified as snow, cold rain, or mixed precipitation event. Qualitative and quantitative assessments were performed taking into account the freezing level and the radar sampling height. For the qualitative assessment, event based diagrams were made to compare each product at its native resolution. For the quantitative assessment, the ASOS was brought to the resolution of HID and compared, then the ASOS and HID were brought to the resolution of MRMS and compared. A weighted sorting method was used to determine the precipitation type and HID for the upscaled time periods. The preliminary results indicate that the performance of the HID and MRMS products depend on the beam height, freezing level, and the intensity of snow.

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THE ROLE OF Mg$^{2+}$ IN STABILIZING tRNA:MA INTERACTIONS

Phoebe Somani$^1$, Emre Tkacik$^1$, Amalia Rivera-Oven$^1$, Matthew McDonough$^{1,2}$, Noel Getachew$^1$, Christy Gaines$^1$, Dr. Michael Summers$^1$

$^1$Department of Chemistry and Biochemistry, Howard Hughes Medical Institute at the University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
$^2$Department of Chemistry and Physics, Western Carolina University, 1 University Way, Cullowhee, NC 28723

Human immunodeficiency virus-1's (HIV-1) matrix domain (MA) of the Gag polyprotein targets Gag to the cell membrane through interactions between MA’s highly basic region and phosphatidylinositol 4,5-bisphosphate [PI(4,5)P$_2$] on the cell membrane. Targeting to the plasma membrane is critical for the assembly of the virus. In addition to interacting with the plasma membrane, other studies have shown that MA specifically binds to certain tRNAs, including tRNA$^{\text{Lys}}^3$ and tRNA$^{\text{GlyGCC}}$, in vivo. Host tRNA$^{\text{Lys}}^3$ acts as a primer for reverse transcription and is packaged into budding virions. Mg$^{2+}$ is crucial for stabilizing the tertiary structure of tRNA, which will allow for the molecule to bind to matrix in vitro. However, when using NMR, increasing the Mg$^{2+}$ concentration in our sample causes peaks to broaden and disappear, making the spectra difficult to interpret. By optimizing conditions that maximize the tightness of binding while minimizing the amount of Mg$^{2+}$ needed for successful folding using the ITC, we will be able to determine the best conditions for further structural studies with the NMR. The studies confirmed that both tRNA$^{\text{GlyGCC}}$ and tRNA$^{\text{Lys}}^3$ form a complex in vitro and that increasing the amount of Mg$^{2+}$ strengthens the tRNA:MA interaction. Additionally, we discovered that at a given Mg$^{2+}$ concentration, tRNA$^{\text{Lys}}^3$ binds more tightly than tRNA$^{\text{GlyGCC}}$. If we can characterize the structure of the MA: tRNA complex using NMR, it would allow for better understanding of the mechanism by which HIV specifically targets the cell membrane and how viral assembly and budding occur.

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OVEREXPRESSION OF CARBON CONCENTRATION MECHANISM GENES CAH1 AND LCI-1 IN CHLAMYDOMONAS REINHARDTII FOR INCREASED ALGAL GROWTH AND OIL PRODUCTION

Kimuel Villanova¹, Newton Mayaka¹, Lian Jackson¹, Amrita Madabushi¹, Stephen M. Miller²
¹Biotechnology Program, Baltimore City Community College, 2901 Liberty Heights Ave, Baltimore, MD 21215
²Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

High fossil fuel consumption and global warming concerns have encouraged exploration of renewable resources, including biofuels produced by algae. A major problem with algae as a source of biofuels is the relatively low amount of oil that algae can produce under optimal conditions. Most efforts towards improving algae as a fuel source are focused on increasing the output and decreasing the generation time of algae growth. Here we attempt to increase the growth rate of the model green alga Chlamydomonas Reinhardtii by overexpression of two genes related to the Carbon Concentrating Mechanism (CCM). The focus here is to use molecular cloning and genetic transformation methods to overexpress LCI-1 (Low Carbon Inducible Membrane Protein) and CAH1 (Carbonic Anhydrase 1), which are responsible for carbon dioxide uptake from the environment into the cell. LCI-1 is located within the plasma membrane and CAH1 is located in the periplasmic space between the cell wall and plasma membrane.

Accumulation of both LCI1 and CAH1 is induced by low carbon dioxide concentrations. Our cloning strategy involves ligating into existing vectors containing LCI1 and CAH1 coding sequence, a bleomycin selectable marker gene that is linked to a viral 2A peptide sequence, to ensure production of our CCM proteins. These plasmids will be transformed into Chlamydomonas reinhardtii and transformants will be tested by western blot for CCM protein accumulation. Transformant growth rates will be analyzed in a multi-cultivator that determines culture OD at regular intervals. If our strategy for improving growth is successful, it could be implemented in related algae better suited for large-scale industrial production of biofuels.

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Human immunodeficiency virus type 1 (HIV-1) has become an epidemic where millions of people are infected worldwide. During the HIV life cycle, the viral assembly is initiated by interactions between the unspliced viral genomic RNA and its translated product, the Gag polyprotein. Although there is an excess of non-viral RNA, Gag protein is able to efficiently package the viral genomic RNA. We seek to understand the molecular mechanism underlying the selective genome packaging for HIV-1. Gag contains three structured domains: Matrix (MA), Capsid (CA), and Nucleocapsid (NC). The NC domain of Gag binds to the 5′-leader (5′-L) within the dimeric HIV-1 genome for selective packaging, while the CA domain mediates Gag-Gag interaction resulting in the hexagonal Gag shell that encapsulates the viral RNA. The basic unit of the immature viral shell is Gag hexamer. Mutations at the hexameric interface cause a significant reduction in Gag’s selectivity towards dimeric viral RNA.

We hypothesize that the hexameric structure of the CA domain contributes to the RNA genome selection. Using Nuclear Magnetic Resonance (NMR) and Isothermal Titration Calorimetry (ITC), about sixteen binding sites for Gag proteins on the 5′-L have been mapped. In order to interrogate whether these binding sites are capable of promoting the formation of the Gag hexamer, chemical crosslinking, engineered disulfide bonds, and cryo-electron microscopy are utilized with modified RNA and protein constructs. Results show that Gag binding alone cannot form a hexamer, and neither can non-specific RNA. Only viral RNA, specifically the 5′L, is able to promote formation of the Gag hexamer. This supports the idea that hexameric Gag protein recognizes the 5′L, and this complex functions as the nucleation site to initiate viral assembly. We aim to solve this protein-RNA initiation complex, which will provide the detailed molecular mechanism for selective genome packaging.

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INCLUSIVE SPECIAL EDUCATION VIDEO CODING AND ANALYSIS

Jessica M. Calle1, Erin Buehler2, Amy Hurst2
1Department of Psychology and Social Work,
University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
2Department of Information Systems,
University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

For the past three spring semesters, UMBC has offered an entrepreneurship course on 3D printing for undergraduates and students with intellectual disabilities (ID) from the Shriver Center’s SUCCESS program. This course teaches 3D modeling and printing skills in the context of entrepreneurship, combining themes related to STEM, real world employability, and social good.

My current research extends this work and seeks to understand how a technology-driven classroom and an inclusive, team-based learning environment impacts student interactions. To answer this question, we created a qualitative coding schema informed by special education experts to analyze video data gathered from this course. We are using BORIS, a software for video/audio coding and live observations, to analyze this data and apply our coding schema. Before implementing the coding schema, I familiarized myself with the software through tutorials and reading the BORIS user guide. After some practice with the software, I created my own coding schema based on behaviors I noted from the videos and I provided input into the final design of the overall coding schema. I also helped incorporate the feedback provided by the special educators concerning positive and negative interactions into the final coding schema.

Out of the 240 post-secondary programs in the U.S. and Canada that offer an inclusive postsecondary certificate program for individuals with intellectual disabilities, UMBC is the only one that has collected data based on inclusive technology-driven classroom and an inclusive team-learning environment. Our goal is to identify detectable patterns from the video data that led to the significant behaviors described by the special educators. Findings from this research may lead to the development of new technologies to help support students with intellectual disabilities in technology-driven classrooms. Future work may focus on the application of our findings to benefit other post-secondary programs in the U.S. and Canada.

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OVEREXPRESSION OF TWO CARBONIC ANHYDRASE ENZYMES TO AMPLIFY ALGAL CELL GROWTH

Sarah Carpe¹, Charity Ensor¹, Lian Jackson¹, Amrita Madabushi¹, Stephen M. Miller²
¹Arts and Sciences, Biotechnology Program, Baltimore City Community College, 2901 Liberty Heights Ave, Baltimore, MD 21215
²Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

Green algae are photosynthetic plant-like organisms that have great promise as a source of sustainable biofuels. Production of biofuels from algae is a sustainable alternative to fossil fuels and is potentially more economical than ethanol produced from corn, another alternative fuel source. *Chlamydomonas reinhardtii*, a single celled microalga, is the most widely used model organism for algal biofuel production research. Many tools are available for molecular genetic manipulation of *C. reinhardtii*, and it is easy to culture, making it an excellent platform for biotechnology research. The limiting factor for algal growth, carbon dioxide (CO₂), is the focus of much algal biofuels research. It is believed that certain enzymes involved in carbon dioxide uptake into cells for conversion to carbohydrates and eventually lipids, can be manipulated to improve photosynthesis and growth. The focus of this study are two genes that encode enzymes involved in CO₂ uptake, *CAH6* and *CAH3*. Carbonic anhydrase 6 (CAH6) converts CO₂ into carbonate in the chloroplast stroma, increasing carbon flow into the pyrenoid, where CO₂ fixation into carbohydrate takes place. Carbonic anhydrase 3 (CAH3) converts carbonate back into CO₂ in the pyrenoid. Our prediction is that over expression of these enzymes should improve photosynthesis and thus cell growth. Utilizing recombinant DNA technology, we are generating vectors with *CAH6* or *CAH3* coding regions connected to *ble* gene sequence (encodes a selectable marker protein), with the hope of increasing expression of these proteins. We will electroporate these vectors into *C. reinhardtii* then use western-blot analysis to determine transgenic protein expression. Finally, we will use an algal multiculivator to observe and compare growth of our transformants to a control strain. If overexpression of CAH3 and CAH6 improves growth in *C. reinhardtii*, then our methods can be applied to other algal species, such as *Chlorella vulgaris*, a biotechnology production organism.

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FOOD INFECTIONS WITH *LISTERIA MONOCYTOGENES*

**Kavneet Chahil**¹, Loran Margevich¹, Tonya M. Santaus¹², Dr. Chris D. Geddes¹²

¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250

²The Institute of Fluorescence, Columbus Center, University of Maryland Baltimore County, 701 E. Pratt Street, Baltimore, MD, USA 21202.

This experiment aims to analyze DNA extraction from various foods infected with *Listeria monocytogenes* (Listeria) such as shredded cheese, spinach, cantaloupe, ground beef, and hot dog, through conventional heating and microwave irradiation. Listeria DNA and proteins will be extracted and detected through Ethidium Bromide gel electrophoresis and Ruby stained sodium dodecylsulfate gel electrophoresis (SDS PAGE).

To carry out the experiment, ground foods were infected with $10^8$ colony forming units (cfu) concentration of *Listeria monocytogenes*, and incubated for 24, 48, and 72 hours respectively. After each period, the foods were either conventionally heated (boiled) or microwave irradiated with and without the use of microscope slides containing gold bow-ties at varying powers and times. Conventionally heated foods were analyzed from 40-80 °C, and microwaved foods from 10% power to 50% power for 60 seconds. The gold bow-tie triangle lysing slides are designed to increase bacterial cellular lysing, DNA fragmentation, and protein degradation. After ethanol precipitation and DNA rehydration, ethidium bromide gels were run to analyze DNA fragmentation. A collaborator at the University of Maryland, Medical Center performed Polymerase Chain Reaction (PCR) to determine if Listeria can be detected from infected foods. SDS PAGE was also run to analyze protein degradation.

The ethidium bromide gels display smearing and banding from combined food and Listeria DNA demonstrating the ability of DNA to be extracted from conventionally heated and microwave irradiated samples. The protein gels show smearing as the temperature and power increases with and without the gold bow-tie structures. It can be analyzed that the use of gold bow-ties aid in the extraction of DNA and proteins from Listeria contaminated food products and can be detected on the PCR platform.

We would like to acknowledge Dr. Chris D. Geddes, the Institute of Fluorescence, UMBC, and collaborators for their support.
PAVING THE WAY FOR REGENERATIVE MEDICINE: CURATING PLANARIAN EXPERIMENTS IN A CENTRALIZED MATHEMATICAL DATABASE

Junaid Bhatti and Daniel Lobo
Department of Biological Sciences, University of Maryland Baltimore County
1000 Hilltop Circle, Baltimore, MD 21250

For more than a century, scientists have been captivated by the regenerative capabilities of the planarian flatworm, which can regenerate a full body from almost any type of amputation. To understand the mechanisms controlling this extraordinary ability, research approaches based on surgical, pharmacological, and genetic manipulations have been used extensively to produce a huge dataset of experimental results disseminated through the literature. The rise of sophisticated machine learning algorithms and computational power has brought upon a novel way of studying these fascinating creatures. The Lobo Lab has developed a mathematical ontology for encoding regenerative experiments, together with an artificial intelligence method to automatically infer mechanistic models of regeneration. In this project, we have curated hundreds of additional experiments into a formal database, called Planform, which stores, in a mathematical language, thousands of planarian experiments and results performed in the past decade. The data includes the details of the experimental interventions, such as drug additions, genetic interference, and surgical manipulations, and their effects on the resultant morphologies. This curated dataset not only deal with the growth and shape of the worm, but also show how the presence, or the lack, of key genes, results in different patterns and morphologies in the worm. Planform is a freely-available, centralized collection of information that helps the scientific community search efficiently for experiments and morphologies published in the literature. More importantly, this resource is essential for the application of artificial intelligence methods to automatically reverse-engineer models that explain the regulatory mechanisms controlling the regeneration in these worms. The new dataset is already published and freely available in the dedicated website (http://lobolab.umbc.edu/planform), helping human scientists and the automated computational approach to find better models of planarian regeneration. All these efforts will pave the way for the next-generation applications in human regenerative medicine.

Acknowledgement: We thank the members of the Lobo lab and the planarian regeneration community for helpful discussions. This work is partially supported by the National Science Foundation (NSF) under award #1566077.
EXAMINING THE ELECTRICAL EXCITATION, CALCIUM SIGNALING, AND MECHANICAL CONTRACTION CYCLE OF A HEART CELL

REU Site: Interdisciplinary Program in High Performance Computing

Wesley Collins¹, Kristen Deetz², Nygel Foster³, Darius Leftwich³, Chad Meyer⁴, Shalin Patel⁵
Graduate assistant: Carlos Barajas¹, Faculty mentor: Matthias K. Gobbert¹,
Client: Zana Coulibaly⁶

¹Department of Mathematics and Statistics, UMBC
²Department of Mathematics, Eastern University,
³Departments of Computer Science and Electrical Engineering, UMBC
⁴Department of Mathematics, University of Pittsburgh,
⁵Department of Computer Science and Mathematical Science, Kean University
⁶Department of Pharmacology, University of California, Davis

As the leading cause of death in the United States, heart disease has become a principal concern in modern society. Cardiac arrhythmias can be caused by a dysregulation of calcium dynamics in cardiomyocytes. Calcium dysregulation, however, is not yet fully understood and is not easily predicted; this provides motivation for the subsequent research. Excitation-contraction coupling (ECC) is the process through which cardiomyocytes undergo contraction from an action potential. Calcium induced calcium release (CICR) is the mechanism through which electrical excitation, is coupled with mechanical contraction through calcium signaling. The study of the interplay between electrical excitation, calcium signaling, and mechanical contraction has the potential to better our understanding of the regular functioning of the cardiomyocytes and help us understand how any dysregulation can lead to potential cardiac arrhythmias.

ECC, of which CICR is an important part, can be modeled using a system of partial differential equations that link the electrical excitation, calcium signaling, and mechanical contraction components of a cardiomyocyte. We extend a previous model to implement a seven variable model that includes the mechanical component of the ECC. We conduct a parameter study to determine how the interaction of electrical and calcium systems can impact the cardiomyocyte’s levels of contraction.

Acknowledgements: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Co-authors Wesley Collins, Nygel Foster, and Darius Leftwich were supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistant Carlos Barajas was supported by UMBC.
QUANTIFYING VARIABILITY OF BASEFLOW OF WATERSHEDS FOR THE CHESAPEAKE BAY

REU Site: Interdisciplinary Program in High Performance Computing

Christian Dixon¹, Gabriel Martinez², Hwan Hee Park³, Maddie Rainey⁴,
Graduate assistants: Nadeesri Wijekoon¹ and Sai Popuri¹,
Faculty mentor: Kofi Adragni¹,
Client: Jeff P. Raffensperger.⁵

¹Department of Mathematics and Statistics, UMBC
²Department of Mathematics, California State University, Fullerton
³Department of Computational & Applied Math, Rice University
⁴Department of Mathematics, Grand Valley State University
⁵United States Geological Survey (USGS)

The U.S. Geological Survey National Water-Quality Assessment Project conducted a study of 225 sites in the Chesapeake Bay watershed to estimate baseflow. Baseflow is the estimated volumetric discharge of water, primarily from groundwater sources, that is relayed to the measurement sites. The study is necessary to address the Nation's water supply for changes in the environment. Baseflow is estimated using a recursive digital filter. Calculating the variability of baseflow water discharge is important to make informed decisions about water regulation. We explore the estimation of variability of the baseflow using two methods: the bootstrap method and the delta method.

Acknowledgments: These results were obtained as part of the REU Site: Interdisciplinary Program in High Performance Computing (hpcreu.umbc.edu) in the Department of Mathematics and Statistics at the University of Maryland, Baltimore County (UMBC) in Summer 2017. This program is funded by the National Science Foundation (NSF), the National Security Agency (NSA), and the Department of Defense (DOD), with additional support from UMBC, the Department of Mathematics and Statistics, the Center for Interdisciplinary Research and Consulting (CIRC), and the UMBC High Performance Computing Facility (HPCF). HPCF is supported by the U.S. National Science Foundation through the MRI program (grant nos. CNS-0821258 and CNS-1228778) and the SCREMS program (grant no. DMS-0821311), with additional substantial support from UMBC. Author Christian Dixon was supported, in part, by the UMBC National Security Agency (NSA) Scholars Program through a contract with the NSA. Graduate assistants Nadeesri Wijekoon and Sai Popuri were supported by UMBC.
ROLE OF PROTEIN PHOSPHATASES IN MELANOPSIN RESENSITIZATION

Jair Flores¹, Juan Valdez-Lopez¹, Phyllis Robinson¹, PhD
¹Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

G-protein coupled receptors (GPCRs) make up the largest superfamily of membrane receptors. Melanopsin, a GPCR, is a unique visual pigment that is expressed in a small population of retinal ganglion cells called intrinsically photosensitive retinal ganglion cells (ipRGCs) in the mammalian retina. This visual pigment is primarily involved in non-image forming vision. It is involved in regulating physiological processes such as circadian photoentrainment and the pupillary light reflex. Our studies of melanopsin signaling have been focused on the activation and deactivation of its signaling cascade. GPCRs must also undergo resensitization to new stimuli to sustain G-protein signaling for extended periods of time. We hypothesize that there are phosphatases of the protein phosphatase family that resensitize melanopsin by removing Carboxytail phosphorylation’s, and this contributes to melanopsin’s ability to sustain prolonged signaling. We performed Reverse Transcriptase-PCR (RT-PCR), Western blot, and immunohistochemistry to test if protein phosphatases are expressed in mouse retina and in Human Embryonic Kidney (HEK). RT-PCR and Western Blot results suggest the expression of protein phosphatase types 1 and 2. Immunohistochemistry suggests protein phosphatase 2α/β is expressed in all cell layers of the retina and in HEK cells. Preliminary dual labeling experiments show co-expression of protein phosphatase 2 with melanopsin but more experiments need to be done. Future work aims to test the significance of phosphatase activity in melanopsin signaling which will be assessed by calcium imaging and conducting crosslinking assays to confirm protein-protein interaction.
OPTIMIZING THE SYNTHESIS OF HEAVY ATOM CARBON NANODOTS FOR PHOSPHORESCENT CHARACTER

Bobga-Herman Gwanvoma¹, Ammar Raza¹, Rachael Knoblauch¹, Dr. Chris D. Geddes²
¹Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 1000 Hilltop Circle, Baltimore, MD 21250
²Department of Chemistry and Biochemistry, The Institute of Fluorescence, The Columbus Center, University of Maryland, Baltimore County, 701 East Pratt Street, Baltimore, MD, 21202

Carbon nanodots (CND) are spherical carbon nanostructures that have been extensively studied for various fluorescence-based applications, primarily because they have been shown to be non-toxic, photostable, thermostable, and somewhat easily collected. This study seeks to achieve a novel phosphorescent signal through the addition of bromine and iodine to the carbon nanodot structure, therefore employing the heavy atom effect. This emission is sought after because phosphorescence decays on a longer timescale than fluorescence, which opens an alternative non-radiative decay pathway via transfer to ambient, dissolved oxygen. The product of this effect is the generation of highly reactive singlet oxygen, which has been shown in literature to induce cell death for bacterial cells.

Consequently, the basis of this study is to optimize reaction conditions to generate phosphorescent signal through bromination of the carbon nanodots (Br-CND) or iodinated carbon nanodots (I-CND). This study explores the effects of reaction time, reaction temperature, heavy atom addition strategies, and varying pH on the intensity of phosphorescence. Once an optimal method for producing high intensity phosphorescent signals is established, we hope to achieve a structure that will produce singlet oxygen for antibacterial applications.

We would like to acknowledge the Institute of Fluorescence, Dr. Chris Geddes, Rachael Knoblauch, and lab members for all of their support and guidance.
Phosphorylation is a post-translational modification (PTM) to a protein by which phosphates are added to the structure of the polypeptide. Among many other roles, phosphorylation regulates the activation of existing proteins and subsequently is fundamental for ideal physiological performance. The identification of the sites where phosphates bind is difficult without the presence of an enrichment protocol to increase the abundance of phosphoproteins. In this study, titanium dioxide (TiO₂) is used as an enrichment tool to identify all the known phosphate sites located on ovalbumin (OVA). This protein is important for study because its structure provides a model for the structure of other serpin proteins. It is also chosen as a model protein because it has only two sites of phosphorylation and its molecular weight of 42.6 kDa makes it moderately sized. The goal of this research is to optimize the capture step of an existing protocol. Ultimately, this work will be combined with concurrent research with the goal of developing an optimized protocols for the capture of proteins of different lengths.

The enrichment from a mixture of controlled proteins is first performed through the use of a TiO₂ ziptip (ThermoFisher, A32993). Parameters of the capture step are varied to find optimal conditions. After capture, the sample is digested according to the protocol. The sample is then analyzed using high performance liquid chromatography (HPLC) coupled to a high resolution mass spectrometer (HRMS), allowing for high mass accuracy detection. The data is analyzed using PEAKS 7 software, providing sequence coverage and allowing for identification of the phosphorylated sites. The sequence coverage of each sample is analyzed to determine which procedure produces the highest sequence coverage and thus is determined to be optimal. Future research includes continued optimization of established protein analysis protocols to create a set of robust techniques for protein analysis.

Acknowledgements: I would like to thank Baltimore Youthworks Summer Jobs Program for funding my experience at the Summer Biomedical Training Program. I would also like to acknowledge the Molecular Characterization and Analysis Complex (MCAC) for their continuous support in allowing this experiment to be completed.
MICROWAVE-ASSISTED LYSING OF GROUND SPINACH AND DETECTION OF 
FE$^{3+}$ AND MG$^{2+}$

Paula Ladd$^1$, Tonya M. Santaus$^{1,2}$, Dr. Chris D. Geddes$^{1,2}$

$^1$Department of Chemistry and Biochemistry, University of Maryland, Baltimore County, 
1000 Hilltop Circle, Baltimore, MD 21250 
$^2$The Institute of Fluorescence, Columbus Center, University of Maryland, Baltimore County, 
701 E. Pratt Street, Baltimore, MD, USA 21202

Spinach is a common food that contains high concentrations of iron and magnesium. There are various methods of extracting these ions of interest and other cellular components, but we are developing a microwave-induced lysis method for rapid cell disruption. Lysis is the process by which cells are broken open to release their intracellular components. We use the Lyse-IT$^\text{TM}$ technology as the primary cellular lysis mechanism. In this poster we study the comparative amounts of DNA released using our lysis assay against other microwave and conventional heating (boiling) methods. We also performed experiments to determine if the metal ions, iron and magnesium, are released with greater efficiency using our lysis assay.

Using a conventional 900W microwave, ground spinach in deionized water is microwave irradiated allowing the cells to break open. Following this lysis step, two fluorescent probes Ferrum 430 and Magnesium 510, are used to look at Fe$^{3+}$ and Mg$^{2+}$ release respectively. We collected fluorescent spectroscopic data of spinach samples treated with the microwave-induced lysis assay as well as other microwave and boiling methods. Gel electrophoresis stained with ethidium bromide was performed to look at the DNA released and fragmented post microwave irradiation.

We have found that as the power of the microwave increases the amount of DNA released increases along with the concentration of iron and magnesium ions.

We would like to thank Dr. Chris D. Geddes, the Institute of Fluorescence, and UMBC for the support and the opportunity to conduct this research.
Engineered nanoparticles (NPs) are increasingly used in wide-ranging technologies due to their unique nanoscale properties. Thus, production of these nanomaterials has increased significantly in the last decade. With the increase in production of NPs, concerns have been raised on their potential impact on human health and the environment. Having learned from history that emerging technologies, as handy as they might be, don’t always come without risks; it is crucial to understand how nanomaterials interact with the environment, so that we might figure out a way to produce minimally invasive and highly sustainable nanomaterials. In this study, we used Gold Nanoparticles (GNPs) as a model. The objective of the study is to understand how different ligands attached to the GNPs affect their interactions with model membranes. Recent studies have shown that the surface chemistry of GNPs, particularly surface charge, has a significant impact on their interactions with model membranes and living organisms. For example, recent studies have shown that GNPs which are wrapped with cationic polyelectrolytes like poly (allylamine hydrochloride) (PAH) have high anti-bacterial activity. In our experiments, we used a new class of cationic polyelectrolytes, poly [oxanorbornenes] (PONs), which allows greater systematic variation of surface charge and hydrophobicity than PAH. UV-vis spectroscopy, Dynamic Light Scattering (DLS) spectroscopy, and Zeta Potential measurements were used to characterize the newly produced PONs-coated gold nanoparticles. The results of these measurements indicate that it is possible to control the surface charge of the nanoparticles by modifying their surface with various PONs. Experiments to determine the antimembranal activity of PONs-coated gold nanoparticles are on-going.
Phosphorylation is a post-translational modification (PTM) to a protein by which phosphates are added to the structure of the polypeptide. Among many other roles, phosphorylation regulates the activation of existing proteins and subsequently is fundamental for ideal physiological performance. The identification of the sites where phosphates bind is difficult without the presence of an enrichment protocol to increase the abundance of phosphoproteins. In this study, titanium dioxide (TiO$_2$) is used as an enrichment tool to identify all the known phosphate sites located on lysozyme. This protein is produced by animals and acts as an antimicrobial enzyme, forming part of the innate immune system. It is also chosen as a model protein because it has only two sites of phosphorylation and its molecular weight of 14.3 kDa makes it a relatively small protein. The goal of this research is to optimize the capture step of an existing protocol. Ultimately, this work will be combined with concurrent research with the goal of developing an optimized protocol for the capture of proteins of different lengths.

The enrichment from a mixture of controlled proteins is first performed through the use of a TiO$_2$ ziptip (ThermoFisher, A32993). Parameters of the capture step are varied to find optimal conditions. After capture, the sample is digested according to the protocol. The sample is then analyzed using high performance liquid chromatography (HPLC) coupled to a high resolution mass spectrometer (HRMS), allowing for high mass accuracy detection. The data is analyzed using PEAKS 7 software, providing sequence coverage and allowing for identification of the phosphorylated sites. The sequence coverage of each sample is analyzed to determine which procedure produces the highest sequence coverage and thus is determined to be optimal. Future research includes continued optimization of established protein analysis protocols to create a set of robust techniques for protein analysis.

Acknowledgements: I would like to thank Baltimore Youthworks Summer Jobs Program for funding my experience at the Summer Biomedical Training Program. I would also like to acknowledge the Molecular Characterization and Analysis Complex (MCAC) for their continuous support.
EXPLORING INDIVIDUAL VARIABILITY IN ESCALATION OF OPIATE INTAKE IN SIGN-TRACKERS AND GOAL-TRACKERS

Janai R. Williams1, Helen M. Nasser2, Guillermo Esber1 & Donna J. Calu 2
1 Department of Psychology, Brooklyn College, CUNY University of New York, Brooklyn, New York, USA, 11210.
2 Department of Anatomy and Neurobiology, School of Medicine at the University of Maryland Baltimore, Baltimore, Maryland, USA, 21201.

Prescription and nonprescription opioid abuse has steadily increased over the past two decades and has had a detrimental impact on the quality of life for the affected individuals. While many individuals try drugs of abuse, only a subset transition to addiction. Studying individual differences in addiction vulnerability is a critical step towards understanding the neurobiological underpinnings of motivation for drugs and their impact on the brain and behavior. Prior studies have indicated that variability in striatal dopaminergic signaling is related both to escalation of drug intake and to individual differences in natural reward approach known as sign- and goal-tracking phenotypes. Sign-tracking rats show heightened motivation for food-predictive cues and subsequently show greater cue-induced drug relapse than goal-trackers. The common neurobiological underpinnings of individual variability in both natural approach and escalation of drug intake suggest that differences in dopamine function prior to drug-exposure may underlie vulnerability to escalate drug intake. We then tested the hypothesis that individual variability in sign- and goal-tracking would predict escalation of heroin and food intake. We first used a Pavlovian lever autoshaping procedure to determine the sign- and goal-tracking phenotype. We then trained sign- and goal-tracking rats to self-administer heroin or food reward. In the intermittent access phase of self-administration training, rats learn to associate that heroin or food reward is only available for 5 minutes every half hour for three hour sessions. Prior work using intermittent access self-administration procedures has shown that variability in escalation of psychostimulant intake that maps onto the sign- and goal-tracking phenotypes. Currently, we are examining individual variability in the acquisition and escalation of heroin and natural reward intake in sign- and goal-tracking rats. This pilot study will allow us to determine whether variability in escalation of opiate intake and natural reward consumption relate to the sign- and goal-tracking phenotypes.

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