18th Annual Undergraduate Research Symposium in the Chemical and Biological Sciences

The College of Natural and Mathematical Sciences;
The Department of Chemistry and Biochemistry &
The Department of Biological Sciences

UMBC
AN HONORS UNIVERSITY IN MARYLAND

Sponsored by:

National Institute of General Medical Sciences of the National Institutes of Health (NIGMS/NIH)

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Schedule of Events

Time	Event
8:00 am	**SYMPOSIUM CHECK-IN & ON-SITE REGISTRATION** *Lobby, University Center, 3rd Floor*
8:00 am	**LIGHT CONTINENTAL BREAKFAST** *UC312, University Center, 3rd Floor*
9:00 am	**OPENING REMARKS & WELCOME ADDRESS** Dr. Freeman Hrabowski, President, University of Maryland, Baltimore County (UMBC) Dr. William R. LaCourse, Dean, College of Natural & Mathematical Sciences, UMBC *Meyerhoff Chemistry and Biochemistry Building, Lecture Hall 030*
9:45 am – 11:45 am	**MORNING POSTER SESSION** **Poster # 1-132 & 263 - 266** A - E: Biochemistry and Molecular Biology F – N: Biological Sciences O – V : Chemical Sciences STEM BUIILD at UMBC *Ballroom, University Center, 3rd Floor*
10:30 am	**WORKSHOPS**

Professional Communication: Making the Right Impression
Ms. Susan Hindle, Career Services, UMBC
UC312, University Center, 3rd Floor

Science, spiritual faith and truth: an interdisciplinary perspective
Dr. Stephen Freeland, Director, Interdisciplinary Studies
CASTLE, UC115D, University Center, 1st Floor

11:45 pm	**BUFFET LUNCH** *(gratis for registered guests with symposium name badge)* *The Commons – Main Street*
12:45 pm – 2:45 pm	**AFTERNOON POSTER SESSION** **Poster # 133 – 262 & 267-269** W – AA: Biochemistry and Molecular Biology BB – JJ: Biological Sciences KK – RR : Chemical Sciences *Ballroom, University Center, 3rd Floor*
1:30 pm	**WORKSHOPS** *Repeat of workshop titles and locations above*
3:00 pm	**PLENARY TALK** **"Emerging contaminants and resources: Environmental solutions to global problems"** Dr. Lee Blaney, UMBC, Department of Chemical, Biochemical and Environmental Engineering *Meyerhoff Chemistry and Biochemistry Building, Lecture Hall 030*
4:00 pm	**AWARDS PRESENTATION** *Meyerhoff Chemistry and Biochemistry Building, Lecture Hall 030*

Morning Poster Session
Group A – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

1. **COMPUTATIONAL STUDIES OF THE MOLECULAR BASIS OF COENZYME SPECIFICITY IN CLASS I AND CLASS II HMG-COA REDUCTASE**

Eri Arai and Yan Kung
Department of Chemistry, Bryn Mawr College, 101 North Merion Avenue, Bryn Mawr, PA 19010

2. **CHARACTERIZATION OF THE HIV-1 5'UTR MONOMERIC CORE**

Hannah Carter, Aishwarya Iyer, Joshua Brown, and Michael Summers
Department of Chemistry and Biochemistry, Howard Hughes Medical Institute, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

3. **UTILIZING STRUCTURAL AND ELECTRONIC ANALYSIS TO PREDICT THE EFFECTS (EFFECTIVENESS) OF PSEUDOPHOSPHORYLATION**

Kelly Daniels, Himal Ganguly, Anil Pandey, and Neal Zondlo
Department of Chemistry and Biochemistry, University of Delaware, 210 South College Avenue, Newark, DE 19711

4. **COMPUTATIONAL ANALYSIS OF THE MECHANISM OF THE UBIQUITIN CONJUGATING ENZYME UBC13**

Walker Jones, Aaron Davis, Serban Zamfir and Isaiah Sumner
Department of Chemistry and Biochemistry, James Madison University, 800 South Main Street, Harrisonburg, VA 22807

5. **CHARACTERIZATION OF 4-HISTIDINE/2-CARBOXYLATE MODEL PROTEINS**

Christine M. Philip and Amanda J. Reig
Department of Chemistry, Ursinus College, 601 E. Main Street, Collegeville, PA 19426

6. **CRYSTALLIZATION OF *E. COLI* GMP SYNTHATASE AND INSIGHT FROM THE STRUCTURES FORMED**

Jonathan Snyder[1] and Walter Patton[1,2]
[1]Program in Biochemistry & Molecular Biology, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003-1400
[2]Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003-1400

COMPUTATIONAL STUDIES OF THE MOLECULAR BASIS OF COENZYME SPECIFICITY IN CLASS I AND CLASS II HMG-COA REDUCTASE

Eri Arai and Yan Kung
Department of Chemistry, Bryn Mawr College, 101 North Merion Avenue,
Bryn Mawr, PA 19010

Isoprenoids are the largest and most structurally diverse class of natural products and are used in a wide range of medical and human health applications. The mevalonate (MEV) pathway is responsible for the conversion of acetyl-CoA into isopentenyl pyrophosphate (IPP) and dimethylallyl pyrophosphate (DMAPP), the precursors of isoprenoids and steroids. The first committed and rate-limiting step of the MEV pathway is the reduction of 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) to mevalonate, a reaction catalyzed by HMG-CoA reductase (HMGR). HMGR plays a key role in the production of isoprenoid natural products and cholesterol and is the sole enzyme target in widely available cholesterol-lowering statin drugs.

Coenzyme usage in the reduction reaction differs among class I and II HMGRs. The structural basis of coenzyme specificity in NADH-utilizing class II HMGR, such as HMGR from *Pseudomonas mevalonii* (PmHMGR) and *Streptococcus pneumoniae* (SpHMGR), has been hypothesized to involve a C-terminal domain that acts as a flexible flap for NADH binding. However, the structural basis of coenzyme specificity in NADPH-utilizing class I HMGR, present in human and yeast, remains unknown.

Comparative analysis between the C-terminal domains of both NADH- and NADPH-binding HMGR was performed using computational modeling programs to gain better molecular understanding of HMGR coenzyme recognition and specificity. Understanding the molecular basis of HMGR coenzyme specificity will enable the construction of modified enzymes that can be used for more efficient microbial production of isoprenoid drug compounds.

This research was funded by the Bryn Mawr College Summer Science Research program and the HHMI New Directions in Research and Teaching Grant.

CHARACTERIZATION OF THE HIV-1 5'UTR MONOMERIC CORE

Hannah Carter, Aishwarya Iyer, Joshua Brown, and Michael Summers
Department of Chemistry and Biochemistry, Howard Hughes Medical Institute, UMBC,
1000 Hilltop Circle, Baltimore, MD 21250

Human Immunodeficiency Virus Type 1 (HIV-1) patients are treated with a drug cocktail that targets various points in the viral life cycle. Unfortunately, this comes with myriad side effects and the possibility of resistance due to the high mutation rate of the virus. There is no drug available, however, that targets the translation or genome recognition portion of the lifecycle, which is character

ized by an equilibrium between the monomer and dimer conformations of the highly conserved 5' Leader (5'-L) in the HIV-1 RNA genome.

In order to study the structure of the monomer conformation using Nuclear Magnetic Resonance (NMR), 5'-L monomer must be isolated and reduced to its smallest functional core. The Primer Binding Site (PBS) region adds broad and crowded signals to our NMR spectrum, making the assignment of peaks difficult. We compared the wild type RNA construct to a construct in which the PBS region was removed to determine how the PBS region affects the dimerization of the RNA. We used gel shift assays in physiological conditions with varying incubation times and concentrations to determine this. So far, results indicate that our construct without PBS dimerizes similarly to our wild type, implying that the ΔPBS construct can act as an analog for the wild type in NMR studies.

This research was funded by NIH/NIGMS grant *1P50GM103297*, and 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. We would also like to thank our research team Seungho Choi, Eric Cormack, and Shyohyn Ghorbanpoor for their assistance.

UTILIZING STRUCTURAL AND ELECTRONIC ANALYSIS TO PREDICT THE EFFECTS (EFFECTIVENESS) OF PSEUDOPHOSPHORYLATION

Kelly Daniels, Himal Ganguly, Anil Pandey, and Neal Zondlo
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Pseudophosphorylation involves substituting a phosphorylated threonine or phosphorylated serine, existing predominantly in their dianionic form, with an aspartic acid or glutamic acid residue, which are inherently monoanionic. This substitution allows for peptide sequences to be mimicked in vitro; as phosphorylated residues are less stable and difficult to selectively phosphorylate and dephosphorylate without site-specific enzymes, pseudophosphorylation allows researchers to study structural and behavioral characteristics of relevant peptide sequences. The utilization of pseudophosphorylation is limited however, as it only successfully mimics phosphorylated serine and threonine in a fraction of its applications. Presently, no pattern has been observed to predict the success of pseudophosphorylation.

It is hypothesized by the Zondlo group that the success of pseudophosphorylation can be predicted. It has been observed that while glutamic or aspartic acid can closely mimic phosphorylated threonine or serine electronically, these substituted amino acids do not produce the same structural effects. Thus, in applications dependent on electronic effects, pseudophosphorylation is predicted to be effective, whereas it is expected to fail in applications dependent on structural conformation. It was observed that phosphorylated Ac-GPPXPPGY-NH_2 peptide, where X is S or T, takes on a polyproline helix conformation that pseudophosphorylation is not able to induce. It is further predicted that pseudophosphorylation will better be able to mimic phosphoserine than phosphothreonine.

Through solid phase synthesis, Ac-DP-OMe, Ac-EP-OMe, Ac-DL-OMe, and Ac-EL-OMe dipeptides have been synthesized. NMR and circular dichroism were used to analyze the structural and electronic shifts of the pseudophosphorylated dipeptides, for comparison to phosphorylated Ac-TP-OMe, Ac-SP-OMe, Ac-TL-OMe, and Ac-SL-OMe. Sequences were selected based on their prevalence in tau peptide, a protein associated with Alzheimer's disease. These findings will make it possible to predict whether pseudophosphorylation will be effective with applications to Alzheimer's research but furthermore it will aid in all research that utilizes pseudophosphorylation to study peptides in vitro.

Research reported in this poster was supported the National Institute of Health. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.

COMPUTATIONAL ANALYSIS OF THE MECHANISM OF THE UBIQUITIN CONJUGATING ENZYME UBC13

Walker Jones, Aaron Davis, Serban Zamfir and Isaiah Sumner
Department of Chemistry and Biochemistry, James Madison University, 800 South Main Street, Harrisonburg, VA 22807

Ubc13 is an E2 enzyme that catalyzes a post translational modification of proteins called lysine ubiquitination, i.e. the addition of ubiquitin to the lysine of a target protein via a thioester aminolysis reaction. Lysine ubiquitination is important because, one of its functions is to signal for the degradation of damaged proteins, and defects in ubc13 are linked to different disorders. The accepted mechanism for Ubc13-catalyzed ubiquitination is a stepwise mechanism that creates an oxyanion intermediate. This intermediate is hypothesized to be stabilized by a nearby asparagine residue, which is known as the "oxyanion hole." However, the validity of the accepted mechanism has come into question because, there has never been a comprehensive study of the ubiquitination mechanism, the accepted mechanism was inferred from the reverse reaction, and recent studies suggest a different role for the oxyanion hole. In our study, we use molecular dynamics to examine the hydrogen bond environment of the active site in two structures of Ubc13 and determine the likelihood for the formation of the oxyanion hole. Furthermore, we present initial data wherein we calculate the energies of the different possible steps of the reaction coordinate with the ONIOM quantum mechanics/molecular mechanics (QM/MM) extrapolation procedure.

Research for this project was funded by the National Science Foundation, grant numbers CHE-1062629 in 2013 and CHE-1461175 in 2015, The Extreme Science and Engineering Discovery Environment (XSEDE), which is supported by National Science Foundation grant number ACI-1053575, and The Thomas F. and Kate Miller Jeffress Memorial Trust, Bank of America, N.A., Trustee.

CHARACTERIZATION OF 4-HISTIDINE/2-CARBOXYLATE MODEL PROTEINS

Christine M. Philip and Amanda J. Reig
Department of Chemistry, Ursinus College, 601 E. Main Street, Collegeville, PA 19426

The relationship between a protein's structure and function is not very well understood. One way we study this relationship is by using a *de novo* model protein because it is a simplified version of a natural protein and is easy to mutate and synthesize in the lab. We are focused on studying the class of binuclear non-heme di-iron enzymes because these proteins are all very similar in structure but carry out different functions. We are working on creating a model for *myo*-inositol oxygenase (MIOX) which plays a role in glycol oxygenation. Our protein model is based on the four-helix bundle G4DFsc protein, which was created as a model system for di-iron carboxylate enzymes. The E11H/E44H double mutant has the same ratio of histidine to carboxylate amino acids as the MIOX active site. To understand the effects of each individual amino acids in directing

function, we also studied the single mutants E11H and E44H. Since the computer-model showed steric clashing with the new mutations we also studied E11H/Y51E and E11H/E44H/Y51E/L81D variants which contained potentially stabilizing mutations. To see how these mutations affected the function, metal binding assays with iron and cobalt were performed, as well as reactivity assays to observe the oxidase and oxygenase properties. Our results showed that the stabilizing mutations did not preserve the 2-to-1 metal-to-protein binding ratio that we would expect to see in the di-iron enzymes. All of our protein variants were able to bind to iron and perform oxidase reactions. None of these proteins were able to perform oxygenase reactions, as expected.

We would like to thank the National Institute of Health for research support (R15-GM110657 to AJR).

CRYSTALLIZATION OF *E. COLI* GMP SYNTHATASE AND INSIGHT FROM THE STRUCTURES FORMED

Jonathan Snyder[1] and Walter Patton[1,2]
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Online access of this abstract is restricted at the request of the Principal Investigator.

Morning Poster Session
Group B – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

7. **DELETION ANALYSIS OF A NOVEL REGULATORY GENE IN *CANDIDA ALBICANS* THAT SWITCHES ITS TARGET PATHWAYS**

Ashi Arora[1], Michelle Hudson[1], and Robert Akins[2]
[1]Wayne State University, 42 W. Warren Avenue, Detroit, MI 48202
[2]Department of Biochemistry and Molecular Biology, Wayne State University School of Medicine, 540 E. Canfield Street, Detroit, MI 48201

8. **INVESTIGATING ANOMALIES IN OUTER MEMBRANE PROTEIN FOLDING: FOLDED AGGREGATE AND PHANTOM OmpA**

Mariama Diallo and Alison Dewald
Department of Chemistry, Salisbury University, 1101 Camden Avenue, Salisbury, MD 21801

9. **NEOFUNCTIONALIZATION OF *CANDIDA GLABRATA* PMU3 AS A THIAMINE PHOSPHATASE**

Pamela Myers, Zefanne Bergado, Ashley Gonzalez, and Kelly Orlando
Department of Biology, Immaculata University, 1145 King Road, Immaculata, PA 19345

10. **CREATION AND CHARACTERIZATION OF RUBRERYTHRIN AND SYMERYTHRIN MODEL PROTEINS**

Jenna Pellegrino[1], Katherine Bell[1], Rachel Z. Polinski[1], Sabrina N. Cimerol[1], Ari Jacbos[2], Edward I. Solomon[2], and Amanda J. Reig[1]
[1]Department of Chemistry, Ursinus College, 601 E. Main Street, Collegeville, PA 19426
[2]Department of Chemistry, Stanford University, Stanford, CA

11. **RIBOSOMAL PROTEIN L4 BINDING DURING RIBOSOMAL RNA MATURATION**

Rebekah Rashford, Jesse Fox, and Lasse Lindahl
Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, Maryland 21250

12. **DETERMINING THE ROLES OF LSM PROTEINS IN *C. ELEGANS* SEX MUSCLE FATE SPECIFICATION**

Neta Shwartz[1], Stephen Sammons[2] and Jun Kelly Liu[2]
[1]Department of Biological Sciences, Towson University, Baltimore, MD 21252
[2]Department of Molecular Biology and Genetics, Cornell University, Ithaca, NY 14853

13. **UNDERSTANDING C/EBP β, A TRANSCRIPTION FACTOR EXPRESSED DOWNSTREAM IN NEUROINFLAMMATORY EVENTS MEDIATED BY HMGB-1**

Camille Werzowa, Beatriz Tenorio, and Christine McCauslin
Department of Science, Mount St. Mary's University, 16300 Old Emmitsburg Road, Emmitsburg, MD 21727

DELETION ANALYSIS OF A NOVEL REGULATORY GENE IN *CANDIDA ALBICANS* THAT SWITCHES ITS TARGET PATHWAYS

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Wayne State University School of Medicine, 540 E. Canfield Street, Detroit, MI 48201

Candida albicans is an opportunistic fungal pathogen, causing widespread vaginal infections and life threatening infections in immunocompromised patients. Resistance may arise by mutations in genes encoding regulatory proteins. More often, resistance occurs by altered regulation rather than mutation. One such regulatory gene is ZCF36 which controls MDR1 expression. The MDR1 gene encodes a multidrug efflux pump. When overexpressed, it pumps out fluconazole to confer resistance. Many point mutations within ZCF36 make the protein hyperactive so that MDR1 is expressed at high levels; such mutants are resistant, but very specifically to fluconazole only. We have shown that a truncation of the carboxy-terminal portion of ZCF36, in which the inhibitory domain resides, confers resistance, but paradoxically not to fluconazole, rather to all echinocandins, when introduced on a high copy number plasmid. These antifungals, in clinical use, target a completely different pathway- cell wall biosynthesis by FKS1-encoded beta-glucan synthase. The hypothesis is that the full length gene will activate MDR1 and cause fluconazole resistance, but that truncated genes will not activate MDR1 and will cause echinocandin resistance. An intact ZCF36 gene and at least two truncated versions were cloned into a high copy number plasmid via PCR. These amplicons and putative clones have been generated. When introduced back into *C. albicans*, these cloned genes will overexpress their encoded products to alter host phenotype. Then, these transformants will be tested for resistance to the two classes of antifungals, and RT-qPCR will be used to measure levels of expression of the cloned genes, MDR1, and select genes in the ergosterol and cell wall biosynthesis pathways. If resistance is conferred, it is likely that the truncated protein is binding to novel gene targets and represents a tool for defining an echinocandin resistance pathway.

This project was funded by and performed in the Akins Lab.

INVESTIGATING ANOMALIES IN OUTER MEMBRANE PROTEIN FOLDING: FOLDED AGGREGATE AND PHANTOM OmpA

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Outer membrane proteins (OMPs) are difficult to study because they must first be folded into something resembling a lipid bilayer. One current method for folding OMPs is by the rapid dilution of the OMP from denaturant to pre-made liposomes. While investigating folding conditions, our lab encountered two mysteries: 1.) folded OmpA in a fraction expected to be pure aggregate and 2.) visually diminishing amounts of OmpA ("phantom OMP") common in literature figures and our own experimental protein folding studies over time. To study the nature of the folded aggregate, we are developing a phosphate assay to test for lipids in the aggregate sample. Phosphate assays are being optimized to produce a standard curve and are performed using diPC12 lipids and OmpA. OmpA was also studied in relation to phantom OMP, a phenomenon where protein appears to "disappear" over time, as visualized by SDS-PAGE. The amount of protein visualized was quantified via gel densiometry. To investigate where the protein could be going we set up experiments testing lipids of differing chain lengths to examine the correlation between chain length and the likelihood of the protein "vanishing". Flash frozen experiments tested the hypothesis that OmpA was folding in the SDS loading buffer and vortexed samples tested the hypothesis that OmpA was aggregating at the bottom of the tube.

We gratefully acknowledge funding through the National Science Foundation Bridges for SUCCESS Summer Research Experience at Salisbury University.

NEOFUNCTIONALIZATION OF *CANDIDA GLABRATA* PMU3 AS A THIAMINE PHOSPHATASE

Pamela Myers, Zefanne Bergado, Ashley Gonzalez, and Kelly Orlando
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Candida glabrata is evolutionarily related to the well-characterized budding yeast *Saccharomyces cerevisiae*, which is commonly utilized in baking and in alcohol fermentation. However, *C. glabrata* lacks homologs for the phosphatases required for a number of vital pathways in *S. cerevisiae*, including thiamine (Vitamin B1) uptake. Survival of single-celled organisms is contingent on this pathway; therefore, there must be other enzyme(s) replacing this necessary function in *C. glabrata*. The Wykoff lab at Villanova University has uncovered a family of genes in *C. glabrata* (PMU1, PMU2, and PMU3) created by gene duplication whose members seem to have neofunctionalized in order to replace various missing phosphatases. Preliminary studies from the Wykoff lab and from our lab suggest that PMU3 encodes an enzyme that can remove phosphate from TPP, a phosphorylated form of thiamine, which then allows it to be taken into the cell. Our objective for this study is to uncover the amino acid changes in the ancestral PMU sequence necessary to generate this novel PMU3 activity. We are testing the phosphatase activity of various fusions of PMU2 (which does not share this neofunctionalized activity) and PMU3 to narrow down the region(s) that confer thiamine phosphatase activity. Once a smaller region is identified we can create point mutations in PMU2 or in the fusions to determine the individual amino acids that confer thiamine phosphatase activity. Our goal is to determine the vital regions of the *C. glabrata* PMU3 gene necessary for its neofunctionalized enzymatic activity.

CREATION AND CHARACTERIZATION OF RUBRERYTHRIN AND SYMERYTHRIN MODEL PROTEINS

Jenna Pellegrino[1], Katherine Bell[1], Rachel Z. Polinski[1], Sabrina N. Cimerol[1], Ari Jacbos[2], Edward I. Solomon[2], and Amanda J. Reig[1]
[1]Department of Chemistry, Ursinus College, 601 E. Main Street, Collegeville, PA 19426
[2]Department of Chemistry, Stanford University, Stanford, CA

The ferritin-like superfamily (FLSF) is a class of proteins that contain a diiron active site and participate in important biochemical pathways, including fatty-acid desaturation and the formation of deoxyribonucleotides. The canonical FLSF sequence contains four carboxylate and two histidine metal-binding ligands in the active site. However, the rubrerythrins (Rbr) and symerythrins (Sym) contain one and two additional carboxylate residues, respectively, in their active sites. Interestingly, these proteins also exhibit enhanced reactivity with hydrogen peroxide relative to other members of the FLSF, but the correlation between the additional carboxylate residues and the altered functionality is currently not well understood.

To investigate this phenomenon, models for Rbr and Sym were created based on G4DFsc, which is a small, *de novo*-designed 4-helix bundle protein that mimics the canonical structure and reactivity of FLSF enzymes. Carboxylate residues, either aspartate (D) or glutamate (E), were introduced at positions G14 and/or G47 to generate Rbr- and Sym-like active sites within the G4DFsc bundle. Metal-binding, protein-folding, and reactivity assays have been used to characterize the geometric and electronic structures of these systems and provide insight into how these particular carboxylate residues in the G4DFsc active site affect its ability to react with hydrogen peroxide.

This work was supported by the NIH (R15-GM110657 to A.J.R.) and the NSF (MCB-1404866 to E.I.S.).

RIBOSOMAL PROTEIN L4 BINDING DURING RIBOSOMAL RNA MATURATION

Rebekah Rashford, Jesse Fox, and Lasse Lindahl
Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, Maryland 21250

Ribosomal RNA (rRNA) and ribosomal proteins are integral parts of the ribosome complex found in all organisms. These particles undergo an extensive synthesis process in which rRNA and ribosomal proteins join together to from mature ribosomes. Much research has been conducted to determine the structure of mature ribosomes (through x-ray crystallography) and the maturation process of rRNA, but at which point the ribosomal proteins join the pre rRNAs is still yet to be better understood.

Using the large subunit (60S) ribosomal protein L4 as the protein of interest, we studied when during the maturation process of rRNA did L4 bind to one of the pre-rRNAs. By utilizing and optimizing co-immunoprecipitation assays specific for the nine amino acid hemagglutinin (HA) protein tag, we were able to target the point at which HA-L4 binds to the pre-rRNA.

Using northern blot analysis and DNA probes specific for the latest pre-60S rRNAs, we found that L4 binds to the 27S and 7S pre-rRNA segments, meaning that L4 binds prior to the C2 cleavage site being cut. The next step is to use different probes on L4 to determine whether L4 is found at earlier cleavage sites (such as A2 or A3). Using this information, we will be able to indicate L4s first encounter with pre-rRNA during the maturation process.

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.

DETERMINING THE ROLES OF LSM PROTEINS IN *C. ELEGANS* SEX MUSCLE FATE SPECIFICATION

Neta Shwartz[1], Stephen Sammons[2] and Jun Kelly Liu[2]
[1]Department of Biological Sciences, Towson University, Baltimore, MD 21252
[2]Department of Molecular Biology and Genetics, Cornell University, Ithaca, NY 14853

A central problem in developmental biology is to understand the process of cell differentiation. *C. elegans* provides an advantage to studying cell differentiation because of its powerful genetics and the availability of a complete cell lineage. The *C. elegans* hermaphrodite egg-laying apparatus consists of the vulva, the egg-laying sex muscles, and the egg-laying neurons that innervate the sex muscles. All the sex muscles are derived from the post-embryonic M lineage and include four type I and four type II vulval muscles, as well as four type I and four type II uterine muscles. An RNAi screen previously completed in the lab revealed genes whose knockdown gives defects in vulval muscle cell fate specification. One of these genes is *lsm-4*, which encodes a protein member of a highly conserved family of genes encoding RNA binding proteins proposed to function together in a ring complex to regulate RNA processing. To determine if other *lsm* family members of *C. elegans* are involved in M lineage cell fate specification, we targeted these genes by RNAi, and analyzed the resulting vulval muscle phenotype.

Thank you to Kelly Liu for her constant attention to detail and care to help me with this project. Thank you to Steve Sammons for mentoring me while in the Liu lab, assisting me with RNNAi techniques and genetic crosses, and answering my numerous questions. Additional thanks go to the Liu lab for their abundant support. Thank you to Volker Vogt, William Brown, Anne Dunford, and the MBG REU program. This research is funded by NIH Grant GM066953 and the Cornell MBG REU program.

UNDERSTANDING C/EBP β, A TRANSCRIPTION FACTOR EXPRESSED DOWNSTREAM IN NEUROINFLAMMATORY EVENTS MEDIATED BY HMGB-1

Camille Werzowa, Beatriz Tenorio, and Christine McCauslin
Department of Science, Mount St. Mary's University, 16300 Old Emmitsburg Road, Emmitsburg, MD 21727

The CCAAT/enhancer binding protein (C/EBP) family of transcription factors is composed of 6 members. When activated, they facilitate transcription of their target genes whose products play a role in cell differentiation, growth, and inflammation. During infection, sterile injury, or through the course of a neurodegenerative disease, High Mobility Group Box 1 (HMGB-1), which is functionally important in regulating transcriptional processes, is rapidly released from necrotic cells into its surroundings where it acts as a cytokine. Studies have shown that the release of HMGB-1 causes the activation of glial cells for an inflammatory response. C/EBP proteins, and specifically C/EBP β, are involved in promoting expression of pro-inflammatory gene products and are upregulated in activated glial cells. Our laboratory is investigating whether the activated transcription factor C/EBP β is a downstream target of HMGB-1 mediated inflammation events. Our hypothesis is that HMGB-1 will activate the glial cells in an inflammatory response, which, in turn, will lead to the pre-existing, and now activated, C/EBP β to be translocated to the nucleus, where it will bind and activate expression of pro-inflammatory gene targets. We have performed time course treatments and Western Blots to examine the effects of HMGB-1 exposure on C/EBP β nuclear localization in a mixed astrocyte cell line derived from primary cells infected with a J2 virus. We noticed an increase in C/EBP β protein levels in our nuclear extracts following HMGB-1 treatment, as well as transcriptional activation of certain pro-inflammatory genes through qPCR. Quantitation of nuclear C/EBP β levels indicates a 2-3 fold increase 24-48 hours after HMGB-1 treatment. Following this information, our laboratory is working on identifying HMGB-1 receptors to understand how it activates our cells.

Morning Poster Session
Group C – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

14. **INCREASED PHARMALOGICAL SELECTIVITY OF COX and LOX ISOFORMS**

Moneeba Khan, Karan Arora, Earl Benjamin, Ellis Benjamin
School of Natural Sciences and Mathematics, Stockton University, 101 Vera King Farris, Galloway, NJ 08205

15. **CLONING THE FULL-LENTH GASTRIN GENE FOR UP-REGULATION IN PANCREATIC CANCER STUDIES**

Victoria C. Koehler, Michael W. Stephan, and John F. Harms
Department of Biological Sciences, Messiah College, 1 College Avenue, Mechanicsburg, PA 17055

16. **TOWARDS SOLVING HIGH RESOLUTION STRUCTURE OF TITIN ZIG9/10**

Allyn Letourneau and Nathan Wright
Department of Chemistry and Biochemistry, James Madison University, 800 South Main Street, Harrisonburg, VA 22807

17. **ALTERED PYRUVATE KINASE MRNA EXPRESSION AND ABNORMAL METABOLIC PROFILES IN MELANOMA CELLS EXPRESSING THE WARBURG EFFECT**

Jonathan McKinney[1,2], Teofilo Borunda[1], and Todd Thompson[1]
[1]Department of Pharmacology, University of New Mexico, Albuquerque, NM 87131
[2] Department of Chemistry, McDaniel College, 2 College Hill, Westminster, MD 21157

18. **MUTAGENESIS OF THE UNSTRUCTURED FELINE IMMUNODEFICIENCY VIRUS MATRIX PROTEIN C-TERMINUS TO IMPROVE STABILITY**

Colin O'Hern, Janae Baptiste, and Michael F. Summers
Department of Chemistry and Biochemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

19. **THE RELATIONSHIP BETWEEN THE 15DKA SELENOPROTEIN AND THE EXPRESSION OF CANCER REGULATORY GENES IN COLON TUMORIGENESIS**

Chukwuka Onyewu[1], Angelica Patterson[1], Jessica Canter[1], Bradley Carlson[2], Cindy Davis[3], Vadim Gladyshev[4], Dolph Hatfield[2], and Petra Tsuji[1]
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[4]Brigham and Women's Hospital, Harvard Medical School, 77 Avenue Louis Pasteur, HMS New Research Building Room 435, Boston, MA 02115

20. **MEASURING METHYLATION OF GNG11 IN HUMAN BREAST CANCER**

Jennifer Young and William Schwindinger
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INCREASED PHARMALOGICAL SELECTIVITY OF COX and LOX ISOFORMS

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Chronic regional and systemic inflammation promotes diseases including Osteoarthritis, Rheumatoid arthritis, Inflammatory Bowel Disease, and Asthma. Although multiple treatments such as corticosteroids and NSAIDs are used for acute flare-ups, long term control is limited by the side effects of these treatments. These side effects include hyperglycemia, insulin resistance, diabetes mellitus, nausea/vomiting, diarrhea, hypertension, and gastric ulceration/bleeding. Currently, research into Cyclooxygenase 2 (COX-2)'s inhibition as a long-term mitigation of inflammation, has lead scientist to search for increased efficacy with the decreased side effects associated with current inhibition of Arachidonic Acid oxygenating proteins. Such examples of these innovative treatments include the addition of NO donating groups to NSAIDs, inhibition of potent inflammation signaling molecules such as TNF-alpha, or dual inhibitors which act on multiple pathways. The goal of this research was to discover new potent anti-inflammatory pharmaceuticals which followed this dual pathway model, by way of inhibition of the COX and LOX isoforms for an increase potentiation of their anti-inflammatory effect. Specifically, this research sought to discover COX-2 and Lipoxygenase-5 (LOX-5) inhibiting isoforms while limiting interactions with the COX-1 protein. This research used two computational docking programs, IGemDock and Open Eye Docking (Hybrid), to determine specific binding ratios between COX1/2 and LOX-5. Direct comparison of molecular structure used VROCS. Based on these comparisons, several molecules were identified.

CLONING THE FULL-LENTH GASTRIN GENE FOR UP-REGULATION IN PANCREATIC CANCER STUDIES

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Online access of this abstract is restricted at the request of the Principal Investigator.

TOWARDS SOLVING HIGH RESOLUTION STRUCTURE OF TITIN ZIG9/10

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Titin domains ZIg9/10 bind to obscurin domains Ig58/59 during myofibrillogenisis. Mutations in this region lead to hypertrophic cardiomyopathy (HCM) in humans. While the cellular consequences of this interaction are well characterized, the molecular determinants governing this structure are unknown. Previous work from our lab has solved the high-resolution structure of the obscurin domains of the complex. Here, we describe the purification and preliminary structure characterization of titin domains ZIg9/10.

Jeffress Memorial Fund, Research Corporation Cottrell College Grant, NSF-REU (CHE-1461175)

ALTERED PYRUVATE KINASE MRNA EXPRESSION AND ABNORMAL METABOLIC PROFILES IN MELANOMA CELLS EXPRESSING THE WARBURG EFFECT

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Many cancer cells preferentially undergo lactic acid fermentation from glycolysis rather than oxidative phosphorylation. Even with adequate amounts of oxygen to metabolize glucose that could yield higher amounts of energy, cancer cells produce lactic acid in a process called the Warburg effect. Moreover, melanoma cancers can exhibit excessive lactic acid production, as was found in the SKMEL19 and SKMEL29 melanoma cell lines that exhibit the Warburg effect. Phenotypic changes that may facilitate the Warburg effect include alterations in the expression of splice variants of pyruvate kinase (PK), which performs the final step in glycolysis, and increased expression of lactate dehydrogenase A (LDH-A), the enzyme responsible for producing lactic acid from pyruvate. Surprisingly, in the current study, gene expression measured using QPCR showed decreased levels of LDH-A mRNA in SKMEL19 and SKMEL29 melanoma cells compared to cancer cells that do not undergo this process (i.e., PC3 and SKMEL103 cells). However, increased LDH protein expression was observed in these cells. Inhibition of LDH via oxamic acid led to decreased growth. Levels of PK mRNA isoforms PKM1 and PKM2 were also determined, the latter being characteristic of cancers exhibiting the Warburg effect. The ratio of PKM2 to PKM1 mRNA was found to be increased in SKMEL19 and SKMEL29 cells, ultimately suggesting PKM2 as a key player in mediating the Warburg Effect. Our results suggest that cancer cells exhibiting the Warburg effect have developed highly coordinated metabolic adaptations that promote cancer growth.

Acknowledgement Statement: This work was supported by the UNM NIH CTSA Grant UL1TR000041. We give thanks to program director; Dr. Jennifer Gillette and program coordinator; Elaine Manzanilla for their direction and supervision throughout the summer. The authors also thank Kirsten White for assistance with melanoma cell lines.

MUTAGENESIS OF THE UNSTRUCTURED FELINE IMMUNODEFICIENCY VIRUS MATRIX PROTEIN C-TERMINUS TO IMPROVE STABILITY

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Feline immunodeficiency virus (FIV) is a retrovirus, similar to human immunodeficiency virus type 1 (HIV-1) in humans, that suppresses and inhibits activity of the immune system in cats. Studying FIV is important because humans and cats have similar immune responses, making cats a plausible animal model for development of HIV-1 treatment. Both viruses feature the Gag polyprotein (Gag), which consists of the following major domains: matrix (MA), capsid, and nucleocapsid. MA is responsible for targeting Gag to the plasma membrane, a step that is vital to the retroviral replication cycle. HIV-1 MA is a well-characterized protein, but limitations in FIV MA stability have impeded long-term experiments by means of nuclear magnetic resonance spectroscopy (NMR). A long, unstructured region at the C-terminus of FIV MA may aid in protein instability that imposes challenges. It is believed that truncating the C-terminus of MA will improve protein stability and facilitate ability to perform long-term studies. To excise this unstructured tail, mutagenesis is conducted to perform the deletion and truncate the unstructured C-terminal tail. Preliminary mutagenesis data indicates successful C-terminus truncation of FIV MA, and sodium dodecylsulfate polyacrylamide gel electrophoresis data suggest that the truncated protein is over-expressed and purified. Implications of this work include the facilitation of further structural analysis of FIV MA by means of NMR and support for application of felines as HIV-1 animal models.

Howard Hughes Medical Institute, NIH/NIAID 5R37AI030917

THE RELATIONSHIP BETWEEN THE 15DKA SELENOPROTEIN AND THE EXPRESSION OF CANCER REGULATORY GENES IN COLON TUMORIGENESIS

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In a model of chemically-induced colon cancer, our previous study showed that mice lacking the 15kDa selenoprotein (Sep15) developed fewer azoxymethane-induced aberrant crypt foci (ACF), which are considered pre-neoplastic lesions, than control mice. In this subsequent study, we investigate the ability of Sep15 knockout mice to develop colon tumors in a model of inflammatory carcinogenesis. As expected, most of our control mice developed many ACF and many small to medium-sized tumors when exposed to azoxymethane with subsequent dextran sulfate sodium treatment. However, much to our surprise, even though our Sep15 knockout mice only developed few ACF, these mice lacking Sep15 systemically developed a similar number of colon tumors as their litter mate controls. To further elucidate these findings, we have isolated mRNA from tumors of both control and Sep15 knockout mice, as well as from normal colon epithelium. Using real-time RT-PCR, we are in the process of comparing the mRNA expression of various genes suspected in the regulation of tumorigenesis in Sep15 knockout mice compared to controls. Preliminary findings indicate that tumor tissues of both Sep15 knockout mice and controls displayed a higher expression of glutathione peroxidases 1 and 2 than in respective normal colon epithelium. Interestingly, mRNA expression of interferon-γ-regulated guanylate binding protein 1 is significantly higher expressed in colon epithelia and tumors of Sep15 knockout mice compared to controls. The contribution of interferon-γ-regulated genes on the regulation of Sep15 remains to be further elucidated.

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MEASURING METHYLATION OF GNG11 IN HUMAN BREAST CANCER

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An area of current cancer research is epigenetic regulation, which investigates changes that do not affect the sequence of DNA. DNA methylation was studied in this project because high levels have been identified in different cancers, and correlated with down regulation of expression in certain genes. Human breast cancer has been shown to have low expression levels of GNG11 and therefore was predicted to have a highly methylated promoter. GNG11 is transcribed and subsequently expressed as a gamma subunit of a G protein. These G proteins are coupled to receptors and function in cell-to-cell communication, breakdown of which can lead to unregulated growth, and potentially tumors. Two regions of the promoter were studied in this project. The methods used to compare the DNA involved using restriction enzymes that were either sensitive to methylation or not. The products were then amplified using PCR and viewed by gel electrophoresis to compare the intensity of the bands and determine a percent level of methylation. The DNA was also treated with sodium bisulfite, then amplified by PCR and sequenced which allowed an examination of the exact sites of methylation and changes that occurred. The restriction digest method failed to show differences in promoter methylation. However the restriction enzymes may not have completely cut the DNA. The bisulfite method found significantly higher levels of methylation at 2 of 3 sites examined in the upstream region of the promoter and 1 out of 8 in the downstream. Based on the results GNG11 was found to have more methylation of the promoter in breast cancer when compared to the adjacent normal tissue and we would predict the expression of this gene to be lowered.

Morning Poster Session
Group D – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

21. **USING CRISPR-CAS9 GENE-EDITING AND GENETICALLY-ENCODED SENSORS TO MODEL NEURODEGENERATION *IN VITRO***

Grisilda Bakiasi[1], Yun Li[2], Laure Freland[2], Maisam Mitalipova[2],
Rudolf Jaenisch[2], and Julien Muffat[2]
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22. **SYNTHESIS AND CHARACTERIZATION OF CROTAMINE AND PEG ADDUCTS FOR APPLICATION AS DRUG CARRIERS**

Andrew Butler, Giovanni Marino, and Richard Karpel
Department of Chemistry and Biochemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

23. **UNDERSTANDING THE ROLE OF CELLULAR PHOSPHORYLATION ON HSF1 AGGREGATION AND TRANSCRIPTIONAL ACTIVITY**

Austin Maduka[1], Giorgio Gaglia[2], Luke Whitesell[2], and Susan Lindquist[2,3]
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Cambridge, MA, 02139

24. **PROTEIN-PROTEIN INTERACTIONS BETWEEN THE LECTIN-LIKE DOMAIN OF THROMBOMODULIN AND COMPLEMENT COMPONENT 3**

Shelby Marchese, Nathan Fritzinger, and Julia R. Koeppe
Department of Chemistry, Ursinus College, 601 E. Main Street, Collegeville, PA 19426

25. **HOMOLOGY MODELING OF CLASS A GPCRS IN THE INACTIVE STATE**

Matthew Skorski[1], Alessandro Deplano[1], Brett Habermehl[1], Mary Mendoza[1],
Jessica Dawson[1], Jia Gao[1], and Stefano Costanzi[1,2]
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Washington, DC 20016
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26. UNDERSTANDING PROTEIN-PROTEIN INTERACTIONS OF ACYL CARRIER PROTEINS USING AN INFRARED ACTIVE THIOCYANATE PROBE

Kathleen Tsai, Grace Thiele, Casey Londergan, and Louise Charkoudian
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27. BIOCHEMICAL CHARACTERIZATION OF THE INTERACTION BETWEEN AN INNATE IMMUNE RECEPTOR NOD2 AND ITS CHAPERONES

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USING CRISPR-CAS9 GENE-EDITING AND GENETICALLY-ENCODED SENSORS TO MODEL NEURODEGENERATION *IN VITRO*

Grisilda Bakiasi[1], Yun Li[2], Laure Freland[2], Maisam Mitalipova[2],
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Neurodegeneration diseases such as Parkinson's or Alzheimer affect millions of individuals. Induced pluripotent stem cells enable us to model these disorders by differentiating them into any cell of interest, including neurons, the electrically active cells of the brain, and glia, whose crucial roles are only recently becoming apparent, after decades of being considered mere "glue".

The aim of our project was to introduce disease-relevant genetic modifications to wild-type human pluripotent stem cells, or to "correct" mutations in diseased cells. I generated constructs carrying the bipartite system CRISPR/Cas9. These constructs, when introduced by transfection into hiPS/hES cells, allow very specific cleavage of the genome at a given locus. Faulty repair mechanisms further ensure that small insertions or deletions (mutations) are introduced in the process. Concurrent delivery of a homologous sequence allows replacement of the endogenous sequence with another. We focused on alleles known to be strongly associated with or causal of various neurodegenerative disorders (including AD and ALD).

In order to assess electrophysiological properties in 2- and 3-dimensional cultures (which may better recapitulate in vivo tissue organization), we used the latest generation of genetically-encoded calcium sensor, GCAMP6. Similarly, we focused on fluorescent sensor HyPer to monitor the contribution of free radicals and oxidative stress to pathologies. To allow efficient delivery of these transgenes to neurons and glia, which are notoriously difficult to transfect, I engineered lentivirus expression vectors carrying these genes into the FUW backbone.

All vectors were cloned, verified by restriction digest and Sanger sequencing, and tested for expression in human 293T cells. These vectors are currently being tested in human iPS cultures. They will help us tease out the role of mutations in several diseases, and allow mechanistic study of the involvement various cell types in neurodegeneration (for example, the effects of astrocytes or microglia on neuronal survival).

Firstly, I would like to express my sincere gratitude to my mentor, Julien Muffat, for the continuous support for my summer project: for his patience, motivation, and immense knowledge. I could not have imagined having a better mentor. My sincere thanks also goes to Dr. Jaenisch, who provided me the opportunity to join his laboratory, and Howard Hughes Medical Institute for their summer funding. Last but not least, I would like to thank my mentor, Gregory Davis, for all of his great advice and support.

SYNTHESIS AND CHARACTERIZATION OF CROTAMINE AND PEG ADDUCTS FOR APPLICATION AS DRUG CARRIERS

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Crotamine is a protein found in the venom of the rattlesnake *Crotalus durissus terrificus,* and has been shown to bind DNA and negatively-charged cell membranes. The biological activity of crotamine has potential as an anticancer agent as previous studies have demonstrated its selective preference for binding cancer cells compared to healthy cells. An effort to make use of this targeting action involves linking crotamine to gold nanoparticles. The central gold particle can serve as a hub to attach additional targeting proteins or other medical compounds into a single combined unit of delivery.

In order to link crotamine to the nano particles, a modified version of polyethyleneglycol (PEG), orthopyridyldisulfide-polyethyleneglycol-succinimidylvalerate (OPSS-PEG-SVA), was employed, as it has been used previously for linking to gold for both crotamine and other proteins. This study focused on the reaction of crotamine with PEG, before attachment to the gold particle.

In an effort to determine efficient methods of synthesis as well as to characterize the products, multiple reactions were performed with varying ratios of crotamine to PEG. The results were analyzed by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) to visualize the ratios of products. High ratios of PEG to crotamine (6:1) resulted in larger yields of adducts. Ultraviolet spectrophotometry (UV-Vis) and fluorescence were also used to assess the binding characteristics of the crotamine-PEG products. The negatively-charged polysaccharide, heparan sulfate, has been implicated in crotamine-cell membrane interaction. Pure crotamine competitively interferes with the binding of a similar polysaccharide, heparin, to the dye, azure A. Although the attachment of PEG to the protein lowers this binding affinity, the crotamine-PEG adducts clearly possess significant binding activity.

Future experiments will involve continuing the sequence of synthesis to attach the crotamine-PEG products to gold nanoparticles, and further characterize the properties of crotamine in conjunction with these compounds.

UNDERSTANDING THE ROLE OF CELLULAR PHOSPHORYLATION ON HSF1 AGGREGATION AND TRANSCRIPTIONAL ACTIVITY

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Heat-Shock Factor 1 (HSF1) is a central regulator of the proteotoxic stress response in eukaryotes. Acting as a transcription factor, HSF1 triggers the expression of heat shock genes, such as cellular chaperones, that help maintain protein homeostasis. Recent work from the Lindquist lab has shown that HSF1 is overexpressed and active in cancer cells, where it regulates a different set of genes to promote and sustain malignancy, making it a novel therapeutic target. Upon heat stress, HSF1 is activated and hyperphosphorylated, forming foci in the nucleus. However, little is known about the many kinases that phosphorylate HSF1 to control activation. In this study, a comprehensive library of kinase inhibitors was screened to test their effect on HSF1 activity by analyzing foci formation over time. Time-lapse live-cell microscopy on a human cancer cell line expressing fluorescently tagged HSF1 was used to measure HSF1 concentration and cellular localization in time. Cells were incubated in kinase inhibitors for three hours and then treated with Velcade, a proteotoxic drug that induces HSF1 foci formation. Automated image analysis in MATLAB was used to measure foci formation during drug treatment. In this screening, 12 of 276 drugs were found to cause alterations to foci formation. Since foci formation is correlated with HSF1 activity, drugs that down-regulated HSF1 aggregation were tested for effects on HSF1 transcriptional function. Changes in transcription levels in basal and heat shock conditions were measured through luciferase reporter assays and quantitative PCR. The compound PIK-75—an inhibitor of DNA-Protein Kinase and Phosphotidylinositol-3-Kinase—was found to change transcript levels of heat shock genes produced during stress, as well as an effect on HSF1 foci formation. These findings will ultimately help shed light on the pathways regulating the activity of this transcription factor, as well as help understanding its role in carcinogenesis.

This investigation was supported (in part) by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences.

PROTEIN-PROTEIN INTERACTIONS BETWEEN THE LECTIN-LIKE DOMAIN OF THROMBOMODULIN AND COMPLEMENT COMPONENT 3

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Protein-protein interactions are vital to the proper functioning of numerous biological systems. Thrombomodulin (TM) is a protein that is involved in the down-regulation of coagulation induced by the clotting protein thrombin. Complement component 3 (C3) is a vital component of the complement system, which is involved in innate immunity against bacteria and viruses. However, dysregulation of C3 can lead to the degradation of host cells.

Evidence suggests that the lectin-like domain of TM, which protrudes into the bloodstream from the host's epithelial cells, may interact with active C3 (C3b) to inactivate it, thus preventing host cell degradation. The research conducted herein required the expression and purification of the lectin-like domain of TM, called TMD1, in yeast cells and the isolation and purification of C3 from bovine blood plasma.

To aid in specific immobilization of the TMD1, two lysine residues were converted to methionine by site-directed mutagenesis via polymerase chain reaction to create two mutant TM proteins, K147M and K86/147M. To quantify possible structural disruption caused by the mutations, analytical tests with circular dichroism (CD) spectroscopy and urea unfolding were carried out. A pull down assay was then performed with the wild-type and mutant proteins to determine protein-protein interactions. Once interactions are confirmed, hydrogen/deuterium exchange followed by matrix assisted laser desorption and ionization time of flight mass spectrometry (MALDI-TOF MS) will be performed to determine which amino acids are involved in binding.

HOMOLOGY MODELING OF CLASS A GPCRS IN THE INACTIVE STATE

Matthew Skorski[1], Alessandro Deplano[1], Brett Habermehl[1], Mary Mendoza[1], Jessica Dawson[1], Jia Gao[1], and Stefano Costanzi[1,2]
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G protein-coupled receptors (GPCRs) constitute a large superfamily of membrane-spanning proteins that, in humans, comprises about 1000 members. They are activated by a wide range of natural ligands and are targeted by many of the marketed and leisure drugs.

Hence, there is great interest in the experimental elucidation and the theoretical modeling of their three-dimensional (3D) structures, which can be employed as platforms to discover or design new ligands that can modulate their activity. To date structures have been solved for a total of 31 receptors, 26 of which belonging to Class A. Most of the solved structures reflect the inactive state of receptors. Although GPCR structural studies are in rapid expansion, a thorough experimental characterization of all the receptors is not feasible due to the large size of the superfamily. However, the available experimental structures can be used as templates for the construction of three-dimensional models of other family members through homology modeling.

With the present work, we are adding a layer of measurability to the line of findings relative to the modellability of Class A GPCRs in their inactive state. In particular, we are aiming at quantitatively studying the relationships between the sequence identity between templates and modeled receptors and the accuracy of resulting homology models. Moreover, we are aiming at comparing the differences in the accuracy of the resulting from the application of different alignment strategies.

To address these questions, we constructed models of a reference GPCR, namely the β_2 AR, using as templates all the other 25 Class A GPCRs that, to date, have been solved in the inactive state. The subsequent comparison of the models with a reference experimental structure of the β_2 AR solved in the inactive state, followed by a correlation study with sequence identity data, are yielding the answers to our questions.

The acknowledge American University, the Schwartz Fellowship of the Chemistry Department, and the Mathias Fellowship of the College of Arts and Sciences for funding.

UNDERSTANDING PROTEIN-PROTEIN INTERACTIONS OF ACYL CARRIER PROTEINS USING AN INFRARED ACTIVE THIOCYANATE PROBE

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Natural product biosynthesis research seeks to understand how organisms synthesize metabolites. Understanding the bacterial enzymes responsible for synthesizing polyketides and fatty acids is important for engineering bacteria to produce metabolites for biomedical and environmental applications. Enzymes involved in natural product biosynthesis exhibit specificity in their protein-protein interactions. In polyketide and fatty acid synthases, acyl carrier proteins (ACP) utilize a phosphopantetheine arm to transfer substrate between enzymes, resulting in elongation and tailoring of the growing natural product. ACPs are highly conserved and are the catalytic center of natural product synthases. Thus, ACPs are crucial bioengineering targets for combinatorial biosynthesis efforts. The installation of an infrared active thiocyanate probe onto the phosphopantetheine arm of an ACP may be useful for detecting protein-protein interactions between an ACP and a binding partner via vibrational spectroscopy. Using ketosynthases (KSs) and ACPs from the deoxyerythronolide B synthase (DEBS) and *E. coli* fatty acid synthase (FAS), we sought to explore the utility of an infrared active thiocyanate probe in detecting interactions between enzymes.

The thiocyanate probe was installed onto the phosphopantetheine arm of ACPs using a simple cyanylation reaction. We confirmed successful installation of the probe using infrared spectroscopy. Circular dichroism spectroscopy confirmed that the probe did not disrupt the structure of the ACP. Isothermal calorimetry was used to quantify binding affinity between KSs and cyanylated ACPs. Finally, fast protein liquid chromatography and non-reducing protein gels determined that the thiocyanate probe may catalyze covalent cross-linking between the *E. coli* fatty acid ACP and KS, a process that may correlate with the enzymes' abilities to communicate with each other.

In the future, we will attempt to quantitatively understand how protein interactions between enzymes affect the infrared vibrational frequencies of the thiocyanate probe. We would also investigate the use of thiocyanate probes in other systems like non-ribosomal peptide synthetases.

This project was funded by the Haverford College Koshland Integrated Natural Science Center and the B.A. Rudolph Foundation. We would like to thank Lou Charkoudian, Casey Londergan, and the Haverford College Department of Chemistry for all of their support and mentorship throughout this process.

BIOCHEMICAL CHARACTERIZATION OF THE INTERACTION BETWEEN AN INNATE IMMUNE RECEPTOR NOD2 AND ITS CHAPERONES

Hannah C. Wastyk[1], Catherine L. Grimes[1,2], Amy Schaefer[1], Ching-Wen Hou[1], Mackenzie Lauro[1], and Vishnu Mohanan[2]
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Online access of this abstract is restricted at the request of the Principal Investigator.

Morning Poster Session
Group E – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

28. **IDENTIFYING THE MINIMUM CATALYTIC FRAGMENT OF *PSEUDOMONAS* EXOTOXIN A**

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29. **INVESTIGATING INTERACTIONS BETWEEN THE LECTIN-LIKE DOMAIN OF THROMBOMODULIN AND COMPLEMENT COMPONENT 3**

Thomas Holt and Julia R. Koeppe
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30. **IMPROVEMENT OF THE PURIFICATION PROCESS OF THE *NEISSERIA MENINGITIDIS* SEROGROUP W-135 CAPSULE POLYMERASE ENZYME**

Kayla Powell, Ophelia Ukaegbu, Jayda Smith and Pumtiwitt McCarthy
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Baltimore MD 21251

31. **ALTERATION OF PSEUDOMONAS PUTIDA β-HBDH COFACTOR SPECIFICITY**

Jorna Sojati, Connor Ott, Nadia Galchak, and Jennifer B. Palenchar.
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32. **MEASUREMENT OF ACTIVITY FOR SELENOCYSTEINE-CONTAINING MOTIFS**

Jay Subramoney, Dominic Santoleri, Rujin Cheng, and Sharon Rozovsky
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33. **PROTEOMIC ANALYSIS OF YEAST HISTONE METHYLTRANSFERASE SET5**

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34. **CLONING NEUREXIN 3A cDNAs REPRESENTING ALTERNATIVELY SPLICED ISOFORMS IN DANIO RERIO**

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IDENTIFYING THE MINIMUM CATALYTIC FRAGMENT OF *PSEUDOMONAS* EXOTOXIN A

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Pseudomonas exotoxin A (PE) is a 67 kDa protein produced by the bacterium *Pseudomonas aeruginosa*. PE has 3 structural domains with individual functions. The N-terminal domain I is divided into two non-sequential but structurally adjacent domains Ia (residues 1-252) and Ib (365-404). Domain II (253-364) is located between domains Ia and Ib. The remaining residues make up domain III (405-613). Functionally, domain I binds to a cell surface receptor, while domain II is involved in intracellular trafficking and domain III has enzymatic activity. PE acts on cells by internalization through receptor-mediated endocytosis, intracellular trafficking to the cytosol, and inactivation of elongation factor 2 (EF2), an essential component of protein synthesis. Inactivation of EF2 halts protein synthesis and ultimately leads to cell death.

Inactivation of EF2 is catalyzed by a fragment of the toxin that transfers an ADP-ribose group from NAD^+ to EF2. Previous research has proposed the catalytic fragment to be residues 395-613 of PE, including some of domain Ib and all of domain III, but no study has rigorously and systematically explored the minimum size of this fragment that retains activity. In order to do this we have begun by using site-directed mutagenesis to make amino- and carboxy-terminal truncations of PE. We plan to test these truncations in an *in vitro* protein synthesis assay to evaluate their catalytic activity. Future directions include using the minimal catalytic fragment of PE to explore the intracellular trafficking of PE and develop antibody-targeted PE fusion proteins for the treatment of cancer.

We would like to thank Dr. John Weldon for proctoring us throughout this project and also for being a constant source of information and guidance. Thanks also to Earl Brooks, whose effort in this project should not go unmentioned. Special thanks to the Fisher College of Science and Mathematics at Towson University for supporting this research.

INVESTIGATING INTERACTIONS BETWEEN THE LECTIN-LIKE DOMAIN OF THROMBOMODULIN AND COMPLEMENT COMPONENT 3

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Protein-protein interactions are vital to the proper functioning of numerous biological systems. Interactions between the lectin-like domain of thrombomodulin (TMD1) and the complement system may provide a link between coagulation and inflammation. Complement component 3 (C3) is at the center of three different modes of activation of the complement system and may provide a good target for regulation of the system, which is involved in innate immunity against bacteria and viruses. However, dysregulation of C3 can lead to the degradation of host cells. Evidence suggests that the lectin-like domain of TM (TMD1) may interact with active C3 (C3b) to inactivate it, thus preventing host cell degradation. C3 has been isolated and purified from bovine blood plasma, and TMD1 was expressed in Pichia pastoris and then purified. A protein pull-down assay was used to verify that the two proteins interact. Hydrogen/deuterium exchange followed by matrix assisted laser desorption and ionization time of flight mass spectrometry (MALDI-TOF MS) was performed and the data from those studies is currently under analysis to determine which regions of each protein are involved in binding. Additionally, interactions between the two proteins will be characterized using fluorescence resonance energy transfer (FRET) experiments. Complement component 4 (C4), which is one step above C3 in the complement cascade and the center of two modes of activation for the complement system, has also been purified and may be used in future studies.

IMPROVEMENT OF THE PURIFICATION PROCESS OF THE *NEISSERIA MENINGITIDIS* SEROGROUP W-135 CAPSULE POLYMERASE ENZYME

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Neisseria meningitidis causes most cases bacterial meningitis worldwide. This bacterium has twelve serogroups each with different capsular polysaccharides. The main purpose is to improve the purification process for the enzyme that makes these sugars in *Neisseria meningitidis* serogroup W-135. We would like to obtain pure protein to study the activity of this enzyme. Our methods include cell lysis, sonication, and nickel affinity column chromatography. The purity of the protein was tested by gel electrophoresis. Initial results showed more protein of interest in the wash buffer, but not as much protein in the elution buffers. The protein was present, but we were unable to obtain all or most of the pure protein. During column chromatography, we changed the method to include six elution buffers with different amounts of imidazole (25mM, 50mM, 75mM, 125mM, 250mM, and 500mM) in each. These results indicated some protein was in the wash buffer, but most of the protein was in the 25mM imidazole and 50mM imidazole elution buffers. The activity of the protein during purification was tested using a fluorescent acceptor DMB-trimer. The fluorescent dye DMB was chemically attached to a trimer (3 linked together) of sialic acid. Activity of the enzyme was followed using thin layer chromatography (TLC) under UV light. The protein had more activity than the control sample after 1 hour and after 2 hours based on TLC. For future work we will continue to improve our process to obtain purer protein of interest from the nickel column.

Louis Stokes Alliance for Minority Participation in Research Program (K.P.)
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American Chemical Society Project SEED Program (J.S.)
K.P. and O.U. contributed equally to this work.

ALTERATION OF PSEUDOMONAS PUTIDA β-HBDH COFACTOR SPECIFICITY

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Prior work has revealed that trypanosome β-hydroxybutyrate dehydrogenase (β-HBDH) can utilize NADP(H), unlike nearly all other β-HBDHs characterized. Using the trypanosome enzymes as a guide, we sought to alter the cofactor specificity of an NADH-dependent β-HBDH. In the work presented, we describe amino acid changes in the Rossman fold region of an NADH-dependent bacterial β-HBDH from *Pseudomonas putida*. The histidine-tagged recombinant enzymes were overexpressed, purified from *E. coli*, and characterized kinetically. In the bacterial β-HBDH, one amino acid change is sufficient to loosen specificity, while a combination of mutations allows for cofactor preference reversal. The kinetic characterization of these mutant enzymes will be presented. Long term, we seek to use this information to alter the cofactor specificity of the parasite enzyme in vivo to better understand the role of this enzyme in the trypanosomes.

We gratefully acknowledge support for this work from the Villanova University Department of Chemistry and the Villanova University Research Support Grant program.

MEASUREMENT OF ACTIVITY FOR SELENOCYSTEINE-CONTAINING MOTIFS

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Online access of this abstract is restricted at the request of the Principal Investigator.

PROTEOMIC ANALYSIS OF YEAST HISTONE METHYLTRANSFERASE SET5

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Histone methylation is a type of post-translational modification, used to regulate the way in which DNA is packaged in chromosomes. This packaging, called chromatin, is important for the regulation of gene expression and DNA repair. Methylation is mediated by histone methyltransferases, which often work in large complexes to alter chromatin and regulate gene expression.

A relatively uncharacterized methyltransferase found in budding yeast, Set5, has been shown to methylate lysine residues on histone H4, and may be responsible for responses to cellular stress. This is the first known enzyme to methylate these targets, and as such, its broader cellular function is important to characterize, for understanding the purpose of these novel epigenetic events. Here, we use a structure-function approach to characterize the binding sites on Set5 that mediate its interaction with H4 and other proteins. We also use a biochemical approach to investigate the proteins associated with Set5 in cells, to lend insight to the pathways in which it is involved. Preliminary data from analysis by mass spectrometry show hundreds of potential protein partners, including many involved in transcription and stress responses, such as carbohydrate and lipid metabolism, which will be explored further in future experiments.

CLONING NEUREXIN 3A cDNAs REPRESENTING ALTERNATIVELY SPLICED ISOFORMS IN DANIO RERIO

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Cell fate specification is the process by which cells of equivalent developmental potential become committed, specialized, different cell types. We seek to better understand this process by examining the genetic regulation of blood cell specification in the model organism zebrafish, and our work has identified a novel gene *neuer1* as an important regulator of this process.

Through protein domain analyses, we realized that a prominent protein domain is shared between Neuer1 and a family of cell-signaling ligands known as Neurexophilins, raising the question whether Neuer1 performs a similar function. Neurexophilins bind to cell-surface receptor proteins known as Neurexins, further raising the possibility that Neuer1 may bind to one of the Neurexins as well. Our goal is to determine which, if any, of the Neurexins is bound by Neuer1 by comparing the expression pattern of each *neurexin* to that of *neuer1*.

In order to determine the expression patterns of *neurexins* using *in situ* hybridization, we attempted to clone *neurexin* cDNAs to be used as templates to synthesize riboprobes that can specifically recognize each of the *neurexin* mRNAs. This proved to be challenging, since there are 6 zebrafish *neurexin* genes, each of which can generate hundreds of mRNA isoforms due to extensive alternative splicing. To circumvent having to clone thousands of cDNA isoforms, we devised a strategy to clone cDNA fragments that represent either common exons shared by all isoforms or alternatively spliced exons that are shared by a small group of isoforms. Here we report this strategy as well as our successful cloning of a group of *neurexin 3a* cDNAs that represent alternatively spliced exons. Analyses of these clones will contribute to the emerging understanding of *neurexin* alternative splicing; utilization of these clones in *in situ* hybridization will provide valuable clues to the expression patterns of different *neurexin 3a* isoforms.

This research project was funded by the McDaniel College Student-Faculty Collaborative Summer Research Fund and the Department of Biology.

Morning Poster Session
Group F - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

35. **OVEREXPRESSION OF NAR1.2 AND LCI1 IN *CHLAMYDOMONAS REINHARDTII***

Courtney Bass[1], Francis Ayehfor[2], Rose Gbemefu[3], Amrita Madabushi[1,] and Stephen M. Miller[2]
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[2]Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

36. **THE EPSILON SUBUNIT OF DNA POLYMERASE III IN THE BACTERIAL RESPONSE TO QUINOLONES**

Kelly DiGeronimo, Amanda Finck, and Zakiya Whatley
Department of Biology, Gettysburg College, 300 N. Washington Street, Gettysburg, PA 17325

37. **EXAMINING THE ROLE OF MATRIX ELASTICITY ON MEGAKARYOCYTIC DIFFERENTIATION OF HEMATOPOIETIC STEM CELLS**

Pragyan Khanal[1,3], Jinlin Jiang[2,3], and Eleftherios T. Papoutsakis[1,2,3]
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38. **DEVELOPMENT AND INVESTIGATION OF A NEW FLUOROGEN ACTIVATING PROTEIN, J3, FOR THE PRODUCTION OF A PROTEASE BIOSENSOR IN A NEW COLOR**

Vivian Pham, Matthew J. Farber and Peter B. Berget
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39. **NOVEL ANTIBACTERIALS FROM DIVERSE OCEAN SPONGES: ONE HALOGEN MAKES A DEADLY DIFFERENCE!**

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40. **INFLUENCE OF FIBER ORIENTATION ON POLARIZATION AND MIGRATION OF FIBROBLAST AND FIBROSARCOMA CELLS**

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OVEREXPRESSION OF NAR1.2 AND LCI1 IN *CHLAMYDOMONAS REINHARDTII*

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Grown under optimal conditions algae have the ability to use free resources (CO_2 and sunlight) to grow rapidly as well as produce significant amounts of biomass that can be converted into biofuel to be used as an alternative to fossil fuels. The aim of this project is to improve the growth rate of *Chlamydomonas reinhardtii*. This green alga is a model organism for lab research because its whole nuclear genome is known, and many molecular genetic tools make it easy to manipulate. We are focusing our efforts on the carbon concentrating mechanism (CCM), which regulates CO2 uptake in the organism, and more specifically on two CO_2 transporters, LCI1 and NAR1.2. LCI1 (low CO_2 induced) localizes in the plasma membrane and NAR 1.2 functions in the chloroplast envelope. Our overall goal is to overexpress these proteins by ligating the coding sequences for the LCI1 and NAR1.2 genes into *C. reinhardtii* nuclear expression vector pARG and then transforming into *C .reinhardtii*. Thus far we have succeeded in generating the expression vectors for both LCI1 and NAR1.2, and have obtained transformants for the NAR1.2 vector. Multiple lines of the transformed algae were cultured and are being tested by western blot analysis. We will do growth curve and biomass accumulation analyses on the best expressing strains. If we are successful in improving algal growth by overexpressing NAR1.2 and/or LCI1, the next step will be to express these enzymes in the *Chlorella vulgaris*, a green alga that is related to *C. reinhardtii* but a much better commercial production organism.

The 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. The program is funded by a grant (REM supplement to NSF-EFRI-1332344) from the National Science Foundation (NSF) Directorate for Engineering (ENG) Office of Emerging Frontiers in Research and Innovation (EFRI).

THE EPSILON SUBUNIT OF DNA POLYMERASE III IN THE BACTERIAL RESPONSE TO QUINOLONES

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Online access of this abstract is restricted at the request of the Principal Investigator.

EXAMINING THE ROLE OF MATRIX ELASTICITY ON MEGAKARYOCYTIC DIFFERENTIATION OF HEMATOPOIETIC STEM CELLS

Pragyan Khanal[1,3], Jinlin Jiang[2,3], and Eleftherios T. Papoutsakis[1,2,3]
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Megakaryocytes (Mk) are the precursor to platelets and differentiate from $CD34^+$ hematopoietic stem cells (HSC) found *in vivo* in the bone marrow. Platelets function with coagulation factors to stop bleeding after injury or vessel damage. Due to their short 3-5 day shelf life, as well as other factors, worldwide supply of platelets is low, despite an increasing demand for platelet transfusions. The production of platelets *ex vivo* is still very inefficient, as many factors, such as matrix elasticity, are involved and not all are understood. Matrix elasticity suggests an important mechanism in cell signaling and influencing differentiation of hematopoietic stem and progenitor cell subsets by directing cell differentiation. HSCs face different elasticity levels in the bone marrow microenvironment during Mk development and maturation. The role of this substrate level mechano-sensitivity on Mk differentiation, however, remains largely unexplored.

In this study, we examined megakaryocytes cultured from human $CD34^+$ HSCs that were seeded on polyacrylamide (PA) hydrogels of different elasticities and coated with extracellular matrix protein fibrinogen (FGN) to explore the role of elasticity on Mk DNA synthesis and polyploidization. Furthermore, we explored proplatelet & Mk microparticles (MkMP) formation across the different hydrogels. No significant differences in each analysis were found, possibly due to low cell attachment on PA hydrogels. Initial testing of polydimethylsiloxane (PDMS) gels, however, yielded more cell attachment and promising results in the role of matrix elasticity on proplatelet & MkMP formation.

This project was supported by the Ronald E. McNair Post-Baccalaureate Achievement Program and the Delaware INBRE Summer Scholars Program, with a grant from the National Institute of General Medical Sciences - NIGMS (8 P20 GM103446-15) from the National Institutes of Health.

DEVELOPMENT AND INVESTIGATION OF A NEW FLUOROGEN ACTIVATING PROTEIN, J3, FOR THE PRODUCTION OF A PROTEASE BIOSENSOR IN A NEW COLOR

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Fluorogen activating proteins (FAPs) are derived from recombinant single chain antibodies, which comprise two parts: a variable heavy chain (Vh) and variable light chain (Vl) connected by a flexible linker of Glycine and Serine residues. These FAPs can bind to specific dyes, inducing their fluorescence. By mixing and matching Vh and Vl domains, certain combinations are inhibitory to dye binding and thus do not fluoresce. Insertion of a protease cleavage sequence into the flexible linker enables the creation of a protease biosensor. In this model, a protease cleaves the linker, the inhibitor domain disassociates, and the FAP binds to the dye, inducing fluorescence. J3 is a newly identified FAP that binds to the dye alpha-cyano-thiazole orange (α-CNTO). This dye fluoresces a green-yellow color at the wavelength of approximately 565nm and thus will permit construction of a biosensor of a new color.

In order to evaluate J3 as a candidate biosensor, restriction enzyme digestion was used to isolate the individual heavy and light chain J3 genes. It was determined that the variable light chain, J3Vl, was capable of binding dye on its own and causing fluorescence. Various different Vh domain genes were then cloned next to the J3Vl domain to test for inhibition of dye binding. J3Vl in conjunction with H4Vh showed little to no fluorescence when measured in the presence of dye. Finally, a linker was engineered in between H4Vh and J3Vl domains to contain a protease cleavage site to test the ability of J3 to be used a protease biosensor of a novel color. The experiment showed that while H4Vh was a successful inhibitor, it did not disassociate fully from the J3Vl domain to induce a significant level of fluorescence.

I would like to thank Dr. Peter Berget and Dr. Matthew Farber for guidance throughout this project. I would also like to thank Dr. Bruce Armitage of Carnegie Mellon University for providing the fluorogenic dye used in this experiment. This project was funded by NIH grant U54- RR022241, as well as the Melvin Firman Undergraduate Research Grant by the University of the Sciences.

NOVEL ANTIBACTERIALS FROM DIVERSE OCEAN SPONGES: ONE HALOGEN MAKES A DEADLY DIFFERENCE!

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INFLUENCE OF FIBER ORIENTATION ON POLARIZATION AND MIGRATION OF FIBROBLAST AND FIBROSARCOMA CELLS

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Morning Poster Session
Group G - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

41. **A COMPARISON OF DNA EXTRACTION METHODS USING *GROMPHADORHINA PORTENTOSA* TISSUE**

Esther Apraku Bondzie and Kenneth G. Sossa
Department of Biology, Notre Dame of Maryland University, 4701 North Charles Street, Baltimore, MD 21210

42. **CHARACTERIZATION OF NANOPARTICLES USING ATOMIC FORCE MICRSCOPY (AFM)**

Chloe A. Kwon, Margaret E. LaCourse, Ian W. Shaffer, Joshua A. Wilhide, and William R. LaCourse
Molecular Characterization and Analysis Complex, UMBC, 1000 Hilltop Circle, Baltimore MD 21250

43. **BIODISTRIBUTION STUDIES OF NEAR-INFRARED LABELED NANOEMULSIONS IN A RAT CCI PAIN MODEL**

Ryan Sanders[1, 2], Emily Nehl[1, 2], Muzamil Saleem[1, 2], Andrea Stevens[1, 2], Jelena Janjic[2, 3], and John Pollock[1, 2]
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[2]Chronic Pain Research Consortium, Duquesne University, Pittsburgh, PA 15203
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44. **CHARACTERIZATION OF THE PUPILLARY LIGHT REFLEX (PLR) IN PATIENTS RECEIVING ULTRA-LOW DOSE KETAMINE INFUSION**

Raissa Audrey Tseumie[2, 3], Kevin Jackson[1,2], Ben Geenspun[2], David Strum[2], Brendon O'Neil[2], and Julia Finkel[1,2]
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45. **THE PERFORMANCE OF IRRITANT-EXPOSED WILDTYPE AND SKN-1A KNOCKOUT MICE IN A COOKIE TEST WITH ODOR BACKGROUND**

Chantel Wilson, Kayla Lemons, and Weihong Lin
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A COMPARISON OF DNA EXTRACTION METHODS USING *GROMPHADORHINA PORTENTOSA* TISSUE

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To understand how genes not only affect structure and function but also behavior, we use the *Gromphadorhina portentosa*, the Giant Madagascar Hissing Cockroach. Specifically, we are interested in genes expressed in the nervous system of this roach, and will therefore need to extract DNA. DNA extractions have been widely performed in insects such as *Drosophila*; however, given the size and thickness of *G. portentosa's* chitinous exoskeleton a clear protocol is required. To determine the most efficient method for extracting the DNA of this cockroach, three methods were used: Pure Link Genomic mini kit (Invitrogen), extraction using DNAzol (Invitrogen) and extraction using SDS and phenol/chloroform/isoamyl alcohol solution. DNA concentrations were determined using a NanoDrop Spectrophotometer. Results showed that extraction using the SDS and phenol/chloroform/isoamyl alcohol solution was the most efficient, providing the largest amount of DNA. Further work is needed to optimize DNA yield but this is the first step towards exploring the genome of this particularly useful model. Future studies will involve knocking down the expression of specific proteins using siRNA to test their role in experience-mediated neuronal plasticity.

Funded by CFR&D of Notre Dame of Maryland University

CHARACTERIZATION OF NANOPARTICLES USING ATOMIC FORCE MICRSCOPY (AFM)

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Evolution of microscopy over the years has greatly improved imaging. In particular, 1986 marked the start for a method of microscopy known as atomic force microscopy (AFM). Using a probe with a tip radius of around 30 nm at the end that is responsible for scanning the surface of the sample, AFM allows users a wider range of applications with minimal sample preparation. A laser beam is deflected off the tip onto a photodiode, which measures the specific mechanical movements of the probe to produce an image of the sample. AFM offers three modes of operation (i.e., tapping, contact, and non-contact) to maximize the options of applications. For instance, AFM is capable of imaging proteins, blood cells, bacteria, viruses, polymers, metals, and minerals.

The goal of this research is to characterize nanoparticles using an Axiovert 100 BioScope II and NanoScope Analysis software. Nanoparticles have extraordinary strength and conductivity and can be utilized in many fields of applications, such as high-powered batteries or in sensors that help detect cancer. Imaging analysis with AFM allows for the particle characterization of height, volume, and three-dimensional topography in a timely, cost effective manner. This research will focus on optimizing sample preparation methods and imaging procedures for a variety of samples, while testing the capabilities and parameters of the AFM instrument. Future applications for the AFM will extend to imaging atomic level structures of a biological nature.

On behalf of the MCAC, I would like to thank Bruker for their assistance with instrumentation and troubleshooting. Furthermore, I would like to personally thank the members of the MCAC for allowing me the opportunity to utilize their facility and instruments to further my understanding and experience as an undergraduate researcher.

BIODISTRIBUTION STUDIES OF NEAR-INFRARED LABELED NANOEMULSIONS IN A RAT CCI PAIN MODEL

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CHARACTERIZATION OF THE PUPILLARY LIGHT REFLEX (PLR) IN PATIENTS RECEIVING ULTRA-LOW DOSE KETAMINE INFUSION

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Ketamine is an analgesic and sedative drug used to treat several chronic pain syndromes. It is used in clinical therapy among patients with opioid tolerance, acute hyperalgesia, and chronic neuropathic pain. Ultra Low Dose ketamine (ULD ketamine) (<0.5 mg/kg/hr) serves to reverse the central sensitization of the nervous system that occurs with neuropathic pain states via NMDA antagonist activity. This therapeutic intervention relieves pain without causing prolonged sedation and respiratory depression. This study aims to characterize the impact of ULD ketamine infusion on the eight parameters that characterize the pupillary light reflex (PLR), through pupilometer measurements taken every two hours from patients in a controlled environment. Each patient is submitted to two assessments per subject, one in the right and one in the left eye is performed with 30seconds between each measurement. We hypothesize that patients will exhibit repeatable changes in PLR parameters upon administration of ULD ketamine every two hours over the eight-hour infusion time. Initial results show noticeable changes in specific PLR parameters upon ULD ketamine administration. For example, the percentage maximum pupil diameter of patient I decreased from 17.5% to 8.7% after 4 hours, to 5.3% after 6 hours and back up to 10.5% after 8 hours. Ongoing and future research will examine the mechanism behind the PLR baseline changes of patients receiving ULD and assess accurate information guiding dosing parameter for patient with chronic pain.

I would like to thank my mentor and PI Dr.Finkel. Thank you to the Sheikh Zahed Institute for Pediatric Surgical Innovation, Children's National Medical Center for funding. I am grateful to Kevin Jackson for guidance and assistance in data collection and interpretation.

THE PERFORMANCE OF IRRITANT-EXPOSED WILDTYPE AND SKN-1A KNOCKOUT MICE IN A COOKIE TEST WITH ODOR BACKGROUND

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Morning Poster Session
Group H - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

46. **CHARACTERIZATION OF CANDIDATE MORBID MUTATIONS IDENTIFIED IN TWO PATIENTS WITH MYCOBACTERIAL OR SEVERE VIRAL DISEASES**

Austin Gabel[1], Serkan Belkaya[2], Jacinta Bustamante[2], Emmanuelle Jouanguy[2], Stéphanie Boisson-Dupuis[2], and Jean-Laurent Casanova[2]
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[2]St. Giles Laboratory of Human Genetics and Infectious Diseases, The Rockefeller University, New York, NY

47. **EXAMINATION OF RETINOIC ACID-INDUCED TRANSGLUTAMINASE ACTIVITY IN NEURITE OUTGROWTH**

Abigail M. Samuelsen and Kristen L. Boeshore
Department of Biology, Lebanon Valley College, 101 College Avenue, Annville, PA 17003

48. **THE ROLE OF N-LINKED GLYCOSYLATION IN DROSOPHILA DEVELOPMENT**

Morgan Thomas and Erica M. Selva
Department of Biological Sciences, University of Delaware, 210 South College Avenue, Newark, Delaware 19711

49. **RNA INTERFERENCE OF DEVELOPMENTAL GENES IN THE PEA APHID EMBRYO**

Maritza Vazquez-Trejo, Erin Bonner, and Gregory Davis
Department of Biology, Bryn Mawr College, 101 N. Merion Avenue, Bryn Mawr, PA 19010

50. **PHYSIOLOGICAL ESTROGENS EXERT OPPOSING EFFECTS ON HEART RATE IN ZEBRAFISH EMBRYOS**

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CHARACTERIZATION OF CANDIDATE MORBID MUTATIONS IDENTIFIED IN TWO PATIENTS WITH MYCOBACTERIAL OR SEVERE VIRAL DISEASES

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EXAMINATION OF RETINOIC ACID-INDUCED TRANSGLUTAMINASE ACTIVITY IN NEURITE OUTGROWTH

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Retinoic acid (RA) is known to increase transglutaminase activity after a nervous system injury has occurred[1,2,3]. There are two isoforms of TG2, a short and a long form that arise from alternative splicing. The two isoforms differ in their ability to bind GTP, which can turn off catalytic activity in the long form. Our questions are: Does RA treatment of injured PC12 cells promote neurite regeneration? As well as: Does RA increase TG-2 transamidation activity in injured PC12 cells? The prediction to both questions is yes. To answer these questions, we differentiated PC12 cells that were mechanically injured then treated with RA or RA + ionomycin. A neurite outgrowth assay was performed and the neurite outgrowth was observed. Then a TGase functional assay was performed, incorporating 5-BAP into target proteins. The proteins were detected with avidin-peroxidase. After running our far-western blot on the STORM imager a second time at a higher sensitivity, bands were visible, verifying that our technique is sensitive enough to detect transamidation activity. Future plans will be to expand the study to compare RA treatment to NGF treatment and addition of treatment lengths.

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THE ROLE OF N-LINKED GLYCOSYLATION IN DROSOPHILA DEVELOPMENT

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Asparagine-linked or N-linked glycosylation is an important post-translational modification pathway that adds a 14-sugar oligosaccharide to target proteins on the luminal side of the endoplasmic reticulum. These glycan "tags" are necessary for multiple cell functions including cell-cell recognition, protein trafficking, and proper protein folding. The focus of this study is to examine the effect of loss of function mutations in the N-linked pathway. A group of human diseases, congenital disorders of glycosylation (CDG), arise from mutations in the genes involved in various steps of this pathway. While CDGs display pleotrophic phenotypes, most include neuronal defects. The two specific genes under study in this project are *alg9* and *alg10.* Each encode a glycosyltransferase that adds a sugar residue to the growing oligosaccharide chain. The *Drosophila* eye is used as a model organ to study the effects of these genes on neuronal development. In adult flies, these mutations yield a small rough eye phenotype, which is more severe in *alg9,* as it acts five steps before *alg10* in the pathway. In order to determine the basis of this phenotype, larval eye discs were dissected, stained for different glycoprotein and neuronal markers, and then imaged using confocal microscopy. We found these mutations interrupt proper glycoprotein trafficking, as Chaoptin accumulates in the cell bodies of *alg9* and *alg10* mutant photoreceptors. Caspase-3 staining showed this accumulation eventually leads to photoreceptor apoptosis. Photoreceptor death likely continues through pupal development resulting in a reduced number of photoreceptors in *alg10* adult eyes and almost complete absence in *alg9* adult eyes. These results indicate that CDG patients may have normal neuronal specification and differentiation, but experience neuronal deficits due to intracellular accumulation of glycoproteins leading to cell death. These data suggest markers of endoplasmic reticulum stress and the unfolded protein response should be examined in the future.

RNA INTERFERENCE OF DEVELOPMENTAL GENES IN THE PEA APHID EMBRYO

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Aphids are hemipteran insects that have evolved alternative methods of reproduction and development in response to environmental cues without undergoing genetic change. We are interested in understanding how the same genome has adapted to allow for both sexual and asexual reproduction, and in elucidating the roles of developmental genes in both mechanisms of development. By knocking down gene expression using RNA interference (RNAi), it is possible to determine the function of a gene. In the pea aphid, *Acyrthosiphon pisum,* RNAi has yet to achieve successful results in the study of gene function *during development*, although previous attempts in our lab using siRNAs have yielded promising results. By injecting double-stranded RNA (dsRNA) for the developmental genes *Distal-less (Dll)* and *Ultrabithorax (Ubx)* into the hemolymph of both sexual- and asexual-producing mothers, we hope to knock down expression of these genes in developing embryos. For comparison, we also injected milkweed bugs (*Oncopeltus fasciatus*) with dsRNA for the same genes, since this hemipteran insect is known to exhibit parental RNAi.

PHYSIOLOGICAL ESTROGENS EXERT OPPOSING EFFECTS ON HEART RATE IN ZEBRAFISH EMBRYOS

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Estrogens are a group of hormones that influence the development and function of diverse organ systems, such as the reproductive, nervous and cardiovascular systems. Acute exposure to 17β-estradiol (E2) increases heart rate in zebrafish embryos, an established model for cardiovascular development. However, the effect of acute exposure to other physiological estrogens on heart rate is unknown. Here we show that the physiological estrogens estrone (E1) and estriol (E3) have opposing effects on heart rate. 50-hour post fertilization zebrafish embryos treated with exogenous E1 had a decrease in heart rate while embryos treated with exogenous E3 had an increase in heart rate. Estrogens signal through two different receptors: nuclear estrogens receptors (ERα, ERβ) that are ligand-dependent transcription factors, or the G-protein coupled estrogens receptor (GPER), an integral membrane protein that activates second messengers in the cytosol. E2 acts via GPER to increase heart rate, but whether E1 or E3 modulate heart rate via GPER is not known. To test whether E1 and E3 act via GPER to change heart rate, we treated GPER mutant zebrafish with E1 and E3 for one hour and measured heart rate. We found that E1 led to a decrease in the heart rate while the E3 did not cause an increase in heart rate suggesting E1 acts in a GPER-independent mechanism while E3 acts in a GPER-dependent mechanism. Our results demonstrate that although estrone, 17β-estradiol and estriol are structurally similar, each has its own unique downstream physiological effect on heart rate.

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Morning Poster Session
Group I - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

51. **EFFECT OF THE 15KDA SELENOPROTEIN ON WNT/B-CATENIN EXPRESSION IN MICE**

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52. **EFFECTS OF CRF_1-RECEPTOR ANTAGONIST ON STRESS INDUCED ALCOHOL CONDITIONED PLACE PREFERENCE**

Eugene Dennis[1], Irvin Gamara[1], Norman Schanz[1], Zhicheng Carl Lin[2] and Emmanuel S. Onaivi[1]
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53. **microRNA MODULATION OF THE NON-CANONICAL WNT SIGNALING PATHWAY IS ESSENTIAL FOR CELLULAR MORPHOGENESIS**

Tyler McCann and Jia Song
Department of Biological Sciences, University of Delaware, 105 The Green, Newark, DE 19716

54. **PRADER-WILLI SYNDROME (PWS) - SNORD116: NON-CODING RNA**

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55. **INVESTIGATING THE MOLECULAR COMPONENTS OF COLD NOCICEPTION IN DROSOPHILA LARVAE**

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56. **MAP1B IMPACTS ON NEURULATION AND MICROTUBULE STABILITY**

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EFFECT OF THE 15KDA SELENOPROTEIN ON WNT/B-CATENIN EXPRESSION IN MICE

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Selenium, primarily through its incorporation into selenoproteins, affects many cellular mechanisms, particularly those involved in cellular homeostasis. Previous research suggests that the 15kDa selenoprotein (Sep15) may be involved in the regulation of colon carcinogenesis, as Sep15 deficiency was shown to protect against the formation of chemically-induced aberrant crypt foci in mice. We are currently examining the expression of typical colon cancer genes associated with the Wnt/β-catenin pathway in response to the expression (control) or absence of expression (knockout) of Sep15 in mice. These mice were not treated with any chemicals, and were maintained on a diet of adequate selenium (0.1 ppm selenite) for 20 weeks. Colon epithelia, serum and other tissues were harvested at the end of the study. mRNA was harvested from colon epithelia, reverse transcribed to cDNA and quantitated using real-time RT-PCR.

Preliminary analyses showed a significant mRNA upregulation of β-catenin variant 2, Cyclin B1 binding protein and Wnt5A in Sep15 knockout mice, whereas mRNA expression of APC, Wnt2B, and Axin appeared slightly decreased but not significantly changed. Several genes within the Wnt/β-catenin pathway were close to the limit of detection. Thus, our preliminary results suggest that Sep 15 may regulate some aspects of the Wnt/β-catenin pathway, which will be further investigated.

Supported by the NIH Office of Dietary Supplements, Towson University's Fisher College of Science and Mathematics, and Jess and Mildred Fisher Endowed Chair funds (P. Tsuji).

EFFECTS OF CRF_1-RECEPTOR ANTAGONIST ON STRESS INDUCED ALCOHOL CONDITIONED PLACE PREFERENCE

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The Hypothalamic-Pituitary-Adrenal (HPA) axis is a primitive pathway. It is one of the first if not the first pathways to develop in mammals. It mediates stress responses and therefore is very essential to the survival of the mammalian species. Hyper or hypo-activation of this axis has often been implicated in stress related disorders like PTSD, ADHD, Schizophrenia, Alzheimer's, autism depression, and alcoholism. Upon the perception of stress, corticotropin-releasing hormone (CRH or CRF in rodents) is released by the hypothalamus. This binds to its receptor causing a cascade that eventually leads to the release of cortisol and the fight or flight response. Two G-protein coupled receptors ($CRF\text{-}R_1$ and $CRF\text{-}R_2$) have been identified to respond to CRF in rodents. Of the two receptors, $CRF\text{-}R_1$ has a high affinity for CRF. Overactivity of the $CRF\text{-}R_1$ contributes to anxiety disorders and depression. This experiment used the conditioned place preference (CPP) paradigm to examine how a $CRF\text{-}R_1$ antagonist (CP-154) would influence stress induced alcohol CPP in mice. Subjects conditioned with alcohol and stress alone showed "conditioning" which is indicated by increase in the time spent in zone 1 of the CPP apparatus. Stress combined with alcohol induced conditioning whiles stress combined with CP-154 also induced conditioning. On the other hand, CP-154 alone induced aversion. Alcohol combined with CP-154, as well as stress and CP-154 together induced aversion of the subjects to zone 1. These results provides some insight into the HPA-axis and the role it plays in stress and could possibly explain how alcohol may play a role in relieving stress. This could also be used to explain how antidepressants which are used to treat stress related disorders like depression work and why they do not work in conjunction with alcohol.

microRNA MODULATION OF THE NON-CANONICAL WNT SIGNALING PATHWAY IS ESSENTIAL FOR CELLULAR MORPHOGENESIS

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The Wnt signaling pathways are highly evolutionarily conserved in regulating cell specification, cell polarity and morphogenesis in development. The non-canonical Wnt pathways (ncWnt) consist of the Wnt/Planar Cell Polarity (Wnt/PCP) and the Wnt/calcium pathways. *DSH* transduces Wnt ligand activation of all Wnt signaling pathways. Previous studies using genetic approaches and drugs indicate that the morphogenic movements of primary mesenchyme cells (PMCs), which give rise to the skeleton spicules that facilitate larval swimming and feeding, in the sea urchin embryo are in part regulated by the ncWnt pathway. To investigate specifically the contribution of ncWnt/calcium pathway in regulating skeletogenesis, we treated sea urchin embryos with inhibitor Cyclosporin A (CsA) and activator Phorbol 12-myristate 13-acetate (PMA) that disrupt the ncWnt/calcium pathway. Both drug treatments led to significantly shorter skeleton spicules compared to untreated embryos. While ncWnt has been established to regulate cellular movements, the role of microRNAs (miRNAs) in modulating the ncWnt pathways in controlling directed migration has not been examined. We hypothesize that miRNAs regulate the ncWnt by suppressing *DSH*. miRNAs are non-coding RNA molecules that mediate post-transcriptional regulation by binding to the 3'untranslated regions of target transcripts. Using luciferase constructs and site-directed mutagenesis, we identified *DSH* to be directly regulated by at least two miRNAs. To examine the *in vivo* impact of miRNA regulation on *DSH*, we treated newly fertilized sea urchin eggs with *DSH* miRNA Target Protector (miRNA TP) designed to specifically block miRNA binding to the *DSH* mRNA. Our results indicate that the removal of miRNA regulation on *DSH* led to morphogenetic defects in skeletogenic cell patterning and gut. This study reveals the regulatory role of miRNAs of the Wnt signaling pathway that contributes to a deeper understanding of underlying causes of developmental defects resulting from failed control of cell migration.

The University of Delaware Undergraduate Research Program and the Howard Hughes Medical Institute (HHMI) provided partial funding for this project.

PRADER-WILLI SYNDROME (PWS) - SNORD116: NON-CODING RNA

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Prader-Willi syndrome (PWS) is a neurodevelopmental disorder caused by the deletion or silencing of the paternal 15q11-13 loci. Within this region, loss of expression of the *SNORD116* gene cluster on the paternal allele is sufficient to cause PWS. *SNORD116* is a non-coding RNA, which is naturally maternally imprinted. When the *SNORD116* gene is spliced, the exons fuse to produce the *SNORD116* Host Gene (*116HG*), which forms an RNA cloud. This RNA cloud regulates the expression of several diurnal genes. Additionally, the introns are processed to become *SNORD116* small nucleolar RNAs (snoRNAs). However, the molecular function of snoRNAs and its relevance to PWS is still unknown.

The formation of RNA cloud from *116 HG* is a vital concept to creating potential therapies for PWS as well as understanding the pathology of other related disorders. The model mice used in this experiment were genetically modified paternal *SNORD*116 deleted Prad mice, Complete transgenic (Ctg) mice, Host Gene transgenic (HGtg) mice, and Sno-Mutant transgenic (SMtg) mice. We hypothesized that restoring RNA cloud formation can rescue some of the symptoms of PWS observed in our PWS mouse model. We worked on locating the transgene in thirteen Sno-Mutant Transgenic Prad mice by using a method called inverse Polymerase Chain Reaction (PCR). First, we used the restriction enzyme DpnII to cut the mice DNA and after ligation with T4 ligase, various primers were used to determine the precise position of the transgene in the genome of the mice samples. Although we are yet to verify the location of the transgene in the Sno-Mutant mice, further research should lead to discovering the factors that tether *116HG* to the DNA to form an RNA cloud.

INVESTIGATING THE MOLECULAR COMPONENTS OF COLD NOCICEPTION IN DROSOPHILA LARVAE

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Online access of this abstract is restricted at the request of the Principal Investigator.

MAP1B IMPACTS ON NEURULATION AND MICROTUBULE STABILITY

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Brain development is a stepwise process that begins with neurulation. It is the process by which the neural tube develops, the precursor to the brain and spinal cord. Disrupting the intricate process of neurulation can result in mild to severe neural birth defects. An important and conserved event in neurulation is neural convergence extension (NCE) of the neural ectoderm. During NCE, neuroepithelial cells elongate mediolaterally and migrate towards the midline, thus narrowing and lengthening the neural plate. An important cellular component that drives NCE are microtubules (MTs). MTs are dynamic cytoskeletal tracts that drive tissue morphogenesis. Insufficient regulation of microtubule dynamics during neurulation is associated with neural tube defects in model organisms. MTs shorten, lengthen, and stabilize at the instruction of intrinsic and extrinsic factors to shape the tissues using cellular processes that are poorly understood. One such process is regulation by Microtubule Associated Proteins (MAPs), which bind directly to microtubules. Most MAPs control dynamics and stability. Specifically, Map1b temporally modulates MT polymerization and axon elongation, crucial aspects of early nervous system development.

In the direction of better understanding Map1b and its impacts, Morpholino (MO) and Dominant Negative (DN) constructs were used to generate Map1b loss of function (LOF) phenotypes in zebrafish embryos. We found that depleting Map1b with MOs delayed NCE. Histological examination of the MO experiments showed that Map1b depletion caused destabilization in microtubule lattices and altered cell morphology during NCE. Our results are congruent with others that markedly implicate Map1b as a key microtubule regulator during neural tube morphogenesis.

After the alleged invalidation of MOs in recently published paper, we began using RNA Dominant Negatives (DN) to knock-out Map1b. Preliminary results from our dominant negative study are forthcoming, however, we anticipate agreement with the MO study.

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*These authors contributed equally to this work
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Morning Poster Session
Group J - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

57. **ANTIMICROBIAL PROPERTIES OF FLUOROQUINOLONE ANTIBIOTICS ON RESISTANT AND NON-RESISTANT BACTERIA IN BALTIMORE WASTEWATER AND SURFACE WATER**

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58. **ANALYSIS OF MODIFIED HISTONE DISTRIBUTION AT THE TISSUE-SPECIFIC IMPRINTED RASGRF1 LOCUS USING ALLELE-SPECIFIC QPCR FOLLOWING CHIP**

Nicole Hamagami and Tamara L. Davis
Department of Biology, Bryn Mawr College, 101 North Merion Avenue, Bryn Mawr, PA 19010

59. **NOVEL MARKERS OF LENS FIBROSIS**

Priyha Mahesh, Yichen Wang, and Melinda K. Duncan
Department of Biological Sciences, University of Delaware,
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60. **VIOLACEIN MEDIATED FOOD PROTECTION FROM PSYCHROPHILE *Janthinobacterium lividum* OVC5, ISOLATED FROM COLD SOIL**

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Department of Biological Sciences, University of Pittsburgh at Bradford,
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61. **DISRUPTION OF CYCLIC DI-GMP SIGNALING BY SMALL MOLECULES TO INHIBIT ALGINATE PRODUCTION BY *PSEUDOMONAS AERUGINOSA***

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ANTIMICROBIAL PROPERTIES OF FLUOROQUINOLONE ANTIBIOTICS ON RESISTANT AND NON-RESISTANT BACTERIA IN BALTIMORE WASTEWATER AND SURFACE WATER

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Fluoroquinolone (FQ) antibiotics are one of the most popular classes of antibiotics and are intensively used in both humans and animals. After consumption, a fraction of the antibiotics is excreted unchanged. As a result, FQ antibiotics have been consistently found in municipal wastewater. Trace concentrations of these antibiotics in wastewater may contribute to the development and spread of antimicrobial resistance. Compared to non-resistant strains, antibiotic-resistant bacteria must be exposed to greater antibiotic concentrations to inhibit growth. The objective of this work was to simultaneously monitor antibiotic concentrations and antibiotic resistant bacteria (ARB) in wastewater and wastewater-impacted surface water. The antimicrobial activity of fluoroquinolone antibiotics against control bacteria and ARB was determined. We hypothesized that fluoroquinolone concentrations can be correlated with the presence of ARB in water samples. To test this hypothesis, water samples were collected from a Maryland-based wastewater treatment plant, as well as surface water from upstream and downstream of the wastewater effluent discharge site. Water samples were analyzed for 15 fluoroquinolone antibiotics using online solid-phase extraction liquid chromatography tandem mass spectrometry. The total fluoroquinolone mass concentration ranged from 27 ng/L in upstream surface water samples to 2090 ng/L in raw wastewater samples. Fluoroquinolone-resistant bacteria were isolated on agar plates containing ciprofloxacin, and multidrug-resistant bacteria were selected using extended spectrum β-lactamase (ESBL) plates. Downstream surface water contained higher FQ and ARB concentrations, confirming concerns over the discharge of antibiotic resistance from wastewater treatment plants. Inhibition profiles were generated from isolates to demonstrate the relative potency of different fluoroquinolone antibiotics. The fluoroquinolone antibiotics were less potent against the downstream surface water isolates compared to the upstream, suggesting that the bacteria become more resistant after wastewater treatment.

This work was funded, in part, through the UNCF/Merck Undergraduate Research Fellowship, NIH/NIBIB, the Meyerhoff Scholars Program, and an Undergraduate Research Award from the UMBC Office of Undergraduate Education.

ANALYSIS OF MODIFIED HISTONE DISTRIBUTION AT THE TISSUE-SPECIFIC IMPRINTED RASGRF1 LOCUS USING ALLELE-SPECIFIC QPCR FOLLOWING CHIP

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Genomic imprinting is an epigenetic mechanism that facilitates the expression of one parental allele over another by attracting or repelling various elements required for DNA transcription. Specifically, posttranslational histone modifications structurally alter the chromatin itself or regulate enzymatic chromatin binding factors that play a role in genetic expression and imprinting.

The imprinted mouse gene *Rasgrf1* is paternally expressed in a tissue-specific manner. Previous studies on the *Rasgrf1* gene have shown that DNA methylation is present on the paternal allele in tissues with both imprinted and non-imprinted expression, indicating that DNA methylation is not solely responsible for the tissue-specific expression patterns observed at this gene locus.

Histone modifications are additional epigenetic factors that may explain the tissue-specific imprinted expression seen at *Rasgrf1*. To determine whether histone modifications explain these tissue-specific expression patterns, we coupled chromatin immunoprecipitation (ChIP) with allele-specific amplification of DNA using quantitative PCR to locate modified histones that are preferentially distributed on either or both parental alleles. Since the chromatin structure at the promoter tends to reflect the expression patterns of the gene itself, we expect to see permissive and repressive histone modifications on the promoter region of the paternal and maternal chromosomes, respectively, in mono-allelic tissues, and permissive modifications on both alleles for bi-allelic tissues. However, we are unsure how these modified histones will be distributed at the DMR site. If the modified histone distribution appears to reflect that of DNA methylation at these regions, then we may conclude that histone modifications are not involved in the differential expression seen in mono-allelic and bi-allelic tissues. Differences in distribution in mono-allelic versus bi-allelic tissues, however, may suggest the involvement of these modified histones in tissue-specific gene expression. Therefore, the goal for our research is to determine whether histone modifications directly correlate with DNA methylation in regulating differential gene expression.

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NOVEL MARKERS OF LENS FIBROSIS

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Fibrosis is a pathological condition affecting multiple organs throughout the body. Fibrosis is characterized by the formation of scar tissue through hyperproliferation of cells, differentiation of cells into myofibroblasts, and excess production and contraction of extracellular matrix. In the ocular lens, composed of epithelial cells and fiber cells, fibrosis is thought to occur through the epithelial to mesenchymal transition (EMT) of these epithelial cells to migratory fibroblasts. EMT occurs when epithelial cells lose their orientation and change their phenotype. While studies suggest that EMT is induced by the activity of the cytokine TGF-β, there are limited molecular markers for these cells undergoing transdifferentiation aside from α smooth muscle actin (αSMA). Previous research suggests that CD44, a receptor for hyaluronan, acts as an early EMT marker and is a fiber cell marker. When CD44 was removed from the lens, another class of hyaluronan receptors known as Receptor for Hyaluronan Mediated Motility (RHAMM) upregulated. This led us to hypothesize that RHAMM could potentially act as a marker for lens fibrosis. Through immunofluorescence staining of mouse lenses, we were able to observe that in the early developing embryo RHAMM is distributed in all lens cells. In older embryonic time points, RHAMM appeared to restrict itself to lens epithelium cells and this pattern of expression was maintained in newborn and adult lens. The next step will be to investigate how the distribution and levels of RHAMM change in lens cells in a mouse model of cataract surgery. Other potential markers of fibrosis will also be explored.

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VIOLACEIN MEDIATED FOOD PROTECTION FROM PSYCHROPHILE *Janthinobacterium lividum* OVC5, ISOLATED FROM COLD SOIL

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Online access of this abstract is restricted at the request of the Principal Investigator.

DISRUPTION OF CYCLIC DI-GMP SIGNALING BY SMALL MOLECULES TO INHIBIT ALGINATE PRODUCTION BY *PSEUDOMONAS AERUGINOSA*

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Pseudomonas aeruginosa is an opportunistic pathogen that affects over 90 percent of individuals with the genetic disorder cystic fibrosis (CF). Individuals with CF carry a mutation in the cystic fibrosis transmembrane conductance regulator (CFTR) gene, which causes an inability to maintain hydration within the epithelial lining of the respiratory tract and leads to the accumulation of dehydrated mucus within the airways. Due to the presence of dehydrated mucus, cilia within the lungs are unable to expel inhaled microbes from the airways, facilitating the growth of pathogens, including *P. aeruginosa*. As a result, many CF patients experience chronic lung infection from a single strain of *P. aeruginosa* that persists for the entirety of their lives. In a phenomenon known as mucoid conversion, these infectious strains accumulate genetic mutations that deactivate the *mucA* gene, causing the bacteria to secrete copious amounts of a virulence factor known as alginate, which causes airway blockage, lung inflammation, and increased morbidity and mortality. Alginate biosynthesis by *P. aeruginosa* is regulated by the bacterial second messenger cyclic di-GMP (c-di-GMP), which binds to the receptor protein Alg44 in order to elicit alginate production. The identification of small molecules that disrupt c-di-GMP signaling by preventing the second messenger from binding to Alg44 could subsequently reduce the ability of *P. aeruginosa* to produce alginate. Here, we demonstrate the identification of a class of small molecules derived from benzo-triazolo-quinazolinone that inhibit the binding of Alg44 to c-di-GMP, and we show their *in vivo* efficacy in reducing alginate secretion by *P. aeruginosa*. The discovery of a class of inhibitors of Alg44 binding to c-di-GMP represents a novel area of study that could result in the development of pharmaceuticals that reduce alginate production by *P. aeruginosa*, allowing CF patients to live healthier lives.

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Morning Poster Session
Group K - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

62. **CHARACTERIZATION OF EPIGENETIC PATTERNS IN THE HUMAN RETINA**

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63. **GOLDENSEAL (*HYDRASTIS CANADENSIS* L.) EXTRACT DOSE-DEPENDENT GROWTH INHIBITION OF *H. PYLORI* BACTERIA AND EFFECTS OF *H. PYLORI* ON GASTRIC CELL FREE AMINO ACID CONCENTRATIONS**

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64. **DNA METHYLATION PATTERNS AT THE IG-DMR**

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65. **THE DEVELOPMENT OF A MMP17 FLUORESCENT BIOSENSOR**

Donna McKeon, Trevor Cross, Tam-Linh Nguyen, Matthew J. Farber, and Peter B. Berget
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66. **THE OVEREXPRESSION OF CHLOROPLAST CARBONIC ANHYDRASES CAH3 AND CAH6 IN CHLAMYDOMONAS REINHARDTII TO IMPROVE GROWTH FOR BIOFUEL PRODUCTION**

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CHARACTERIZATION OF EPIGENETIC PATTERNS IN THE HUMAN RETINA

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The vertebrate retina is a stratified layer of neuronal cells that serve a highly specialized function: converting photons of light absorbed by photosensitive cells to an electrochemical signal which are then perceived as vision in the brain. The development and function of retinal cells is dictated by epigenetic regulation of retina-specific genes. One particular epigenetic modification, DNA methylation, is essential for the governance of gene expression by modulating the recruitment of activating and repressive proteins in gene regulatory regions. Our experiments characterize DNA methylation levels near the promoter regions and transcriptional start sites (TSS) of retina-specific genes in post-mortem human retina and cultured human embryonic stem cells (hESC). Previous studies have demonstrated mRNA and protein expression in the human retina and hESC organoids, but retina-specific epigenetic regulation of gene expression has yet to be fully characterized in these systems. We hypothesize that the mRNA expression of retina-specific genes is inversely correlated with cell-specific patterns of DNA methylation in regulatory regions and around the TSS. Preliminary data demonstrates lowered levels of DNA methylation in promoter regions of the retina-specific genes, *PDE6b*, *OTX2, RCVRN*, *RHO*, and *CRX,* in post-mortem human retina compared to non-retinal tissues using bisulfite pyrosequencing. Our future studies will analyze DNA methylation as well as mRNA transcription of additional retina-specific loci in both post-mortem humans and hESC organoids.

This project is supported by funding from the Commonwealth Health Research Board, and JMU 4-VA. We also thank the Nasonkin Lab at University of Pittsburgh Medical Center.

GOLDENSEAL (*HYDRASTIS CANADENSIS* L.) EXTRACT DOSE-DEPENDENT GROWTH INHIBITION OF *H. PYLORI* BACTERIA AND EFFECTS OF *H. PYLORI* ON GASTRIC CELL FREE AMINO ACID CONCENTRATIONS

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Gastric cancer is the third deadliest form of malignancy, responsible for 723,000 deaths worldwide in 2012. The greatest identifiable risk factor for gastric cancer development is infection with class I carcinogen *Helicobacter pylori* (*H. pylori*) which increases the risk of disease 6 to 8 times. Due to *H. pylori*'s estimated presence in 2/3^{rds} of the human population, prevalence in under-developed regions, and elevated risk of associated gastrointestinal complications, natural supplements are being investigated for their antibacterial efficacy.

Goldenseal extract (*Hydrastis canadensis* L.) has demonstrated potent *H. pylori* growth inhibition effects; however, standardization of plant-based supplements is challenging. In order to further evaluate Goldenseal extract's antibacterial properties, we endeavor to determine Goldenseal extract's effects on *H. pylori* growth and gastric cancer cells' free amino acid metabolism, as a means of standardization and to better understand Goldenseal's mechanism of action. First, 7.13WT *H. pylori* was cultured with varying Goldenseal extract concentrations to monitor dose-dependent growth inhibition effects. Second, the non-flagellated 7.13 *flaA* knockout strain was also employed to assess the flagella's significance to bacterial survival and detect Goldenseal extract's potential influences on the *flaA* gene pathway. Bacteria from the minimum inhibitory Goldenseal extract concentration were co-cultured with AGS human gastric cancer cells. Amino acid analysis of growth media at various time points was conducted for 90hr. We hypothesize that decreased concentrations of non-essential amino acids utilized by *H. pylori* for survival, will be observed in the presence of Goldenseal. These changes in free amino acid concentrations may also alter cellular downstream pathways and provide further insight regarding *H. pylori*'s mechanism of pathogenicity and carcinogenic potential. Overall, this research will contribute to *in vitro* standardization of Goldenseal products and provide evidence of Goldenseal's efficacy regarding the prevention of *H. pylori* infection related complications including gastrointestinal cancer and ulcers.

Support for this work was funded by NIH 2R44AT003365 (HH) and 5R25HL108825-05 (UAMS-SURP).

DNA METHYLATION PATTERNS AT THE IG-DMR

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Genomic imprinting is a form of epigenetic regulation of gene expression in mammals. For most genes, both the maternal and paternal alleles are expressed. However, imprinted genes have one allele marked early in development, depending on their parental origin, through either DNA methylation or histone modification. These markings, which are differentially distributed, cause one allele to be expressed. So far approximately 150 imprinted genes have been discovered in humans and mice. The imprinting of these genes is necessary for proper growth and development, and multiple disorders are known to develop when imprinted genes are improperly expressed. Research into the mechanisms and patterns of DNA methylation is necessary to better understand and develop treatments for these human disorders.

DNA methylation, an epigenetic mark regulating genomic imprinting, involves adding a methyl group to the cytosine at CpG dinucleotides on both complementary strands. 96% of mammalian DNA is symmetrically methylated, however previous research showed an unusually high percentage of asymmetrical methylation at the *Dlk1* differentially methylated region (*Dlk1*-DMR) in mice. DNA methylation of the Dlk1-DMR is acquired during embryonic development, and the low level of symmetrical methylation (65%), correlates with an overall more variable DNA methylation pattern. Our current study is analyzing a more stably methylated region, intergenic DMR (IG-DMR), which is methylated during spermatogenesis. We hypothesize that this region will be mostly symmetrically methylated because regions that are methylated earlier in development are correlated with more stable methylation patterns. By connecting complimentary strands via hairpin linker and replacing the unmethylated cytosines with thymines, we are able to distinguish the symmetry of the methylated groups. We found that the IG-DMR has very little asymmetrical methylation over multiple developmental stages and crosses, and the least amount of asymmetrical methylation of the three DMRs analyzed at the Dlk1-Gtl2 cluster. Therefore, our findings support our hypothesis.

I would like to thank Dr. Tamara Davis, the Summer Science Research Program at Bryn Mawr, the National Science Foundation grant 1157819 awarded to Tamara L. Davis, and my fellow lab members for their support and guidance in this project.

THE DEVELOPMENT OF A MMP17 FLUORESCENT BIOSENSOR

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Matrix Metalloproteinases (MMPs) influence cell signaling behaviors by cleaving substrates such as gelatin in the extracellular matrix (ECM), allowing cell-cell migration to take place. Abnormal activity of MMPs is associated ailments such as rheumatoid arthritis (RA) by cleaving proteins that are important for cartilage structure. Understanding MMP activity will contribute to new treatments and cures for the disorders that are caused by their dysfunction by enabling the creation of selective inhibitors.

Single chain variable proteins (ScFvs) can be manipulated to understand the activity of MMP17, a protease found to be associated with RA. ScFvs are recombinant proteins composed of the variable light chain and variable heavy chain of an antibody, connected with a flexible linker. ScFvs often retains the binding capacity of the full antibody. The ScFv used in this study is a heavy chain-only domain which is capable of binding and fluorogenically activating a dye molecule. By attaching an inhibitory domain to this ScFv, we can control the dye binding ability of the heavy chain. In this manner, we can encode a protease cleavage sequence in the flexible linker to create a protease biosensor. Upon cleavage of the linker, the inhibitory domain will disassociate and allow the activating domain to bind to dye and fluoresce. Using this model system, it is possible to build a biosensor that will be selectively activated by the activity of MMP17, and not the related MMP25, by introducing a protease-sensitive polypeptide sequence into the linker. Full-length biosensors with three hypothesized selective sequences, VAPLIL, QVFRLI, and RASRLV, were tested for selectivity for MMP17 by being treated with MMP17 or MMP25.

The results of this study revealed that the VAPLIL linker showed the highest selectivity for MMP17. Discovery of this selective substrate will enable more selective assays for MMP17 in the future.

I would like to thank Trevor Cross, Tam-Linh Nguyen, Dr. Matthew Farber, and Dr. Peter Berget for their mentorship throughout this project. Research was funded by NIH grant U54- RR022241 and the USciences Summer Undergraduate Reseach Fund (USURF), a grant made possible through the donations of Marvin Samson, CEO of Medical Technologies and Chairman of USciences Board of Trustees.

THE OVEREXPRESSION OF CHLOROPLAST CARBONIC ANHYDRASES CAH3 AND CAH6 IN CHLAMYDOMONAS REINHARDTII TO IMPROVE GROWTH FOR BIOFUEL PRODUCTION

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Microalgae grown under the right conditions can produce significant biomass, which can be converted to biofuel and provide an alternative energy solution as a renewable fuel. Many efforts are underway to discover how algae can be grown more efficiently to reduce the cost of algal biofuels. The objective of this project is to learn how to improve the growth of the model green alga Chlamydomonas *reinhardtii*. This species was chosen because its complete genome sequence is known and it is easy to manipulate, and what is learned about its biology should be applicable to industrial production algal species. *C. reinhardtii* and other algae rely on CO_2 concentrating mechanisms (CCMs) to elevate the concentration of CO_2 near the enzyme rubisco. A key component of the CCM are carbonic anhydrases (CAs), which catalyze the interconversion of CO_2 and HCO_3^-. In this project, we are attempting to overexpress two chloroplast carbonic anhydrases genes, *CAH3* and *CAH6*, which are believed to play a key role in the CCM. CAH6 is found in the chloroplast stroma while CAH3 is in the thylakoid lumen in close proximity to rubisco, the enzyme responsible for CO_2 fixation. *CAH3* and *CAH6* coding regions were gene synthesized then ligated into a nuclear expression vector. The *CAH6* construct was transformed into two different C. *reinhardtii* strains, CC48 and CC4350. Several lines of transformed cells were cultured and extracts were prepared for western blot analysis to determine which accumulate high levels of protein. We will perform growth curve analysis of the highest CAH6 producers to determine the effect of CAH6 overexpression on growth rate and biomass production.

These results were obtained as part of the Research Experience and Mentoring (REM) program in the Department of Biological Sciences at the UMBC. This program is funded by a grant (REM supplement to NSF-EFRI-1332344) from the National Science Foundation (NSF) Directorate for Engineering (ENG) Office of Emerging Frontiers in Research and Innovation (EFRI).

Morning Poster Session
Group L - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

67. **GENERATION OF LIN28A/B HUMAN EMBRYONIC STEM CELL REPORTER CELL LINE USING CRISPR TECHNIQUE**

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68. **EFFECT OF HYDROGEL MODULUS ON MACROPHAGE PHENOTYPE**

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69. **INVESTIGATION INTO THE ROLE OF RALDH2 IN INFLAMMATION AND REMYELINATION IN THE CNS**

Alisha N. Dua, Sonia Nanescu, and Jeffrey K. Huang
Department of Biology, Georgetown University, 37th and O Street NW, Washington, DC 20057

70. **EFFECTS OF DIFFERENT DOSES OF EGCG ON OOGENESIS IN WILD-TYPE AND EGFR MUTANT DROSOPHILA**

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71. THE 15KDA SELENOPROTEIN IN THE REGULATION OF COLON CANCER SIGNALING PATHWAYS IN A MOUSE MODEL OF SELENIUM DEFICIENCY

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72. **CROSS TALK BETWEEN CELLULAR ORGANLLES DURING TAIL REGRESSION IN TADPOLES**

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GENERATION OF LIN28A/B HUMAN EMBRYONIC STEM CELL REPORTER CELL LINE USING CRISPR TECHNIQUE

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Lin28 is an RNA binding protein first discovered in C. elegans. Mammals have two Lin28 homologs: Lin28 A and B. Our lab and others have demonstrated roles of the Lin28 paralogs in processes ranging from tumorgenesis to reprogramming and pluripotency. Studies have also shown that loss of function in Lin28 accelerates stem cell differentiation while gain of function in Lin28 promotes self-renewal and delays differentiation in stem cells.

The goal of this project is to generate a Lin28A/B human embryonic stem cell reporter cell line utilizing CRISPR. This will allow us to study the role of Lin28 in human stem cell biology. We have successfully cloned Lin28 from genomic DNA via PCR. We have also made an H1 hESC cell line inducible for cas9, as well as cloned the guides that will direct cas9 to the Lin28A/B locus. We have constructed the donor plasmid for Lin28B using the fluorescent protein tdtomato and are working on constructing the donor plasmid for Lin28A using eGFP. We have also been working on nucleofecting hESCs with fluorescent proteins and plan to optimize our guide RNAs. With this Lin28 reporter cell line, we will be able to track expression of the proteins and study its role in maintenance of pluripotency and differentiation potential in human embryonic stem cells.

EFFECT OF HYDROGEL MODULUS ON MACROPHAGE PHENOTYPE

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Online access of this abstract is restricted at the request of the Principal Investigator.

INVESTIGATION INTO THE ROLE OF RALDH2 IN INFLAMMATION AND REMYELINATION IN THE CNS

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Multiple sclerosis (MS) is a demyelinating, progressive neurodegenerative disease characterized by chronic inflammation and inefficient remyelination. The vitamin A metabolite, retinoic acid (RA), has been previously implicated in improving remyelination efficiency in elderly mice. While the RA synthesizing enzyme, Raldh2, is known to have significant differential expression over the course of remyelination after injury, it is unclear which cell types synthesize and utilize RA in the lesioned environment. Preliminary data from immunochemistry of mixed glial cell cultures and lysolecithin lesioned spinal cord tissue indicates that oligodendrocyte progenitor cells (OPCs) and microglia are the primary producers of RA.

To test the role of RA signaling in microglia, purified microglia cultures were treated with LPS to induce M1-like polarization. Exogenous addition of RA to these polarized cells resulted in decreased expression of the nitric oxide synthesizing enzyme, iNOS, confirming the anti-inflammatory effect of RA described in previous research. Additionally, RA receptor activation was significantly decreased in LPS-treated, M1-like polarized microglia cultures as compared to resting microglia. This data suggests that RA signaling in microglia maybe dependent on polarization state and that RA may be a regulator of inflammation in lesioned tissue. Further analysis of Raldh2 expression and RA signaling in M1-like versus M2-like microglia is a necessary next step in understanding the function of RA in inflammation. This work will provide insight into the role of RA in MS pathology and the efficacy of RA as a potential therapy for MS.

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EFFECTS OF DIFFERENT DOSES OF EGCG ON OOGENESIS IN WILD-TYPE AND EGFR MUTANT DROSOPHILA

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Increasing the dosage of EGCG given to *Drosophila melanogaster* appeared to have positive effects on oogenesis in EGFR mutant fruit flies, but not in the wild-type. EGCG (a green tea extract) was fed to wild-type and EGFR mutant flies at concentrations of 0 mM, 2 mM, and 4 mM. According to the hypothesis, a higher concentration of EGCG would have resulted in fewer eggs laid and a larger number of the remaining eggs having DA (dorsal appendage) defects, and these effects would have been even more pronounced in the EGFR mutants than the wild-type, due to the drug interfering with the EGFR pathway. This was not quite what was observed, however. In the wild-type, the number of eggs laid stayed the same and then decreased, but in the EGFR mutants, the number decreased slightly and then increased dramatically. DA defects in the wild-type eggs showed a slow but steady increase. DA defects in the EGFR eggs decreased and then increased back to their original level and showed no continuous linear trend in either direction. These results suggest that EGCG does not inhibit the EGFR pathway further as the dosage increases, as was predicted. Since a number of cancerous tumors have EGFR mutations, knowing how EGCG affects that pathway can have a significant impact on cancer treatment. Further experimentation is required to demonstrate exactly how EGCG affects the EGFR pathway.

Thank you to Fox Chase Cancer Center for providing the necessary materials used to complete this project.

THE 15KDA SELENOPROTEIN IN THE REGULATION OF COLON CANCER SIGNALING PATHWAYS IN A MOUSE MODEL OF SELENIUM DEFICIENCY

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Through the function of selenoproteins, dietary selenium can affect many cellular mechanisms, including those involved in cellular homeostasis and cancer. Previous research suggests that the dietary selenium-sensitive 15kDa selenoprotein (Sep15) may be involved in the regulation of colon carcinogenesis, as Sep15 deficiency was demonstrated to protect against the formation of chemically-induced aberrant crypt foci in a Sep15 knockout mouse model. The protection was greatest under conditions of adequate dietary selenium. However, the regulatory mechanism behind Sep15 and its role in selenium deficiency still remain unclear.

The Wnt cell signaling pathway regulates cellular homeostasis and cell proliferation, and its dysregulation has been linked to sporadic colon cancer. Presently, I am examining the mRNA expression of Wnt pathway genes, their downstream gene targets, as well as genes involved in the regulation of inflammation from colonic epithelia of mice with (control) and without Sep15 (knockout) to elucidate possible effects of Sep15 in the regulation of colon cancer. All mice were maintained on a selenium deficient (0.04 ppm selenite) diet over the span of 20 weeks, which reduces, but not eliminates, the expression of Sep15 compared to a selenium-adequate diet. Preliminary results showed a significant upregulation of Wnt2B in mice unable to express Sep15 compared to litter mate controls. Interestingly, mRNA expression of interferon-γ-regulated genes were also upregulated in colonic epithelia of Sep15 knockout mice. The effect of Sep15 in colon cancer will be further investigated, including the possible interaction with other selenoproteins that are sensitive to dietary selenium levels.

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CROSS TALK BETWEEN CELLULAR ORGANLLES DURING TAIL REGRESSION IN TADPOLES

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During metamorphosis, the anuran body changes dramatically to adapt from the aquatic to terrestrial habitat. Larval specific organ/tissue such as the tail completely degenerates by several different mechanisms including those triggered by reactive oxygen species (ROS). Presently we have carried out *in situ* staining for ROS and mitochondria, peroxisomes, calcium as well as mitochondrial permeability transition pore (MPTP) assay during different stages of metamorphosis in tadpoles, *Xenopus laevis*. During early stages of metamorphosis, there is moderate production of ROS in tail; but prior to and during tail regression, a significant increase in ROS was noted. However, there was no double immunolocalization for ROS and mitochondria indicating that mitochondria are not the source of ROS production.

Tail epidermis shows significant increase in peroxisomal density as metamorphosis progressed. Progressive condensation of nuclei from the tip of the tail towards the body also corresponded with a reverse gradient for peroxisome localization. Ventral fin showed signs of cell death before the dorsal fin as wedges of cell death overlapped with ROS localization and peroxisomal staining. We conclude that ROS responsible for cell death in regressing tail, is partly derived from peroxisomes and they seem to be ubiquitous organelles playing a key role in both the production and scavenging of ROS during. Our results on *in situ* staining for calcium as well as MPTP assay showed an increased expression of these parameters as metamorphosis progressed. Ca^{2+} signaling has long been known to be critically involved in both the initiation and effectuation of cell death. Oxidative stress accompanied by calcium overload leads to permeability pores of mitochondria to open as tail regression begins. Mitochondrial dysfunction is probably a consequence of calcium overload. Interrelationship between cellular organelles and mechanisms involved in tail regression are discussed.

Morning Poster Session
Group M - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

73. **MOLECULAR CHARACTERIZATION OF TWO HUMAN LENS EPITHELIAL CELL LINES AND THEIR SUITABILITY TO STUDY FUNCTION OF CATARACT GENES**

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74. **ANTIOXIDANT EFFECTS OF NORDIHYDROGUAIARETIC ACID ON OXIDATIVE STRESS IN *CAENORHABDITIS ELEGANS***

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75. **TOWARDS THE RATIONAL DESIGN OF POTENT PEPTIDE ANTIBIOTICS**

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76. **ROLE OF ECDYSONE IN THE MIGRATION OF BORDER CELLS IN *DROSOPHILA MELANOGASTER* EGG CHAMBERS**

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77. **ESTABLISHING OPTIMAL TIMING FOR INTRAVITREAL DELIVERY OF AN AAV2-CRE VIRAL VECTOR TO MURINE RETINAL GANGLION CELLS**

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MOLECULAR CHARACTERIZATION OF TWO HUMAN LENS EPITHELIAL CELL LINES AND THEIR SUITABILITY TO STUDY FUNCTION OF CATARACT GENES

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The eye lens is a transparent tissue that focuses light onto the retina, allowing high-resolution vision. The loss of lens transparency causes an eye disease called cataract, which is the leading cause of blindness worldwide. Half of the U.S. population above age 80 is affected by cataracts, for which surgical treatment is the principal therapeutic intervention. Cataracts are caused by environmental insults such as ultraviolet (UV) radiation as well as susceptibility due to alterations in the genome. Thus, characterization of factors that affect lens transparency is the first critical step toward development of new therapies. Recent discoveries have revealed a surprising link between proteins that are components of cytoplasmic RNA granules (RGs) and mammalian cataract. RGs like Processing Bodies (PBs) and Stress Granules (SGs) represent specialized cytoplasmic sites for transcript regulation. To investigate RGs in a lens cell culture model, we molecularly characterized two human lens epithelial cell lines (LECs), SRA01/04 and HLE-B3. Analyzing genome-specific STR by PCR, we authenticated that the LEC lines are human-derived. We next analyzed gene expression in these LECs by microarray analysis. Our data demonstrates that both LECs retain expression of genes that are enriched in native lens epithelial cells. Further, we find that both LECs support formation of RGs, and exhibits formation of SGs under UV-stress conditions. Thus, these studies present the first cellular models for investigating the molecular biology of UV-radiation exposure in lens cells, and therefore represent a new resource for study of factors that are linked to age-associated cataract in humans.

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ANTIOXIDANT EFFECTS OF NORDIHYDROGUAIARETIC ACID ON OXIDATIVE STRESS IN *CAENORHABDITIS ELEGANS*

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Reactive oxygen species (ROS) that accumulate during metabolism may act as signaling molecules, or when in excess, may cause oxidative stress. ROS react with and can damage important biomolecules, leading to dysfunction and disease. Antioxidants react with ROS so that cellular components are not damaged. Nordihydroguaiaretic acid (NDGA), a polyphenolic lignan found in the long-lived desert plant *Larrea tridentata*, inhibits lipoxygenases and may reduce ROS. NDGA has been shown to have potential medicinal benefits for the cardiovascular, immune, and neurological systems, including anti-tumorigenesis. Levels of oxidative stress in the presence or absence of NDGA can be quantified in the model organism *Caenorhabditis elegans* using a microplate reader to measure green fluorescent protein (GFP) expression. *C. elegans* strains expressing GFP driven by the oxidative stress-responsive promoters of heat shock protein (hsp-16.2), superoxide dismutase (sod-3), and glutathione-S-transferase (gst-4) were exposed to heat and chemical oxidants to induce oxidative stress. Fluorescence was quantified using a suppressor of activated let-60 Ras (sur-5) promoter-GFP fusion as a negative control, since this strain showed no increased in GFP accumulation after heat shock. Preliminary data demonstrate the expected, strong increase in GFP expression for the hsp-16.2- and sod-3-regulated strains between ten and sixteen hours after exposure to a forty five minute heat shock at 37ºC, while the gst-4p::GFP strain showed a more moderate response. Ongoing experiments include NDGA treatment of *C. elegans* to determine if it alters oxidative stress responses. Strains treated with NDGA expressed GFP at levels comparable to DMSO controls, suggesting that NDGA may not be effective at lowering oxidative stress levels under the conditions tested. Future experiments include using age-synchronized worm populations and dual fluorescence reporter strains to correct for worm numbers analyzed.

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TOWARDS THE RATIONAL DESIGN OF POTENT PEPTIDE ANTIBIOTICS

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Antimicrobial peptides (AMPs) are seen as possible molecules to combat drug-resistant bacteria because they target bacterial membranes. However, AMPs have not been successful because there is limited information on their interactions with membranes and their mechanism of disruption. Moreover, many of the discovered AMPs are toxic to mammalian cells, rendering them useless for medicinal applications. The purpose of this project was to utilize computer simulations and experiments to understand the function, rationally design, and expedite the discovery of optimized AMPs.

Microsecond atomic-scale molecular dynamics (MD) simulations were used in order to model how the peptides interact with membranes. The system, containing the peptides and the lipid bilayer, was solvated with TIP3P water as well as sodium, chloride, and potassium ions. The MD simulations showed that the peptide channels were able to conduct chloride ions across the membrane at 733 events per microsecond. In addition, the water conduction of the channels was 26 times more powerful than aquaporin, suggesting a mechanism for killing bacterial cells. The MD simulations were then analyzed to determine critical regions of the channels, such as hydrophilic and hydrophobic components. A 2916-member combinatorial peptide library was rationally designed based on the observations and included mutations in certain amino acids of the sequence. Solid-phase peptide synthesis was completed in order to create more potent antimicrobial peptides, and MALDI mass spectrometry and HPLC confirmed the high purity of the synthesis. Further research is being done to test the peptides on models of bacterial and mammalian membranes to identify optimized peptide sequences from the library. This study provides insight into how AMPs function and utilizes a new, simulation-based approach towards designing potent peptides, which saves time and money in the drug development process. The results gleaned from this research pave the way for the next-generation of treatments against drug-resistant bacteria.

Thank you to Martin, Charles, Sarah, Yukun, and Kalina for providing me the opportunity to work on this project in their labs.

ROLE OF ECDYSONE IN THE MIGRATION OF BORDER CELLS IN *DROSOPHILA MELANOGASTER* EGG CHAMBERS

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Cell migration is an essential mechanism during animal development. Understanding how cells migrate can help build information that future generations can use in the fight against birth defects and diseases like cancer. As in humans, steroid hormones in *Drosophila melanogaster* (fruit flies) control the timing of key developmental events, including cell movements, so it is important to investigate how these hormones signal. The goal of this project is to study the role of the steroid hormone Ecdysone in the border cells of fruit fly egg chambers. Border cells are a cluster of motile cells that must migrate during egg development to fulfill their functions. Steroid hormone signaling controls the timing of when border cells exit from the epithelium at the anterior end of the egg chamber and move to the oocyte. Work from multiple labs has identified several factors that are regulated by steroid hormone signaling, such as the role of the protein Abrupt in signaling, and cell adhesion regulators. We have identified many other potential downstream targets of ecdysone signaling through expression analysis. Through genetic experiments, we are determining which of these targets most significantly contribute to cell migration. Prior work has suggested that one of the potential target genes found, *PH4αEFB* (Prolyl-4-Hydroxlase alpha, Embryonic Fat Body) may play a role in border cell migration. Mutations that cause an abnormal phenotype are being further characterized. These results will inform us about the important signaling effectors downstream of ecdysone steroid hormone in cell migration. Gaining knowledge about these effectors will help with understanding how similar pathways function.

This investigation was supported in part by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences.

ESTABLISHING OPTIMAL TIMING FOR INTRAVITREAL DELIVERY OF AN AAV2-CRE VIRAL VECTOR TO MURINE RETINAL GANGLION CELLS

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The vertebrate retina is a light-sensitive, stratified layer of neuronal cells that lines the interior of the eye. Photons of light absorbed by photosensitive retinal neurons are converted into an electrochemical signal ultimately passed to the brain to process visual images. Retinal ganglion cells (RGCs), the terminal neurons within the retina, pass these signals to the brain via axons bundled in the optic nerve. RGCs in the retina as well as their axons in the optic nerve are highly sensitive to damage observed in glaucoma and other optic neuropathies. Due to their position in the inner most portion of the retina, RGCs are accessible to molecular treatments via intravitreal injections. Our experiments explore the short and long-term survival of RGCs following intravitreal injection of AAV2-Cre recombinase or a control virus in wild type mice. Retinal flat mounts and axonal cross sections were assayed for viral transduction and RGC survival at various time points post injection. Short-term experiments demonstrate that AAV2-mediated Cre recombinase expression is efficient and well tolerated in the wt mouse retina. However, Cre toxicity of RGC was observed at 5 weeks post injection of AAV2-Cre. Collectively these findings suggest that viral delivery of Cre recombinase may be an effective strategy for short-term experimental studies, but not as useful for studies requiring persistent Cre expression.

This project is supported by funding from the Commonwealth Health Research Board, JMU 4-VA, and The BrightFocus Foundation. We also thank the Merbs Lab at Johns Hopkins University.

Morning Poster Session
Group N - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

78. **STUDIES ON NEURITE OUTGROWTH BY EXTRACELLULAR MATRIX PROTEINS USING RAT CEREBELLAR GRANULAR NEURONS**

Sumiah Ali, Umefarwa Wasim, Alia Saddique, Ijaz Ahmed, and Alam Nur-E-Kamal
Department of Biology, Medgar Evers College, 1638 Bedford Avenue, Brooklyn, NY 11225

79. **A NOVEL ENVIRONMENT TO EXAMINE SOCIAL STRESS AND HIERARCHY IN MICE**

Meagan Darling, Hannah Belski, and R Parrish Waters
Department of Biology, University of Mary Washington, 1301 College Avenue,
Fredericksburg, VA 22401

80. **MODULATION OF VALENCE-SPECIFIC LEARNING IN THE BASOLATERAL AMYGDALA BY NEUROTENSIN**

Kritika Chugh[1], Praneeth Namburi[2], and Kay Tye[2]
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[2]Picower Institute for Learning and Memory, MIT,
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81. **STUDYING THE ROLE OF THE RIGHT HEMISPHERE IN LANGUAGE RECOVERY AFTER STROKE**

Miriam Flynn[1], Laura M. Skipper-Kallal[2], and Peter E. Turkeltaub[2,]
[1]Department of Biochemistry, Trinity Washington University, 125 Michigan Avenue NE,
Washington DC 20017
[2]Department of Neurology, Georgetown University Medical Center, 4000 Reservoir Road NW,
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82. **THE EFFECT OF CYCLIN A2 DELETION ON CORTICAL NEURON MORPHOLOGY**

Kenechukwu Mbonu[1], Patrick Gygli[2], and Jose Otero[2]
[1] Department of Biological Sciences, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
[2] Department of Pathology, The Ohio State University,
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83. **EXAMINATION OF TRANSGLUTAMINASE ISOFORM EXPRESSION IN NEURITE OUTGROWTH**

Carrie Pfleiger, Holly Langdon and Kristen L. Boeshore
Department of Biology, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

84. **BEHAVIORAL COMPARISON OF OLFACTION RECOVERY IN WILD TYPE AND SKN-1A KNOCKOUT MICE AFTER IRRITANT EXPOSURE**

Julianna Sun[1], Kayla Lemons[1], Mario Rodriguez[2], David Dunston[1], and Weihong Lin[1]
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[2]Department of Biology, University of Miami, Coral Gables, FL 33124

STUDIES ON NEURITE OUTGROWTH BY EXTRACELLULAR MATRIX PROTEINS USING RAT CEREBELLAR GRANULAR NEURONS

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Injury in nervous system including spinal cord injury triggers a cascade of degenerative changes leading to cell death and cavitation. Severed axons fail to regenerate across the scar tissue and are only capable of limited sprouting. Extracellular matrix proteins such as Laminin, fibronectin, tenascin-C etc. are known to stimulate neurite (axon) growth. We have mastered *in vitro* model to culture primary neurons isolated from the cerebellum of young rat pups and demonstrated that extracellular matrix proteins stimulate neurite (axon) growth. Extracellular matrix proteins (e.g. laminin fibronectin) was coated on cell culture dishes. Cerebellar granule neurons (neuronal precursor cells) were seeded onto these culture dished. Neuron were allowed to grow for followed by staining with anti-beta tubulin III antibodies and visualized under a fluorescent microscope. Length of neurite was determined using ImageJ software.
Results: We have successfully developed a primary neurite culture condition. Neuronal precursor cells isolated from young rat pups were found to develop neurites in our culture condition. It was also found that laminin stimulates the growth of neurite as measured by their length.

A NOVEL ENVIRONMENT TO EXAMINE SOCIAL STRESS AND HIERARCHY IN MICE

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Department of Biology, University of Mary Washington, 1301 College Avenue, Fredericksburg, VA 22401

Mice form social hierarchies in which dominant mice display aggressive behavior toward subordinates. These interactions have profound effects on the behavior and physiology of both dominant and subordinate mice. Furthermore, these effects in mice are often used to model human disorders, such as depression and anxiety, which typically stem from exposure to social stress. We group-housed male CD1 mice (five per cage) in a novel cage setup, and assessed dominance status using home cage behavior and performance in the tube test. To compliment these behavioral measures, we assessed fecal levels of testosterone and corticosterone using ELISA. Finally we evaluated central monoamine (dopamine, serotonin, norepinephrine) levels using HPLC in select brain areas involved in psychological stress response. Individuals that exhibited the most frequent aggressive behaviors, such as chasing or attacking cagemates, were assigned a dominant status. Subordinate mice received more aggressive advances than other mice. Unexpectedly, both of our behavioral measures demonstrated shifts in the social hierarchy throughout the experiment. Our data suggest that a decline in social rank is associated with an increase in fecal corticosterone, while a rise in social rank is associated with an increase in fecal testosterone. Central monoamine analysis suggests that a subordinate status is associated with increased norepinephrine in the amygdala, and that falling in hierarchical rank is associated with increased global dopaminergic and serotonergic activity.

Special thanks to the Irene Piscopo Rodgers ’59 and James D. Rodgers Student Research Fellowship II, University of Mary Washing Biology Department, and Summer Science Institute for providing necessary funding for this project. The authors would also like to thank the University of Maryland, Baltimore County for providing the opportunity to present our findings.

MODULATION OF VALENCE-SPECIFIC LEARNING IN THE BASOLATERAL AMYGDALA BY NEUROTENSIN

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Online access of this abstract is restricted at the request of the Principal Investigator.

STUDYING THE ROLE OF THE RIGHT HEMISPHERE IN LANGUAGE RECOVERY AFTER STROKE

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Approximately 20% of stroke survivors experience chronic aphasia, a language deficit that can occur after stroke due to left brain hemisphere damage. People with aphasia activate their right hemisphere when doing language tasks more than healthy people. However, it is debated whether the right hemisphere is helping or hindering recovery. If non-invasive brain stimulation is used to inhibit right brain hemisphere activity during language therapy, will it improve aphasia recovery? Participants in this study fell into three groups. The first were people who suffer from chronic aphasia who received brain stimulation paired with language treatment. This group was then compared to people who also have chronic aphasia but receive "sham" stimulation (placebo group), and a third group of individuals without neurological injury. Participants were assessed before treatment, immediately after treatment and three months later, to see whether brain stimulation aided in their recovery. They also experienced an MRI scan at all time points, to see whether brain stimulation changed the way their right hemisphere responded. Participants looked at pictures of objects while being scanned in the MRI, and were asked to say the name of the object out loud. I scored trials based on whether a participant's response was correct or incorrect during testing. Special techniques were used to identify critical areas of the brain required to successfully name the pictures. I overlapped participant brain scans over the images of these critical areas, and precisely calculated the proportion of the critical area that was lesioned for each patient. This was used to test the degree to which lesion location accounted for each participant's language deficit. I also scored various cognitive tests including measures of attention, spatial awareness, and language production. I gained skills in scoring a wide range of cognitive measures and basic neuroimaging techniques during my internship.

This project is supported by the Doris Duke Charitable Foundation (2012062) and NIH/NCATS KL2TR000102 to P.E.Turkeltaub.

THE EFFECT OF CYCLIN A2 DELETION ON CORTICAL NEURON MORPHOLOGY

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[2] Department of Pathology, The Ohio State University,
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Unrepaired DNA damage in neurons is extremely detrimental to the developing nervous system. If the DNA damage is not corrected it can lead to neuron cell death one of the causes of many neurological disorders including Alzheimer's and Parkinson's disease. If neurons corrected DNA damage before it killed brain cells these disorders may be avoidable. It has been shown that Cyclin A2, a cyclin involved with the S phase of the cell cycle, is present in neurons and may be involved with DNA damage repair. Using a cre lox transgenic mouse approach the lab has noticed that mice where Cyclin A2 has been deleted in forebrain neurons display varicosities on the dendrites of cortex neurons more than mice with Cyclin A2. In order to objectively quantify this difference I used StereoInvestigator software to count the varicose neurons after staining them using the Golgi Impregnation method. I found that the control mice had a lower number of varicose neurons in comparison to the mutant and heterozygous mice. The next step is to quantify additional mouse brains and then look at the behavior of these mice to see if there is a difference in the mice without Cyclin A2 compared to the control mice.

This investigation was supported, in part, by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences.

EXAMINATION OF TRANSGLUTAMINASE ISOFORM EXPRESSION IN NEURITE OUTGROWTH

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Retinoic acid (RA) has been shown to promote axonal regeneration from injured neurons after an acute spinal cord injury. In the work presented here, the role of RA in promotion of neurite regeneration was examined in PC12 cells, a well-established cell line used for studying the mechanisms involved in differentiation of neurons. Specifically, the ability of RA to induce transglutaminase-2 (TG-2) expression was investigated. Western blot analysis was used to determine if RA stimulation of injured cells increases expression or alters the relative expression of the long (TG-2L) and short (TG-2S) isoforms. Since transamidation activity of TG-2 has been linked to promotion of neurite outgrowth, it was expected that RA stimulation would increase expression of TG-2S, a form which exhibits greater transamidation activity. Additionally, real-time RT-PCR was carried out to examine relative expression of TG-2 isoforms at the level of the mRNA. Both isoforms of TG-2 were detected in cells at various amounts. Replication of the experiment and quantification of the isoforms are ongoing. Due to complications with the current PCR parameters, optimization of real-time RT-PCR using new samples and parameters is continuing.

This work was supported by the Biology Department at Lebanon Valley College.

BEHAVIORAL COMPARISON OF OLFACTION RECOVERY IN WILD TYPE AND SKN-1A KNOCKOUT MICE AFTER IRRITANT EXPOSURE

Julianna Sun[1], Kayla Lemons[1], Mario Rodriguez[2], David Dunston[1], and Weihong Lin[1]
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Online access of this abstract is restricted at the request of the Principal Investigator.

Morning Poster Session
Group O – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

85. **CARBON DIOXIDE CYCLES: EXPLORING AND INTEGRATING THE SIGNIFICANCE OF CO_2 IN THE UNDERGRADUATE LABORATORY**

Jessica Bursler and Trevor Daly
DuPont Interdisciplinary Science Learning Laboratories, University of Delaware, Newark, DE 19717

86. **OUTER PRODUCT ANALYSIS (OPA) FOR CALIBRATION TRANSFER**

Cannon Giglio and Steven D. Brown
Department of Chemistry and Biochemistry, University of Delaware, 163 The Green, Newark, DE 19716

87. **ENHANCING STUDENT COMPREHENSION OF SPECTROSCOPY THROUGH THE IMPLEMENTATION OF NEW INSTRUMENTATION INTO THE CHEMISTRY CURRICULA**

Marissa Gouker, Perry Wood, and Debra Ellis
Department of Science, Frederick Community College, 7932 Opossumtown, Frederick, MD 21702

88. **EXPANDING INSTRUMENTATION ACCESS THROUGH THE INCORPORATION OF PORTABLE X-RAY FLUORESCENCE SPECTROMETRY INTO THE CURRICULUM**

Alexander Jarnot, Angela Mansfield, Kevin Bennett, and Christopher Stromberg
Department of Chemistry, Hood College, 401 Rosemont Avenue, Frederick, MD 21702

89. **OPTIMIZATION OF NATURAL PRODUCT EXTRACTION FOR INCORPORATION INTO AN UPPER-LEVEL UNDERGRADUATE ORGANIC SYNTHESIS LABORATORY: EFFICIENT ISOLATION AND DERIVATIZATION OF SHIKIMIC ACID**

Gina G. To and Steven M. Kennedy
Department of Chemistry, Millersville University, 1 S. George Street, Millersville, PA 17551

CARBON DIOXIDE CYCLES: EXPLORING AND INTEGRATING THE SIGNIFICANCE OF CO_2 IN THE UNDERGRADUATE LABORATORY

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In a new integrated introduction to Biology and Chemistry program at the University of Delaware, carbon dioxide is weaved throughout the curriculum. Two sister experiments were developed for the undergraduate laboratory, allowing students to explore the roles played by carbon dioxide in blood buffers and leaf decomposition. Chemical buffers and cardiovascular physiology are two concepts discussed in Chemistry and Biology, respectively, and the topic of blood buffers integrates these subjects while utilizing a real world example. Carbon dioxide plays a large role in the creation of the blood buffer and can be controlled with respiration, an idea students will explore more in depth in lab. On the large-scale side of the curriculum, the carbon cycle is a major topic, integrating ecology concepts and chemical equilibrium. In the second lab, students will explore the carbon cycle by analyzing leaf decomposition and relating their findings to the big picture. Following the created blood buffer lab procedure, accurate and precise outcomes were produced consistently. Therefore, the lab is being taught this fall for two sections of the introductory biology and chemistry courses. While the leaf decomposition lab matches the curriculum well, it still needs further work before implementation. Eventually, these two labs will be laced together, exploiting the prevalence of carbon dioxide in biology, chemistry, and real world concepts.

OUTER PRODUCT ANALYSIS (OPA) FOR CALIBRATION TRANSFER

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In chemical data analysis it is desirable to build calibrations that work across multiple instruments, but this is often difficult due to small but important differences that occur between instruments. A mathematical transform called Outer Product Analysis (OPA) has been combined with Piecewise Direct Standardization (PDS) to improve the results obtained from multi-instrument calibration. Using a data set consisting of samples of corn meal measured on three near-infrared (NIR) spectrometers, calibration models were built by using PDS, as well as by using OPA and PDS. It was found that models produced using OPA and PDS gave lower prediction errors on data from the three instruments than those developed using PDS alone.

This research was supported by the Department of Chemistry and Biochemistry Alumni Undergraduate Research Fellowship, sponsored by Mr. David Plastino, and by NSF Grant No. 1506853

ENHANCING STUDENT COMPREHENSION OF SPECTROSCOPY THROUGH THE IMPLEMENTATION OF NEW INSTRUMENTATION INTO THE CHEMISTRY CURRICULA

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The Spectronic 20 has been heavily relied upon at Frederick Community College's (FCC) chemistry laboratory experiments such as visible spectroscopy for the identification and quantitation of colored compounds. Outdated and approaching disrepair, the non-visual display of the Spectronic 20 inhibits full student comprehension of visible spectroscopy and the relationships between its respective measurements.

Recently the Science Department at FCC has purchased a more student-friendly alternative: the SpectroVis Plus Visible to Near-Visible Spectrometer and Fluorometer. The SpectroVis can be partnered with either the Vernier LabQuest 2 data collector or laptops available for student lab-use. Both instruments provide students with a full-spectrum display that enhances their conceptual understanding of various aspects of spectroscopy including absorbance, transmittance, the visible spectrum, and kinetics in action.

In order to make the transition between the Spectronic 20 and the SpectroVis as seamless as possible in FCC chemistry labs, the current in-house general chemistry lab manuals required a detailed procedure on how to interact with the interface of the SpectroVis and utilize the data-collecting functions of either the laptop or the LabQuest 2. Three currently in-use general chemistry (CH 101 & CH 102) lab procedures were revised to incorporate the SpectroVis Plus and appropriate data collector(s). In addition, a new experiment was created to increase CH 102 student's expertise in the use of the new instrumentation. These experiments have been included in the CH 101 and CH 102 lab manuals for the 2015-2016 school year.

EXPANDING INSTRUMENTATION ACCESS THROUGH THE INCORPORATION OF PORTABLE X-RAY FLUORESCENCE SPECTROMETRY INTO THE CURRICULUM

Alexander Jarnot, Angela Mansfield, Kevin Bennett, and Christopher Stromberg
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X-Ray Fluorescence is a phenomena in which an incident x-ray ejects an inner shell electron from an atom, whereby an electron from a higher shell falls down to fill the vacancy left by the ejected electron. The energy from the electron falling is given off in the form of a fluorescent x-ray. These fluorescent x-rays can be detected and the energies measured to quickly identify the elements present in the sample. For lighter elements, the inner shell electron comes from the K-shell. However, in the case of heavier elements, the K-shell electrons are held too tightly. Therefore, an L-shell or an M-shell electron is ejected.

Hands-on experience with different instrumentation is crucial for developing a proper understanding of the fundamentals of different analysis techniques. Therefore, laboratory experiments were developed to be incorporated into General Chemistry, Quantitative Analysis, Instrumental Analysis, and Quantum Mechanics classes in order to expose students to a wider variety of instrumentation. The General Chemistry lab will focus on the non-destruction nature of the instrument and its use with complex mixtures. For Quantitative Analysis, the students will learn about the quantitative effects of the sample matrix on the signal. The Instrumental Analysis lab will delve into signal-to-noise ratios. Finally, the Quantum Mechanics experiment will examine the theory behind x-ray fluorescence by predicting characteristic x-rays. The experiments are designed to focus on different aspects of the instrument. Therefore, come graduation, the students will have gained a deeper understanding of the theory and applications of x-ray fluorescence. This experience will better prepare students for a future in the sciences, whether it be in the workforce or graduate school.

This work was funded by the National Science Foundation's Improving Undergraduate STEM Education program (award number DUE-1431522).

OPTIMIZATION OF NATURAL PRODUCT EXTRACTION FOR INCORPORATION INTO AN UPPER-LEVEL UNDERGRADUATE ORGANIC SYNTHESIS LABORATORY: EFFICIENT ISOLATION AND DERIVATIZATION OF SHIKIMIC ACID

Gina G. To and Steven M. Kennedy
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A new, user-friendly, method for the rapid extraction of Shikimic acid (SA) from star anise, based on the work of Just and coworkers, has been optimized for an upper-level organic laboratory. Currently, our studies are focused on the development of a modified procedure that can be adapted into an undergraduate laboratory. The synthesis of SA derivatives is also being explored. To date, approximately 2.0 g of SA can be isolated and purified within an average of a 120 min time period, starting from 20 g of star anise. The ease and efficiency of this method allows SA, which is commercially available, but cost prohibitive (2.0 g = $206), to be used as the starting material for a six-week multi-step synthesis laboratory. SA derivatives have been shown to exhibit useful biological activities. Particularly, they act as viral neuraminidase inhibitors, display anticancer, antiviral and antibiotic behavior, or exhibit anticoagulant and antithrombotic activity. During the first half of a sixteen-week semester, students will use SciFinder Scholar to help them plan a step-wise organic synthesis that employs functional group protected SA derivatives as intermediates. After isolating SA with our modified extraction protocol, students will implement their multi-step synthesis.

These results were obtained during the summer of 2015 as part of the Independent Research program in the Department of Chemistry at Millersville University. This work was funded in part by the generous contributions of Professor Emeritus Robert K. Wismer. We would also like to thank the Chemistry Department Faculty at Millersville University, Mr. Stephen Peurifoy, and the Dean of the College of Science and Technology.

Morning Poster Session
Group P – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

90. **TOTAL SYNTHESIS OF PYROPHEN**

Hannah Burdge and Keith Reber
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91. **SYNTHESIS OF ANTISENSE NUCLEIC ACID MONOMERS**

Matthew Bowen*, Matthew Davisson*, and Debbie. Mohler
Department of Chemistry and Biochemistry, James Madison University, 901 Carrier Drive, Harrisonburg, VA 22807

92. **CO(II)-SALEN CATALYZED CARBON-CARBON BOND FORMATION VIA C–H FUNCTIONALIZATION FOR THE ELABORATION OF HETEROCYCLES**

Andrew De Los Santos[1], Andrew Schafer[2], and Simon Blakey[2]
[1]Department of Chemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250
[2]Department of Chemistry, Emory University, 1515 Dickey Drive, Atlanta, GA 30322

93. **SYNTHESIS OF AMIDE PENDANT RUTHENIUM HOST SYSTEMS**

Molly McBride and Marc Harris
Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

94. **OPTIMIZING THE MANUAL SOLID-PHASE SYNTHESIS OF CRITICALLY RACEMIZATION-PRONE ARYLGLYCINE-CONTAINING PEPTIDES FOR USE IN ANTIBIOTIC-RELATED RESEARCH**

Aurelio Mollo, Joshua Bulos and Louise Charkoudian
Department of Chemistry, Haverford College, 370 Lancaster Avenue, Haverford, PA 19041

95. **SYNTHESIS OF PRELIMINARY HETEROCYCLIC SALTS FOR CYANINE DYES**

Dominique Munson* and Angela Winstead
Department of Chemistry, Morgan State University, 1700 East Cold Spring Lane, Baltimore, MD 21251

96. **THE SYNTHESIS OF ETHYL AND METHYL BENZOATE WITH A REDUCED REFLUX TIME**

Paola Kleimann[1], Matthew Koury[1], Jacqueline Rowan[1], Patricia Kreke[2],
[1]Department of Science, Mount St. Mary's University, 16300 Old Emmitsburg Road, Emmitsburg, MD 21727

97. **ALKENE MIGRATION STUDIES EN ROUTE TO ALTERSOLANOL P**

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TOTAL SYNTHESIS OF PYROPHEN

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A total synthesis of the novel compound pyrophen, isolated from the marine fungus *Aspergillus niger* and known to have antifungal activity, is reported for the first time. Beginning from L-phenylalanol, this synthesis is achieved in seven steps. The key steps of this synthetic route are an aldol reaction uniting the two major fragments of the molecule and a two-step, one-pot reaction in which a dioxinone is opened with a sacrificial alcohol, and the resulting tricarbonyl compound is cyclized under basic conditions. The title compound was isolated in approximately 11% yield overall, and efforts to extend this synthetic sequence to access a related family of natural products, the campyrones, are currently in progress.

We would like to thank Towson University for supporting this project through both startup funding and a Faculty Development and Research Committee grant (#150174).

SYNTHESIS OF ANTISENSE NUCLEIC ACID MONOMERS

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Due to RNA's ability to be used as a natural therapeutic for its gene silencing capabilities, antisense oligonucleotides have been studied for their wide variety of uses as therapeutic drugs or for use as a tool for studying gene function. However, to avoid problems with degradation of the sugar-phosphate backbone, synthetic oligonucleotide analogs that lack traditional ribose-phosphate backbone are being developed and will be studied for their ability to bind DNA and to silence gene expression.

We would like to thank the NSF: REU grant *CHE-1461175* for funding.

CO(II)-SALEN CATALYZED CARBON-CARBON BOND FORMATION VIA C–H FUNCTIONALIZATION FOR THE ELABORATION OF HETEROCYCLES

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Heterocycles are highly useful and often necessary motifs in medicinal and pharmaceutical products. Furthermore, many methods have been developed for the synthesis of aromatic heterocycles with benzylic or alkyl substitution due to their ability to help in binding drugs to their targets in the body. While a great number of methods for preparing heterocycles exist, often times these methods rely on significant functional group manipulation, adding synthetic steps and therefore significant cost and potential byproduct formation issues to their production. For this reason, we have been looking into reactions involving metal-catalyzed carbene and nitrene C–H insertion to prepare heterocycles via direct activation of C–H bonds to prepare pharmaceutically relevant heterocycles.

We believe that through the use of a chiral metal salen complex, we may exert stereocontrol for the addition of allylic carbons to the heterocycles from the reactive hydrazones. We hypothesize that through the use of a cobalt(II)-salen complex, we can conduct enantioselective reactions between heterocycles and *N*-tosylhydrazones to synthesize heterocycles of specific enantiometric configuration With this method in hand we can mass synthesize these heterocycles for medicinal and pharmaceutical applications.

Throughout the course of this project, we were able to produce an *N*-tosylhydrazone from acetophenone to react with benzoxazole, a 1,3 azole, and a cobalt(II)-salen ligand in order to synthesize heterocycles of specific enantiometric configuration. Through various screenings of temperatures, bases, and solvents, we were unable to collect any yield of the desired product. Due to this we intend to continue the screening of various temperatures, bases, and solvents to find the optimal conditions for the reaction.

This material is based upon work supported by the Howard Hughes Medical Institute Science Education Program award #52006923 to Emory University, and the NSF Center for C-H Functionalization (NSF CHE-1205646). Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the Howard Hughes Medical Institute or Emory University.

SYNTHESIS OF AMIDE PENDANT RUTHENIUM HOST SYSTEMS

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Key bipyridine containing amide pendant ligands were synthesized and coordinated to Ruthenium, a photoactive transition metal. This allows for ion binding studies to determine how efficient and selective a host is towards a specific ion. The photo activity of these host-guest systems can be probed using fluorescence titration studies. Ligands and host complexes were characterized by mass spectrometry and ^{1}H NMR spectroscopy.

This project was funded by the LVC Research Endowment Fund.

OPTIMIZING THE MANUAL SOLID-PHASE SYNTHESIS OF CRITICALLY RACEMIZATION-PRONE ARYLGLYCINE-CONTAINING PEPTIDES FOR USE IN ANTIBIOTIC-RELATED RESEARCH

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Complestatin is a heptapeptide with promising antibacterial, anticomplement and anti-HIV properties. Although the molecule has been synthesized, current methods require numerous steps and complex transition metal chemistry, and suffer from critically low yields. We are developing a biomimetic synthetic strategy which involves the synthesis of the linear heptapeptide, followed by enzymatic cross-linking of the aromatic side-chains to impart the crucial rigidification of the structure required for biological activity. To this end, we seek to understand the molecular and enzymatic bases of the two crucial oxygenase-catalyzed linkages: an aryl-aryl bond and an aryl-ether-aryl bond. In preparation for turnover assays with the oxygenase ComJ (putatively involved in aryl-ether-aryl bond installment), we developed a manual solid-phase peptide synthesis (SPPS) protocol to obtain the relevant tripeptide substrates for cyclization.

For most peptides, SPPS is a standard process. In our case, however, the presence of the base-labile 4-hydroxyphenylglycine residues in our target products render standard SPPS obsolete. In response, we developed an efficient synthetic route that is void of harsh reaction conditions. Key steps such as resin loading and capping, amino acid coupling, peptide capping and acid-catalyzed peptide cleavage were investigated in depth with the ultimate goal of reducing racemization in these critically racemization-prone peptides.

In the future, we will apply this route to generate the peptide substrates in their carrier-protein bound form to evaluate the underpinning mechanisms for aryl-ether-aryl bond formation in complestatin biosynthesis.

We would like to thank Haverford College's Marian E. Koshland Integrated Natural Sciences Center and the Research Corporation for Science Advancement Cottrell College Scholars Award (L.K.C.) for funding.

SYNTHESIS OF PRELIMINARY HETEROCYCLIC SALTS FOR CYANINE DYES

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In the past, scientists have studied and used pentamethine (Cy5) dyes as fluorescent probes for pathogen detection. They have found that most Cy5 dyes fluoresce at 670nm. Unfortunately, some biomolecules fluoresce around 650nm which causes background interference when Cy5 dyes are used. Heptamethine (Cy7) cyanine dyes have become of interest to scientists because of their significant ability to luminescence in the near-infrared region (650-900nm) which causes minimal background interference.

Sulfoindocyanine dyes are one of the many Cy7 dyes that have been used as a fluorescence probes. Sulfoindocyanine dyes are highly water soluble compounds that are ideal for biological life forms. This study will focus on the synthesis of sulfoindocyanine dyes. The specific aims of this approach are synthesis of the potassium sulfoindole starting material, sulfoindole heterocyclic salt derivatives, and water soluble dye derivatives. The starting material was synthesized via conventional organic techniques, yielding 68%. The sulfoindole heterocyclic salt derivatives were synthesized by using a Biotage microwave yielding between 50-80%. Ethyl and carboxyl sulfoindocyanine 7 dye were synthesized with yields ranging from 50-75%. All structures were successfully determined by 1H NMR. The dyes were also successfully characterized using Cary50 UV/Vis spectrometer and HPLC.

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THE SYNTHESIS OF ETHYL AND METHYL BENZOATE WITH A REDUCED REFLUX TIME

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The synthesis of ethyl benzoate or methyl benzoate is a procedure that all organic chemistry students at Mount Saint Mary's University complete. The reflux step takes 4 hours to complete, and shortening this step would make the experiment less time consuming for the students and professors.

This experiment is an effort to shorten the pre-existing procedure in the synthesis of ethyl benzoate and methyl benzoate. Alterations were made to the reflux step in order to shorten the time required. A stirring bar was used to test if stirring made the process more efficient and then the time was halved to observe changes in the purity and yield of the ethyl benzoate. Stirring did not have a significant effect on the yield and purity of the ethyl benzoate. When only half of the time was used in the reflux, the ethyl benzoate product was still as pure as when the full time was used, and the yield was also the same. These results indicate that using half of the reflux time would be acceptable as it yields similar results. The same procedure will now be followed with methyl benzoate. The results for methyl benzoate are predicted to be similar to ethyl benzoate or will show a greater yield and purity compared to the ethyl benzoate.

ALKENE MIGRATION STUDIES EN ROUTE TO ALTERSOLANOL P

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Studies toward the total synthesis of altersolanol P (AP), a new member of the altersolanol family of compounds, have been conducted in our laboratory. AP was recently isolated from an unknown Hypocreales fungus collected at a forest in Puerto Rico. AP exhibited broad-spectrum activity against Gram-positive bacteria and inhibited the growth of Gram-negative *Haemophilus influenzae*. Currently, a synthetic intermediate containing the complete carbon framework of AP has been synthesized via Lewis acid-mediated Diels-Alder cycloaddition on multi-gram scale (in 80% yield and 8:1 regioselectivity). An oxidization reaction of the cycloadduct provided efficient access to a 1,4-diene intermediate. From the 1,4-diene, a sequence of alkene isomerization followed by dihydroxylation should give rise to AP. Our efforts are focused on a methods study to isomerize the 1,4-diene to the 1,3-diene. The long-term goal of this project is to find an efficient route to synthesize Altersolanol P and related derivatives so that their biological activities can be further studied.

The authors acknowledge the Niemeyer-Hodgson Student Research Grant, the Noonan Endowment Award, the Millersville University Student Research Grant. We would also like to thank the Chemistry Department Faculty at Millersville University, Mr. Steven Peurifoy, and the Dean of the College of Science and Technology.

Morning Poster Session
Group Q – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

98. **STUDIES TOWARD THE TOTAL SYNTHESIS OF HUNANAMYCIN A**

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Department of Chemistry, Millersville University, 1 S. George Street, Millersville, PA 17551

99. **MICROWAVE ASSISTED ORGANIC SYNTHESIS OF HEPTAMETHINE CYANINE DYES**

Jahnn Drigo and Angela Winstead
Department of Chemistry, Morgan State University, 1700 E. Cold Spring Lane,
Baltimore, MD 21251

100. COMBINATION OF BIRCH REDUCTION-ALKYLATION, ESTERIFICATION AND ALLYLIC OXIDATION IN THE SYNTHESIS OF COMPLEX BIOACTIVE MOLECULES

Hamna Shahnawaz and William P. Malachowski
Department of Chemistry, Bryn Mawr College, 101 N. Merion Avenue, Bryn Mawr, PA, 19010

101. SYNTHESIS AND CHARACTERIZATION OF PENDANT PHENYL ESTER-SUBSTITUTED THIOPHENE BASED COPOLYMERS

Devon M. Shircliff , Vincent J. Pastore, Michael L. Poltash, and Brycelyn M. Boardman
Department of Chemistry and Biochemistry, James Madison University,
Harrisonburg, VA 22807

102. **SYNTHESIS OF A BIOORTHOGONAL TYROSINE LABELING LINKER**

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103. **FLUORESCENCE PROPERTIES OF 6,7-DIPHENYL-2(1H)-THIOXO-4(3H)-7,8-DIHYDROPTERIDINONE**

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104. **COCRYSTALLIZATION OF N,N'-DIPHENYLUREAS AND TRIPHENYLPHOSPHINE OXIDE BASED ON HETERODIMER ENERGIES**

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STUDIES TOWARD THE TOTAL SYNTHESIS OF HUNANAMYCIN A

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Studies toward the synthesis of Hunanamycin A (HA) have recently been initiated in our laboratory. HA is a natural product isolated in small quantities (< 1 mg) from *Bacillus hunanensis*. It exhibits antibacterial activity for various pathogens such as *Salmonella* and *E. coli.* Conceivable synthetic routes to HA have been designed based on literature precedent. Test reactions (*e.g.* reductive amination, amine acylation, and cyclization) are being optimized on model systems to explore multiple pathways of producing the target product. Once an efficient route is elucidated, further biological testing of HA and related derivatives could allow for a calculated modification of the antibacterial properties displayed by this class of molecules. Currently, two methods, reductive amination and amine acylation, have achieved formation of a prenylated aromatic amine intermediate. Our most progressive route employs an intramolecular electrophilic cyclization of the prenylated aromatic amine to provide a tetrahydroquinoline intermediate.

Our research moves forward with funding from the Neimeyer-Hodgson Student Research Grant, the Noonan Endowment Fund Grant, Millersville University Student Research Grant, and Millersville University Faculty Grant. We would also like to thank the Chemistry Department Faculty at Millersville University, Mr. Steven Peurifoy, and the Dean of the College of Science and Technology.

MICROWAVE ASSISTED ORGANIC SYNTHESIS OF HEPTAMETHINE CYANINE DYES

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Fluorescent labeling is important in detecting infectious diseases; cyanine dye labels are therefore indispensable to controlling the spread of pathogenic disease. The pentamethine cyanine (Cy5) dyes popularly employed suffer biological interference from molecules that fluoresce in their region of the electromagnetic spectrum (650nm-700nm).

To alleviate this shortcoming, heptamethine cyanine (Cy7) dyes that fluoresce at 780nm-1000nm have been synthesized using Microwave-Assisted Organic Synthesis (MAOS), a more efficient and environmentally friendly technique compared to traditional reflux heating. Ethyl- and propyl-benzoindolenine salts were synthesized and characterized with yields of 89% and 86%, respectively, by combining 2, 2, 3-trimethylbenzoindolenine with the corresponding alkyl halide. The dyes were then synthesized in condensation reactions between the salt precursors, bisimine, sodium acetate, and a suitable solvent in a Biotage Microwave Initiator 2.0.

A 77% average yield of the benzo-ethyl dye was obtained, compared to 44% yields cited in the literature. The benzo-propyl dye was synthesized with 85% yield in only ten (10) minutes, an enormous reduction when contrasted with 18 hours stated in publications wherein the conventional method was used. In addition, methyl bromovalerate ester was used to prepare the 5-bromovaleric acid derivative of trimethylbenzoindolenine. Future work includes the synthesis of salts with other R-groups and other dyes with the excellent spectral properties noted in the ones already made.

The author wishes to thank Dr. Angela Winstead, Grace Nyambura, Dr. Richard Williams, and Tijesunimi Odebode for their invaluable contributions to this project, as well as the US Department of Defense W911NF-11-1-0157 sponsors, without whom these achievements would not have been possible.

COMBINATION OF BIRCH REDUCTION-ALKYLATION, ESTERIFICATION AND ALLYLIC OXIDATION IN THE SYNTHESIS OF COMPLEX BIOACTIVE MOLECULES

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The formation of carbon-carbon bonds are crucial for the synthesis of biologically active molecules however the reactions that form these bonds have proven to be a challenge. The challenges faced in the organic synthesis of natural products include constricted rings, complex stereochemistry and all carbon centers. It is essential to try to overcome these problems as natural products and natural product derivatives play a crucial role in the design of novel therapeutic agents. A large number of bioactive molecules contain four carbon quaternary centers and so it is essential to explore the formation of these molecules with a high yield and in an enantiomerically selective way. The three step sequence provided is the start to the creation of highly complex and functionalized molecules in an enantioselective way.

I would like to thank the National Science Foundation for financial support for the NMR instrument and Bryn Mawr Summer Science Research for their financial support of this project.

SYNTHESIS AND CHARACTERIZATION OF PENDANT PHENYL ESTER-SUBSTITUTED THIOPHENE BASED COPOLYMERS

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Recently applications in polymer solar cells (PSCs) have been of increasing interest in the field of solar energy due to these polymers strong light harvesting capabilities, low production costs and solubility in a wide variety of organic solvents. PSCs containing poly-3-hexylthiophene (P3HT) as the electron-donor material and [6,6]-phenyl-C_{61}-butyric acid methyl ester ($PC_{61}BM$) as the organic electron-acceptor are the most extensively studied. However, these subsequent devices only exhibit power conversion efficiencies up to 4-5%. Low band-gap polymers have also recently been investigated. These structures absorb a wide range of the solar spectrum but suffer from low device efficiencies due to poorly ordered structures. In order to improve the ordered solid state packing of these polymers, the addition of π-conjugated side chains has been of increasing interest. However, the addition of these modifications negatively affects the solubility of these materials and has proven to be rather synthetically challenging.

In an attempt to synthesize low band gap copolymers, three new monomers 3-ethyl-2-(2,5-dibromothiophene)-3-ylacetate (ETA), 3-phenylethyl-2-(2,5-dibromothiophene)-3-ylacetate (PTA), and 3,3-diphenylethyl-2-(2,5-dibromothiophene)-3-ylacetate (DPTA) were synthesized. ETA, PTA, and DPTA were copolymerized with 2,5-bis(trimethylstannyl)thiophene to yield poly [3-ethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene (PETAT), poly [3-phenylethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene (PPTAT) and poly [3,3-diphenylethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene (PDPTAT) respectively. Additionally, these mildly electron-donating monomers as well as 2,5-bis(trimethylstannyl)thiophene were incorporated with the electron withdrawing monomer, 4,7,dibromobenzo[*c*]-1,2,5-thiadiazole (BT), to develop three push-pull polymeric systems; poly [3-ethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene-2,6-diyl-alt-2,1,3-benzothiadiazole-4,7-diyl] (PETATBT), poly [3-phenylethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene-2,6-diyl-alt-2,1,3-benzothiadiazole-4,7-diyl] (PPTATBT), and poly [3,3-diphenylethyl 2-(thiophene-3-yl)acetate-2,2'-thiophene-2,6-diyl-alt-2,1,3-benzothiadiazole-4,7-diyl (PDPTATBT). Copolymers PETAT, PPTAT, and PDPTAT exhibit band gaps from 2.2 – 1.9 eV respectively, where as PETATBT, PPTATBT, and PDPTATBT show a reduction in the band gaps to 1.9-1.7 eV respectively. The impact of the phenyl moiety was investigated using atomic force microscopy; increased order within films of PPTAT and PPTATBT can be attributed to the increased π-π stacking in the solid state from the pendant phenyl moiety.

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SYNTHESIS OF A BIOORTHOGONAL TYROSINE LABELING LINKER

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Bioconjugation chemistry with proteins is restricted by the structures of the 20 naturally occurring amino acids that make up each protein, the functionality of them, and the presence of multiple of the same amino acids within each protein. Tyrosine is a nucleophilic amino acid with distinct reactivity due to the presence of a phenol ring with an acidic proton. Tyrosine is also able to undergo a click-reaction with 4-phenyl-3*H*-1,2,4-triazoline-3,5(4*H*)-dione (PTAD), which can be used in order to label tyrosine amino acids incorporated in the protein's amino acid sequence. The click-reaction of tyrosine and PTAD has been reported to have quantitative yields, showing great potential for use with tyrosine labeling.

A 4-phenyl-3*H*-1,2,4-triazoline-3,5(4*H*)-dione (PTAD) derivative with the presence of an azide functional group was synthesized through a series of reactions. The azide group on the derivative will be converted to an amine moiety, to which will be further attached to a previously synthesized linker that contains a cyclooctyne unit. After the protein labeling step, this cyclooctyne end can easily react with azide-functionalized synthetic dyes using Strain Promoted Azide-Alkyne Cycloaddition (SPAAC) bioorthogonal reaction.

Using fluorogenic dyes, which will become fluorescent in the red-near infrared region only after being bound to the previously modified protein in the selective and fast bioorthogonal reaction, the background fluorescence and auto-fluorescence can be minimized and the signal-to-noise ratio can be greatly improved. Finally, since the linker and dyes will be introduced into cells and applied under *in vivo* conditions, toxicity will be monitored carefully.

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FLUORESCENCE PROPERTIES OF 6,7-DIPHENYL-2(1H)-THIOXO-4(3H)-7,8-DIHYDROPTERIDINONE

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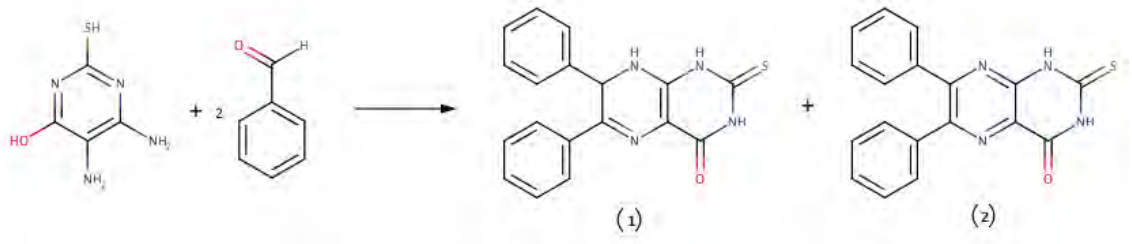
6,7-diphenyl-2(1H)-thioxo-4(3H)-7,8-dihydropteridinone, (**1**), a novel compound synthesized via the reaction scheme shown, has incredible fluorescent properties. When dissolved in DMSO, compound **1** fluoresces at 490nm with a quantum yield of 0.54 upon excitation at 418nm. The fluorescent intensity has been proven to be solvent dependent, with the highest fluorescence intensity in DMSO. Different percentages of ethanol and acetonitrile directly affect the intensity of fluorescence of compound **1**. We believe that the compound exists in either a twisted conformation or a flat conformation dependent on solvent. A related known compound, 2-mercapto-6,7-diphenylpteridin-4-ol (**2**), is a more highly conjugated system, but its fluorescence is 2-3 orders of magnitude less intense, which suggests that the benzene rings are twisted out of the pteridine plane, interfering with the conjugation. The conjugated part of compound 1 is hypothesized to be planar in solvents such as DMSO where the highest intensity is observed. The fluorescence intensity is also dependent on exposure to light, decreasing exponentially with continued light exposure. NMR shows that the fluorescence decreases because the dihydropteridine is being oxidized to the fully aromatic pteridine.



COCRYSTALLIZATION OF N,N'-DIPHENYLUREAS AND TRIPHENYLPHOSPHINE OXIDE BASED ON HETERODIMER ENERGIES

Taylor A. Watts[1], Marina A. Solomos[2], and Jennifer A. Swift[2]
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One-dimensional H-bonding motifs are commonly observed in diphenyl urea (PU) crystals. In previous work, meta-substituted PUs with electron withdrawing groups were shown to cocrystallize with the H-bond acceptor triphenylphosphine oxide (TPPO) in a 1:1 ratio. The cocrystals all showed dimer formation between urea and TPPO. Density functional theory (DFT) provided a thermodynamic rationale for cocrystal formation. Only when the interaction energy of urea-TPPO > urea-urea by ~ 5.3-6 kcal/mol, was cocrystal formation realized experimentally.

In this study, we investigate PUs with ortho- and para- electron withdrawing substituents in an effort to outline similar thermodynamic trends of cocrystallization. The chemical synthesis and characterization of 8 PUs, as well as efforts to cocrystallize them with TPPO are described. Characterization techniques include ^{1}H NMR, differential scanning calorimetry (DSC), powder x-ray diffraction (PXRD), optical microscopy, and single crystal x-ray diffraction (XRD). DTF predicts cocrystal formation in 3/8 cases. The structures of 2 PUs and 2 PU:TPPO cocrystals are reported.

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Morning Poster Session
Group R – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

105. **STREAM ACID MITIGATION PLAN FOR TWO JEFFERSON NATIONAL FOREST STREAMS**

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106. **UV SPECTROPHOTOMETRIC DETERMINATION OF SULFATE USING BARIUM ION AND THE CHROMATE/DICHROMATE EQUILIBRIUM: RESULTS TO DATE**

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107. **CAPACITY OF ANTIOXIDANTS IN ARONIA MITSCHURINII WHEN EXPOSED TO INCREASED TEMPERATURES**

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108. **EFFECTIVENESS OF NON-WOVEN GEOTEXTILE FABRIC IN SLOW SAND FILTRATION**

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109. **THE EFFECT OF HOFMEISTER SERIES COUNTERIONS ON THE COLLOIDAL AND ANTIMICROBIAL PROPERTIES OF A TRIPLE-HEADED SINGLE-TAILED AMPHIPHILE**

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110. **COMPARING ANTHROPOGENIC LAND USE AND ITS IMPACT ON AGATE LAKE AND CROW WING LAKE**

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STREAM ACID MITIGATION PLAN FOR TWO JEFFERSON NATIONAL FOREST STREAMS

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North Fork Potts Creek and North Fork Stony Creek are two Appalachian Mountain streams in the Jefferson National Forest of West Virginia and Virginia. Both streams flow from the same mountain on opposite sides of the ridge. Both have acceptable cold water and thermal habitat for brook trout (*Salvelinus fontinalis)* but originate within watershed geology of little natural carbonate bearing minerals. The combination of the near absence of natural buffer and acid precipitation have created conditions of relatively low pH (pH < 5) and other water quality values which result in poor trout biomass and recruitment. In the present study water chemistry has been evaluated for the purpose of designing a mitigation strategy by the single point, single application method of introducing base material ("liming") near the headwaters. Physical size, watershed geology and discharge values have been evaluated for the two streams. Water samples have been collected and analyzed for pH, acid neutralizing capacity (ANC), base cations (Ca^{+2}, Mg^{+2}, K^{+}, and Na^{+}), strong acid anions (Cl^{-}, SO_4^{-2}, and NO_3^{-}), and aluminum. These physical and chemical parameters have been used along with results of previous liming studies to propose a single point, single application of 50 and 100 tonnes, respectively, for a 4 to 5 year treatment.

Research done with help by the NSF-REU Grant CHE-1461175, United States Forest Service-George Washington and Jefferson National Forests Dawn Kirk, USFS fisheries biologist

UV SPECTROPHOTOMETRIC DETERMINATION OF SULFATE USING BARIUM ION AND THE CHROMATE/DICHROMATE EQUILIBRIUM: RESULTS TO DATE

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In this presentation, the spectrophotometric determination of sulfate by use of a barium/dichromate reagent will be investigated, with an eye toward utilization of this method for determination of sulfate in abandoned mine drainage (AMD). The purpose of this investigation is to develop a portable, inexpensive method for sulfate determination in AMD that is more rapid than the traditional gravimetric method, and can be scaled down to minimize waste due to toxic barium- and chromate-containing reagents that must be disposed of by proper protocols. The dichromate/chromate method relies upon the solubilities of barium chromate and barium sulfate, with precipitation of sulfate as its barium salt at pH ≈ 2, and colorimetric detection of unprecipitated chromate (pH ≈ 8.5-9) at 365 nm. Additionally, the dichromate/chromate equilibrium that is core to this indirect determination of sulfate will be studied, particularly with respect to determination of an equilibrium constant for this process. Principal component analysis (PCA) will be employed for characterization of the dichromate/chromate equilibrium. Appropriate statistical methods will be used for analysis of the sulfate results obtained by the barium/chromate method.

Sample collection and preparation protocols, and details of the analytical determination of sulfate by the barium/chromate method, will be presented and discussed, as will results of the sulfate determinations, calibration data and results, and future directions for this research.

CAPACITY OF ANTIOXIDANTS IN ARONIA MITSCHURINII WHEN EXPOSED TO INCREASED TEMPERATURES

Courtney Rhoades[1], Blessing Aroh[1], Kelsey Chandler[2], Andrew Ristvey[3], and Victoria Volkis[1]
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Aronia mitschurinii, also referred to as the black chokeberry, is a fruit-bearing shrub native to Maryland. Antioxidants are an important nutrient needed for capturing naturally formed free radicals in living organisms, and prevention of oxidation and cancer formation. The aronia berry is a dark purple color which can be attributed to its increased content of anthocyanins, one kind of antioxidant. Aronia's reputation of being a super berry entices small farms to use it as a perspective specialty crop. The berry's large content of polyphenols also makes it a likely ingredient in several new products such as, organic teas and vitamin supplements. All these and classic food applications require high temperature pasteurization as a component of manufacturing. There are three major effects higher temperatures can have on antioxidants; isomerization, decomposition, or the loss of water. Here we present the data for the antioxidant content of *Aronia mitschurinii* as a function of the variation in temperature and the time of exposure to these temperatures. The aim of this project is to determine the thermal process that would avoid significant decomposition of antioxidants in aronia by studying the change present in antioxidant profiles before and after heating. Detailed measurements and analysis of anthocyanin, flavonoids, polyphenol content, and ORAC will be presented and discussed. The future direction of this project is to study the mechanisms of decomposition which take place in the aronia juice and the pulp of the aronia berry.

The project is supported by the MAES 2013-2014 SEED grant and Thurgood Marshall Undergraduate Research to Retain and Graduate Students in STEAM grant. Also, thanks both to the Richard A. Henson Honors Program and to the Department of Natural Sciences at the University of Maryland Eastern Shore for their continued assistance and support.

EFFECTIVENESS OF NON-WOVEN GEOTEXTILE FABRIC IN SLOW SAND FILTRATION

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In this study layers of non-woven geotextile fabric were used in addition to the sand filter media to determine if the fabric improves the performance of slow sand filters. Performance was measured by the removal of turbidity and total organic carbon (TOC) from the water source as well as from the mass accumulated on the fabric. Separate runs were performed with sand and 1-4 layers of fabric and a control column with sand but no fabric. Turbidity removal was shown to increase with each layer added until reaching a maximum removal at 3 layers. TOC removal in the effluent was found to decrease with each layer of fabric added. Mass accumulation was found to follow the same trend as turbidity removal. The results for turbidity removal and mass accumulation were consistent with previous studies. The results for TOC removal where unexpected but may be explained by carbon leaching off the fabric. Batch testing showed as more layers were added more TOC leaching occurred, which is consistent with column results. These findings suggest that TOC is an important factor to consider in the application of geotextile fabrics in slow sand filters. Further research is required to determine if TOC leaching can be minimized.

This research was supported by the National Science Foundation grant EEC-1263257.

THE EFFECT OF HOFMEISTER SERIES COUNTERIONS ON THE COLLOIDAL AND ANTIMICROBIAL PROPERTIES OF A TRIPLE-HEADED SINGLE-TAILED AMPHIPHILE

Kirstie Thompson[1], Elizabeth Rogers[2], Kyle Seifert[2] and Kevin Caran[1]
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Antibacterial resistance is becoming increasingly prominent causing a great need for novel antibacterial products. Several series of novel amphiphilic molecules have been synthesized with promising antibacterial results. In an amphiphile series with three hydrophilic heads and one hydrophobic tail, a linear hydrocarbon chain with 18 carbons is more antibacterial than those with longer or shorter tails. Previously, this compound has been synthesized with three bromide counterions. In this work, a group of compounds with varying anionic Hofmeister series counterions have been prepared via ion exchange. Each new amphiphile in this series has three quaternary ammonium headgroups and an 18-carbon hydrophobic tail. The effects of varying the anionic counterions on antibacterial and colloidal properties will be reported.

Funding for this project came from the Jeffrey E. Tickle ‘90 Endowment and NSF-REU (CHE-1461175).

COMPARING ANTHROPOGENIC LAND USE AND ITS IMPACT ON AGATE LAKE AND CROW WING LAKE

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When precipitation occurs, nutrients, waste and any particulates of organic matter are transferred through streams that will eventually flow into a lake. The direction of the streams moving towards the lake are determined by elevation which also creates boundaries of the watershed. My research poster compares the watershed and water quality of two Brainerd, MN area lakes: Agate Lake and Crow Wing Lake, which are different in terms of their water quality and land use within their watersheds.

Secchi depth measurements were taken to measure water clarity, chlorophyll *a* concentrations were determined by a certified laboratory, and total phosphorus concentrations were determined by both a certified laboratory and the interns in Central Lakes College's laboratory. Water samples for chlorophyll *a* and total phosphorus analysis was collected by a 2-meter integrated sampler. Watershed land use analysis was determined by Geographic Information System software (ArcGIS).

ArcGIS reveals that Crow Wing Lake's watershed is approximately three times the size of Agate Lake's watershed. Trophic state index calculations were determined: Agate Lake is mesotrophic and Crow Wing Lake is eutrophic. After the watershed boundary is determined via ArcGIS, land use within the watershed can be determined from imagery, such as developed, agricultural, brush, and forested land. It was found that there is less developed/agricultural land and more forested land in Agate Lake's watershed compared to Crow Wing Lake's watershed. A major highway also passes through the Crow Wing Lake watershed. These relationships help lake associations better understand the impact of land use on the lake's water quality.

I would like to thank the anonymous donor, contributing through the Gallaudet University Development Office, for providing financial support to the internship, and to the District of Columbia Space Grant Consortium and NASA for my stipend and lodging. Many thanks to Jeronimo Ocampos, my fellow intern, for the ArcGIS data and to Ms. Teresa LaDoucer, Science Laboratory Technician at Central Lakes College, for her generous help in the laboratory.

Morning Poster Session
Group S – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

111. **THE CORRELATION OF ZOOPLANKTON AND TOTAL PHOSPHORUS IN MINNESOTA LAKES**

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112. **MARCELLUS SHALE GAS DRILLING AND TIOGA COUNTY WATER QUALITY**

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Michele Conrad[1], and Paul Wendel[2]
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113. **ANTIFOULING PROPERTIES OF *ARONIA MITCHURINII*, *FUCUS SP.*, AND *CLATHRIA PROLIFERA***

Deirdre Johnson[1], Baruch Volkis[1], Andrew Ristvey[2], Paulinus Chigbu[1], and Victoria Volkis[1]
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114. **DEVELOPMENT OF AN ENHANCED METHOD FOR IONS IN SEAWATER**

Margaret E. LaCourse, Ian W. Shaffer, Joshua A. Wilhide, and William R. LaCourse
Molecular Characterization and Analysis Complex, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

115. **ASSESSMENT OF WATER QUALITY PARAMETERS FROM THE LOWBER ABANDONED MINE DRAINAGE TREATMENT FACILITY, PART 2: FURTHER STUDIES AND RESULTS**

Tell M. Lovelace, Aaron K. Hirshka, Luke J. Metzler, and Mark T. Stauffer
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THE CORRELATION OF ZOOPLANKTON AND TOTAL PHOSPHORUS IN MINNESOTA LAKES

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The main goal of my internship was to find whether there is a correlation between zooplankton and total phosphorous levels in Minnesota lakes.
The purpose of this research is to find a quicker and cost-efficient way to monitor the lakes' health. Total phosphorus concentrations are analyzed, because they are associated with nutrient loading in lakes, such as runoff from the lakes' watershed. Determining total phosphorus concentrations are determined by laboratory-based experiments, which can become expensive when water samples are tested by certified laboratories or by students in the laboratory with instrumentation. Why did we choose the zooplankton as a possible alternative? Zooplankton are readily accessible to everybody, easily collected with a zooplankton net, and counted with a microscope after samples are splitted. Zooplankton also are very important to the lake ecosystem, because they act as the keystone species in the food chain, which will support the fish in lakes.

Water samples from several Brainerd, MN area lakes were collected, including samples from pour points in Crow Wing Lake, and analyzed for total phosphorus and zooplankton. To determine the zooplankton density, the volume of the water column (where the zooplankton net collected zooplankton) was calculated first. Four types of zooplankton were counted: ostracod, rotifer, copepod, and cladoceran. The zooplankton count was multiplied by the splitting factor, if the plankton splitter was used, and a formula was used to calculate the estimated density of zooplankton per cubic meter. A bivariant analysis was then done to see whether there was a correlation between the zooplankton density and total phosphorus for each site. Results suggest that there is no correlation; however, there may not be enough data to support the hypothesis. This study will be continued by future interns to increase the sample size.

I would like to thank the anonymous donor, contributing through the Gallaudet University Development Office, for providing financial support to the internship. And to the Gordon Brown Fund, administered by the Gallaudet University Career Center, for my stipend and lodging.

MARCELLUS SHALE GAS DRILLING AND TIOGA COUNTY WATER QUALITY

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Since the advent of hydraulic fracturing in Pennsylvania, monitoring residential well water quality has become increasingly important. The work done in this research furthered two prior phases of research, all of which aimed to track the impacts of the Marcellus Shale gas drilling in Tioga County, Pennsylvania, on residents' water. A variety of potential contaminants were examined in conjunction with EPA limits and potential health impacts.

Water samples were collected at 82 randomly chosen wells across the county. A sampling rig equipped with a probe measured temperature, pH and specific conductivity. GPS coordinates were taken at each water well in order to evaluate the quality with respect to proximity of gas wells. Samples were carefully collected and preserved according to USGS procedure at each site. Using ion chromatography, inductively coupled plasma, gas chromatography, mass spectrometry, and alkalinity titrations, a comprehensive picture of each well was developed. Ten cations were monitored: B, Ba, Ca, Cr, Fe, K, Mg, Mn, Na, and Sr. Three anions were analyzed: Cl^-, Br^-, and SO_4^{2-}. Furthermore, the concentrations of methane and the alkalinity of each sample were reported. The analysis of the water, as well as the EPA recommended maximum concentrations and remediation techniques to correct potential problems, was mailed to each patron. A clearly defined link between gas drilling and degradations in water quality is difficult to observe, but the next phase of research in 2017 will continue monitoring fluctuations in problematic contaminants that could eventually be linked to drilling.

We would like to acknowledge the Tioga County Commissioner's Office for the Act 13 Grant that funded the supplies and stipends, the homeowners that allowed us to test their water, and the Mansfield University Chemistry Department for giving us the opportunity to conduct this research.

ANTIFOULING PROPERTIES OF *ARONIA MITCHURINII*, *FUCUS SP.*, AND *CLATHRIA PROLIFERA*

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Marine biofilm formation is the accumulation of micro/macro organism on a submerged objects. This process occurs on worldwide trade ships and results in significant increase of fuel consumption and damage to ship hulls. As well, all petroleum platforms, piers and other under-water objects are affected. Previously, there have been solutions such as tributyltin in paint products to prevent biofilm formation, but this has been proven to be harmful to marine life and are banned in most of countries. Now, the need to find an environmentally friendly compound that is able to successfully prevent biofilm formation is important in order to save money on fuel costs for ships. In this project we tested extracts from plants and algae with strong antioxidant and antibacterial properties as potential antifouling agents. Qualitative methods were developed to test anti-fouling potential of such algae, berry and sponge extracts. Extracts of berries, algae and sponges were blended with polymers to test the effectiveness of antifouling properties. Quantitative and statistical methods for comparison of the effect of different plants are still in process of development.

DEVELOPMENT OF AN ENHANCED METHOD FOR IONS IN SEAWATER

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Aquariums are an important part of modern society, providing a way to explore the wonders of the ocean, a source of entertainment, and a platform for aquatic research. Many aquariums require the production of artificial seawater with a formulation of various salts in order to match its composition to that of natural seawater to provide aquatic life with the proper nutrients needed to thrive. Assaying artificial seawater is essential for the proper maintenance of aquariums, and it is critical that ion concentrations be known with precision and accuracy. The goal of this project is to develop, optimize, and validate a robust method for the determination of halide ions (F^-, Cl^-, Br^-, I^-) in aquarium water with the purpose of maintaining a healthy ecosystem for marine life in captivity. Previous efforts have resulted in a method for determination of chloride and bromide in seawater, but the determination of fluoride and iodide is more challenging as their concentrations in seawater are relatively low, making it more difficult to detect them. The detection of fluoride can be improved by changing chromatographic conditions such as eluent composition and flow rate. However, the detection of iodide is especially difficult due to the high concentrations of chloride and sulfate in seawater. Fluoride, chloride, and bromide are best detected using conductivity detection, while the detection of iodide is best achieved using UV detection because high concentration anions in solution do not interfere with its determination as they are not detected. The aim of this project is to develop and validate a single method to detect halide ions in aquarium water using ion chromatography for separation with UV-Vis and conductivity detection in line to allow for shortened analysis, permitting validation of a comprehensive method for halide ions.

We would like to thank the National Aquarium in Baltimore, especially Jill Arnold and Kim Gaeta for their continued support, insight, and valuable discussions. I would also like to acknowledge the Office of Undergraduate Education at UMBC for sponsoring the Undergraduate Research Award program, allowing me to receive funding to perform this research.

ASSESSMENT OF WATER QUALITY PARAMETERS FROM THE LOWBER ABANDONED MINE DRAINAGE TREATMENT FACILITY, PART 2: FURTHER STUDIES AND RESULTS

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This presentation will provide an update on continued assessment of abandoned mine drainage (AMD) from the old Marchand coal mine, which is remediated at the Lowber AMD treatment facility in southwest Pennsylvania. This study is sponsored by the Rho Theta Chapter of the Gamma Sigma Epsilon Chemistry Honor Society, in collaboration with the Sewickley Creek Watershed Association (SCWA). The focus continues to be on determination of alkalinity, acidity, pH, conductivity, dissolved oxygen, sulfate, iron, aluminum, manganese, and calcium. The goals of this research continue to be: 1) assessment of selected analytes in AMD from the Lowber facility in the field and laboratory, and 2) lending assistance to the SCWA toward evaluation of AMD and development of remediation strategies for use at potential sites throughout the Sewickley Creek Watershed. Sample collection and analytical methodologies will be presented and discussed, as will results of determinations performed so far and their significance in light of the effectiveness of the passive treatment process used at Lowber, and future plans for this study.

Morning Poster Session
Group T – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

116. **SYNTHESIS OF ARTIFICIAL BACTERIAL CELL WALL FRAGMENTS AS TOOLS TO STUDY THE INNATE IMMUNE SYSTEM**

Gabriel Gregorzak, James Melnyk, Kristen DeMeester, and Catherine Leimkuhler Grimes
Department of Chemistry and Biochemistry, University of Delaware, Newark, DE 19716

117. **MULTIMERIZATION OF SOLUTION-STATE PROTEINS BY WATER SOLUBLE PORPHYRINS**

Daniel Marzolf, Aidan McKenzie, Alex Hudson, and Oleksandr Kokhan
Department of Chemistry, James Madison University, 800 S. Main Street, Harrisonburg, VA 22807

118. **COLLOIDAL AND BIOLOGICAL PROPERTIES OF M-E**

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[2]Department of Biology, James Madison University, 800 S. Main Street, Harrisonburg, VA 22807

119. **A FLEXIBLE APPROACH TO TREATING THE EBOLA VIRUS**

Natalie Steenrod, Matthew Shirley, Therese Ku, and Katherine Seley-Radtke
Department of Chemistry and Biochemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

120. **SYNTHESIS AND STUDY OF POLYCATIONIC AMPHIPHILES AS POTENT ANTISEPTICS AND NOVEL COLLOIDS: EXPLORING STRUCTURE ACTIVITY RELATIONSHIPS**

Brenna J. C. Walsh[1], Kunny Kou[2], John N. Marafino[1,2], Kristin McKenna[1], Kirstie Thompson[1], Tara M. Gallagher[2], Kyle Seifert[2], and Kevin L. Caran[1]
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121. **UNDERSTANDING A FUNDAMENTAL FORCE IN PROTEIN FOLDING: TUNING THE $n \rightarrow \pi^*$ INTERACTION VIA DESIGNED PEPTIDES**

Nicole Wenzell, Himal Ganguly, Glenn Yap and Neal Zondlo
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SYNTHESIS OF ARTIFICIAL BACTERIAL CELL WALL FRAGMENTS AS TOOLS TO STUDY THE INNATE IMMUNE SYSTEM

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Online access of this abstract is restricted at the request of the Principal Investigator.

MULTIMERIZATION OF SOLUTION-STATE PROTEINS BY WATER SOLUBLE PORPHYRINS

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Interactions between charged porphyrins and complimentary and oppositely charged proteins provide important models systems for studies of electron transfer processes, artificial photosynthesis, and control of protein-protein interactions. Typically, the experimental results are analyzed and discussed assuming that the proteins exist in a monodisperse state. However, this report will demonstrate that some proteins, positively and negatively charged, can multimerize in the presence of water-soluble porphyrins. Small angle X-ray scattering data shows that hen egg lysozyme, horse heart cytochrome *c,* PpcA, and cytochrome *c4* can multimerize in the presence of negatively and positively charged porphyrins. While most of the multimerization and binding occurred between oppositely charged proteins and porphyrins, some of the complimentarily charged proteins and porphyrins bound to each other despite having like charges, demonstrating that there may be another force that overcomes the electrostatic repulsion, such as a hydrophobic interaction. These results lay the groundwork for more experiments to show the reason for multimerization when attaching photosensitizer porphyrins to proteins, as there are more aspects to the attachment of porphyrins to proteins, heme containing cytochrome proteins and otherwise, than an assumption of monodispersity would allow, as there are mechanisms resulting in multimerization that are unknown at this point in time.

We gratefully acknowledge the VFP program through the Department of Energy for funding, Dr. David Tiede, Argonne National Lab, for support, and the staff of the APS and CNM at Argonne National Lab for instrument time.

COLLOIDAL AND BIOLOGICAL PROPERTIES OF M-E

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Bacteria are becoming resistant to current antibiotic drugs necessitating new therapeutic tactics to combat these resistant bacteria. Amphiphiles are molecules with both hydrophilic and hydrophobic regions. Some amphiphiles have antimicrobial properties. In this study we synthesized amphiphiles with a mesitylene core, three head groups and two hydrophobic tails. One head group is an N,N-dimethylethanolammonium group and the others are dimethylalkylammonium groups. The alkyl chains on the dimethylalkylammonium groups range in length from 8 to 16 carbon atoms. Compounds in this series are named *M-E,#,#*, where *M* represents the mesitylene core, *E* represents the ethanolammonium group and # represents the number of carbon atoms in each linear alkyl tail. Conductivity studies and isothermal titration calorimetry (ITC) studies were performed to determine colloidal properties, including critical aggregation concentration (CAC) and thermodynamics of aggregation. A plot of log(CAC) versus chain length shows a negative linear correlation as is typical for series of homologous amphiphiles. In addition, biological studies indicate the antibacterial activity of the amphiphile. The amphiphile with two linear 12 carbon chains was found to be the most potent antibacterial compound of the series. The pendent alcohol group on these molecules will allow for further functionalization toward developing more potent and specific antibacterial compounds.

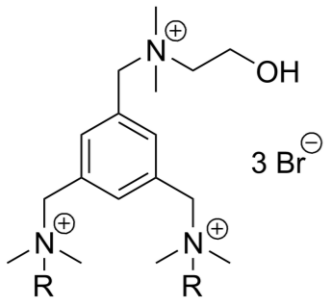

Figure 1. General structure of M-E,#,# amphiphiles. (R = C_8H_{17}, $C_{10}H_{21}$, $C_{12}H_{25}$, $C_{14}H_{29}$ or $C_{16}H_{33}$)

A FLEXIBLE APPROACH TO TREATING THE EBOLA VIRUS

Natalie Steenrod, Matthew Shirley, Therese Ku, and Katherine Seley-Radtke
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The Ebola virus is at the center of the world's stage due to the recent outbreaks in West Africa. Filoviruses such as Ebola are among the deadliest pathogens known, with fatality rates reaching near 90 percent. Despite dire need, there is no FDA-approved treatment or cure. Presently there are se

veral nucleoside analogues being investigated, including the carbocyclic nucleoside 3-deazaneplanocin A (3-deazaNpcA). The proposed mechanism of action for 3-deazaNpcA is the inhibition of S-adenosylhomocysteine hydrolase (SAHase). Inhibitors of this enzyme indirectly inhibit DNA methyltransferase through a biofeedback mechanism. This halts S-adenosylmethionine-dependent methylations of the 5'-cap of mRNA, leading to defective viral transcription and translation, inhibiting viral replication. For this project, the modified adenine base in 3-deazaNpcA will be "split" into its imidazole and pyridine components, remaining connected by a single C-C bond to give the target compound Flex-3-deazaNpcA. This will allow the base to adjust to form non-canonical binding interactions, without losing the integrity of the functional groups required for recognition, hence adopting an optimum conformation within the enzyme binding site. This strategy has been successful in multiple preliminary studies. Thus, endowing the 3-deaza scaffold with flexibility should prove strategic in terms of increased potency against Ebola.

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. This research was also supported by an Undergraduate Research Award provided through UMBC.

SYNTHESIS AND STUDY OF POLYCATIONIC AMPHIPHILES AS POTENT ANTISEPTICS AND NOVEL COLLOIDS: EXPLORING STRUCTURE ACTIVITY RELATIONSHIPS

Brenna J. C. Walsh[1], Kunny Kou[2], John N. Marafino[1,2], Kristin McKenna[1], Kirstie Thompson[1], Tara M. Gallagher[2], Kyle Seifert[2], and Kevin L. Caran[1]
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Several series of novel amphiphilic molecules have been designed and synthesized in an effort to develop novel and efficient antiseptics. Each of the new series include amphiphiles with non-conventional structures; molecular architectures include compounds with multiple hydrophilic head groups and one or two hydrophobic tails. In addition to these amphiphiles, we will also present progress on current research of amphiphiles with a hexasubstituted hydrophilic head group. The synthesis, purification, and preliminary analyses, both colloidal and antibacterial, of these amphiphiles will be presented. Relationships between amphiphile structure, colloidal properties, and biological properties will be discussed. We would like to thank the National Science Foundation and the Research Corporation for the Advancement of Science for their funding and support of our research.

UNDERSTANDING A FUNDAMENTAL FORCE IN PROTEIN FOLDING: TUNING THE $n \rightarrow \pi^*$ INTERACTION VIA DESIGNED PEPTIDES

Nicole Wenzell, Himal Ganguly, Glenn Yap and Neal Zondlo
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An $n \rightarrow \pi^*$ interaction is a local interaction that orders two consecutive residues by delocalizing the lone pair of electrons from oxygen of the donor carbonyl to the antibonding orbital of the acceptor carbonyl carbon. This interaction is a recent discovery that gives insight into the inherent biases that proteins have to drive them to their folded state. By changing the electronics of the donor carbonyl on model proline-based peptides, we can tune the strength of this $n \rightarrow \pi^*$ interaction.

Proline can readily populate both the *trans* and *cis* conformations in a peptide bond. The $n \rightarrow \pi^*$ interaction is shown to stabilize the *trans* conformation. Thus, in our model system the equilibrium constant of *trans-cis* isomerization ($K_{trans/cis}$) acts as a readout for the strength of $n \rightarrow \pi^*$ interaction; greater electron donation causes a stronger interaction, resulting in higher $K_{trans/cis}$ values. The substitution of the electron-withdrawing monochloroacetyl for acetyl resulted in a 0.2 kcal/mol weaker $n \rightarrow \pi^*$ interaction.

Further evidence of the electron donation involved in the $n \rightarrow \pi^*$ interaction is seen via the chemical shifts of the ^{13}C NMR. The chemical shift of the acceptor carbonyl moves downfield with stronger electron-donating substituents; carbonyls uninvolved in the interaction remain constant. The $n \rightarrow \pi^*$ interaction is also indicated through the distance and angles determined through X-ray crystallography of model peptides. The more electron donating character of the acetyl compared to the formyl group results in a 0.3Å shorter distance (3.3-3.0Å) and an angle (115˚vs 90˚) that more closely resembles an ideal nucleophilic attack (109.5˚).

The electronics of the donor carbonyl in the model peptides were modulated by synthesizing derivatives with varying electron-donating capabilities on the donor carbonyl. The comparison of the $K_{trans/cis}$ values, the carbonyl chemical shifts, and the angles and distances of the donor-acceptor carbonyl pair show that more electron-donating character leads to a stronger $n \rightarrow \pi^*$ interaction.

Morning Poster Session
Group U – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

122. **SYNTHESIS OF AU-PCL NANOPARTICLES**

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123. **UTILIZING CHLOROFORM POST-TREATMENT TO IMPROVE THE ADHESION OF AU THIN FILMS ONTO PMMA**

Kathleen T. Krist[1], Harry Hu[1], Brian H. Augustine[2], and Wm. Christopher Hughes[3]
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124. **EXFOLIATION OF BISMUTH TELLURIDE**

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[2]Department of Chemistry, Howard University, 525 College Street NW, Washington DC 20059

125. **SYNTHESIS AND CHARACTERIZATION OF PALLADIUM NANOPARTICLE DOPED 3D GRAPHENE NANOSHEETS FOR USE AS ELECTROCATALYST SUPPORTS IN FUEL CELLS**

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[2]Department of Chemical and Biological Engineering, University of New Mexico, Albuquerque, NM 87131

126. **EFFECT OF DOPANT CONCENTRATION ON SOLAR CELL PERFORMANCE**

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127. **TEMPLATE-DIRECTED CRYSTALLIZATION OF DIPHENYL UREA**

Serena Seshadri, Marina Solomos, and Jennifer Swift
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SYNTHESIS OF AU-PCL NANOPARTICLES

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Oleylamine coated gold nanoparticles (Au-OA) have been synthesized, thiolated with mercaptoundecanol (Au-MUD), and then polymerized with polycaprolactone (Au-PCL), which will allow the nanoparticles to form micelles. The goal of this research is to synthesize gold polycaprolactone (Au-PCL) nanoparticles, which form micelles to be used in a targeted drug delivery system. The system would provide healthier and more effective treatment, as it specifically targets malignant cells, leaving healthy cells unharmed. The gold nanoparticles build up in tumor sites due to their enhanced permeability and retention (EPR) effects and have the ability to convert light energy into heat, providing the potential for a drug to be released into the body using a laser. The drug could be placed in the gold micelle and delivered to the tumor site for more efficient treatment of cancer.

UTILIZING CHLOROFORM POST-TREATMENT TO IMPROVE THE ADHESION OF AU THIN FILMS ONTO PMMA

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The metallization of Au onto plastics is an important processing step in applications such as the aerospace and automotive industries, the field of microelectronics, and the fabrication of microfluidic devices. While its corrosion resistance and excellent electrical and thermal conductivity make Au a useful choice, its inertness results in poor adhesion to polymer surfaces. Previous studies have indicated that exposing commercially available poly(methyl methacrylate) (PMMA) sheets to chloroform vapor following Au deposition significantly improves adhesion. In this study, we utilized electron-beam evaporation to deposit 6 nm of Au onto 1.50 mm thick PMMA and exposed the samples to vapor released from chloroform heated on a hot plate set at 70°C. The force required to remove the Au thin films was determined by placing samples on a polisher spinning at 150 rpm and utilizing UV-VIS spectroscopy to measure the transmittance of 700 nm light through the films to quantify their removal as a function of applied polishing force. The Au thin films were also characterized using atomic force microscopy (AFM). AFM images demonstrated a progressive roughening of the surface corresponding to an increase in applied force. Additionally, these images support a model in which the chloroform treatment softens the PMMA surface, producing a softened layer that the polisher removes simultaneously with the Au thin film. Following the attainment of quantitative data demonstrating the effectiveness of chloroform post-treatment, the vapor exposure procedure was applied to selectively pattern a series of PMMA samples.

This research was supported by the JMU Center for Materials Science and the National Science Foundation (NSF-RUI DMR #1305808).

EXFOLIATION OF BISMUTH TELLURIDE

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Bismuth Telluride (Bi_2Te_3) is a good candidate for thermoelectric applications at room temperature as it has low thermal conductivity and high electrical conductivity resulting in a high thermoelectricity figure of merit (ZT). We are interested in developing a synthesis protocol for bismuth telluride nanomaterials and measure the properties of the synthesized nanomaterials.

In this poster, we present the progress that we have made towards synthesis of bismuth telluride by Chemical exfoliation using n-butyl lithium. The exfoliated bismuth telluride was then deposited on silicon wafers and then analyzed using Scanning Electron Microscope, Optical Microscope, Energy Dispersive X-ray Spectroscopy (EDS), and Raman Microscope.

In the future, we plan to use other techniques including Atomic force microscopy and profilometer to determine the thickness of deposited materials. We also plan to use MatLab to analyze the density of the deposited material.

This work was supported by grants from the National Science Foundation (NSF DMR 1231319 and NSF #1205608). We thank Howard University, Howard Nanofabrication Facility (HNF), Howard University Chemistry Department, Gallaudet University, and Dr. Tito Huber's Research Laser Facility for their support.

SYNTHESIS AND CHARACTERIZATION OF PALLADIUM NANOPARTICLE DOPED 3D GRAPHENE NANOSHEETS FOR USE AS ELECTROCATALYST SUPPORTS IN FUEL CELLS

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The depletion of fossil fuels along with the increasing demand for energy has given rise to the development of sustainable energy technologies such as fuel cells. Fuel cells require catalysts in order to have the reaction move forward and catalysts require supports at both the cathode and anode of the fuel cell. Due to its high stability, surface area, and electrical conductivity we will consider using 3D graphene nanosheets (GNS) as our support. Using the Sacrificial Support Method (SSM) we were able to control the morphology of the nanosheets. This was done by altering three factors: the weight ratio of SiO_2 to GNS, the Si based sacrificial compound used, and the reduction method for turning graphene oxide into GNS. After changing these factors for different samples, Palladium (Pd) nanoparticles were doped on the surface of each sample. Energy dispersive spectroscopy (EDS) was used to confirm that all the graphene oxide was reduced and that no Si based compound remained. After looking at the Brunauer-Emmett-Teller (BET) analysis of each sample it was shown that chemical reduction method with the SiO_2 sacrificial support yielded the highest surface area. Scanning Electron Microscope (SEM) images show multiple anchoring sites for the catalysts on the surface of all samples besides the thermally reduced tetraethyl othrosilicate (TeOS) samples which had a two dimensional morphology. Raman spectroscopy showed that the chemically reduced SiO_2 sample had the most defects and that all samples had multilayers of GNS. A final characterization test involving cyclic voltammetry was done to test the electrochemical activity of the samples. After all the characterization tests it was concluded that the highest surface area and greatest amount of defects does not necessarily translate to the best electrocatalyst support. The thermally reduced GNS sample had higher electrochemical activity than chemically reduced and was the best support.

This investigation was supported by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences.

EFFECT OF DOPANT CONCENTRATION ON SOLAR CELL PERFORMANCE

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Quantum dot sensitized solar cells show promise in improved efficiency of these devices. Most of the research has focused on cadmium selenide nanocrystals as the quantum dots, but these two elements are of concern from environmental and toxicological standpoints. Zinc sulfide nanocrystals doped with manganese could serve as possible replacements for cadmium selenide nanocrystals. The main goal of this research was to determine what concentration of manganese dopant will make a solar cell more energy efficient in the conversion step.

To help accomplish this goal, doped zinc sulfide nanocrystals were prepared by reacting aqueous solutions of zinc acetate, which contained a selected percentage of manganese acetate ranging from 0.1% to 2%, with aqueous solutions of sodium sulfide. Polyvinylpyrrolidone (PVP) was added to the zinc solution as a capping agent. Particle sizes were determined to be around 4 nm using UV/Vis absorption spectroscopy, and the emission properties were studied using fluorescence spectroscopy. The quantum yield of the doped nanocrystals will be determined by comparing results with those from the fluorescence standard quinine.

These doped nanocrystals will be used in the preparation of solar cells consisting of a graphite coated electrode and a quantum dot sensitized titanium dioxide coated electrode, with a postassium triiodide in ethylene glycol electrolyte. In the initial phase of this part of the project, improvements were made in the fabrication of the titanium dioxide film. A paste was created by combining TiO_2, acetic acid, and a surfactant, and this paste was then applied to the conducting side of a piece of fluorine doped tin oxide coated glass. An initial solar cell was assembled using raspberry dye as the sensitizer and voltages were measured to test device performance. Future work will involve the fabrication of solar cells using manganese doped nanocrystals.

Acknowledgment is made to the Donors of the Lebanon Valley College Chemistry Endowed Research Fund for support of this research.

TEMPLATE-DIRECTED CRYSTALLIZATION OF DIPHENYL UREA

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Many 1,3-bis(X-phenyl)ureas crystallize as concomitant mixtures of polymorphs. In previous work, we showed it is possible to control polymorphism by heterogeneous nucleation on glass-siloxane monolayer templates. The present study focuses on 1,3-bisphenylurea (PU), which crystallizes from various solvents as a concomitant mixture of α and β polymorphs. Slow evaporation growth in the presence of select siloxane templates and solvents yields rectangular α-PU crystals arranged in unusual circular patterns approx.150-400 μm in diameter alongside or independent of long α-PU needles and small, angular β-PU crystals. The ongoing use of Raman spectroscopy to study the polymorphic phase of the individual crystals within the circles has indicated that α-PU dominates the circular patterning. Fast evaporation drop-cast crystallization of PU onto the same siloxanes yields a mixture of α- and β-PU fibers with similar curvature indicative of a preliminary bending pattern to the circles produced through slow evaporation. While α-PU is the dominant polymorph yielded from slow evaporation, β-PU is primarily observed through drop-cast crystallization. These results suggest a growth model in which flexible 1D urea fibers are kinetically stabilized by the surface but fracture as stresses develop during their subsequent growth to macroscopic sizes.

We are grateful for the support provided by the National Science Foundation (CHE-0959946, CHE-1337975, and CHE-1428079), the Clare Boothe Luce Foundation, and the Georgetown undergraduate GUROP program.

Morning Poster Session
Group V – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

128. **eSpaneX: A NEW TECHNIQUE FOR HIGH QUALITY EXFOLIATION OF GRAPHENE**

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129. **SYNTHESIS OF IMMOBILIZED PALLADIUM NANOCATALYSTS**

Alexandria Lehman, Kristen Kelsall, and Anderson Marsh
Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

130. **ULTRAFAST LIMITS OF PHOTO-INDUCED ELECTRON TRANSFER RATES IN PpcA, A MULTI-HEME C-TYPE CYTOCHROME**

Aidan M. McKenzie, Daniel R. Marzolf, Matthew C. O'Malley, and Oleksandr Kokhan
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131. **THE SYNTHESIS OF COBALT TETRAPYRROLE MACROCYCLES AND ACTIVITY FOR DIOXYGEN REDUCTION TO WATER OR HYDROGEN PEROXIDE**

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132. **DESIGNING FUNCTIONAL MODELS OF THE MCCA SULFITE REDUCTASE IN MYOGLOBIN AND CYTOCHROME *C* PEROXIDASE**

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eSpaneX: A NEW TECHNIQUE FOR HIGH QUALITY EXFOLIATION OF GRAPHENE

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Micromechanical cleavage or scotch-tape method is one of the most used exfoliation method to produces monolayer graphene but the presence of tape residue on the exfoliated layers will degrade its quality. In this research a new tool for exfoliation, known as eSpaneX, which enables a clean contact between the crystal and the substrate is investigated. The tool enables clean and tapeless exfoliation of graphene layers. Mainly using the press and shear mode, a suitable method for the exfoliation of the crystal was developed. Analysis of the exfoliated layers shows the deposition of thin graphite layers.

Massachusetts Institute of Technology (MIT)
Department of Physics; Pablo Jarillo-Herrero
Harvard School of Engineering and Applied Science (SEAS)
National Science Foundation (NSF) Grant #: NSF DMR-1231319
Center for Integrated Quantum Materials (CIQM)

SYNTHESIS OF IMMOBILIZED PALLADIUM NANOCATALYSTS

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One of the goals of catalysis in the 21st century is to develop materials that exhibit high activity and selectivity during reactions. These two catalytic actions may be manipulated by tuning catalyst properties such as particle size, particle shape, and catalyst support. The tools of nanotechnology may be utilized to control such properties to design materials that display specific functionalities.

To this end, we have synthesized colloidal palladium nanoparticles using solution based reduction methods. Alcohol reduction and seeded growth methods were used to obtain various nanoparticle sizes, with polyvinylpyrrolidone (PVP) serving as a capping agent to control growth and to stabilize the nanoparticles. For comparison, uncapped nanoparticles were also were synthesized by using palladium acetate and sodium borohydride. Particle sizes of as-prepared nanomaterials were determined from transmission electron microscopy (TEM) measurements. To examine the effect of supporting the nanoparticles on catalytic activity and selectivity, immobilized nanocatalysts were prepared by adsorbing nanoparticles from solution onto silica microspheres. The loading of palladium nanoparticles on the microspheres was verified by TEM analysis and was measured using flame atomic absorption spectroscopy (FAAS). Results from FAAS experiments for the immobilized nanocatalysts were compared to those from a commercial 5% palladium on silica catalyst.

The catalytic materials were subsequently used in the aqueous phase hydrogenation of phenol. By studying the effects of reaction conditions (temperature and reactant concentrations), as well as nanocatalyst properties (particle size, PVP capping agent, and support), we hope to ascertain the molecular-level ingredients necessary to design an environmentally-friendly catalyst.

Acknowledgment is made to the Donors of the American Chemical Society Petroleum Research Fund for support of this research.

ULTRAFAST LIMITS OF PHOTO-INDUCED ELECTRON TRANSFER RATES IN PpcA, A MULTI-HEME C-TYPE CYTOCHROME

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Conversion of light energy to electrochemical equivalents is at the core of any photosynthetic system. In our attempts to build enzymes for artificial photosynthesis, we need to have complete control of all charge transfer rates, including the initial charge separation step. Here we demonstrate an unexpectedly large range ($5x10^7$ to $5x10^{11}$ s^{-1}) of the observed photo-induced electron transfer rates between PpcA, a 3-heme member of cytochrome c_7 family, and $Ru(bpy)_3$, an artificial photosensitizer covalently-linked in close proximity to PpcA hemes. HPLC-MS characterization confirmed purity of our isolated protein and its correct assembly as well as stoichiometric binding of photosensitizers. Combined small- and wide-angle X-ray scattering studies performed at the Advanced Photon Source of Argonne National Laboratory demonstrated the absence of any significant structural changes from the initial compact globular form in mutated and covalently-labeled protein forms. All-atom molecular dynamics simulations provide further insight into protein structure and dynamics responsible for the large range of electron transfer rates. These results demonstrate the limits of tuning charge transfer rates and offer opportunities to use inexpensive photosensitizers based on abundant elements.

We gratefully acknowledge the Visiting Faculty Program through the Department of Energy for funding, Dr. David Tiede and Argonne National Laboratory for support, and the staff of the Advanced Photon Source and the Center for Nanoscale Materials at Argonne National Laboratory for instrument time and help with data collection.

THE SYNTHESIS OF COBALT TETRAPYRROLE MACROCYCLES AND ACTIVITY FOR DIOXYGEN REDUCTION TO WATER OR HYDROGEN PEROXIDE

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Online access of this abstract is restricted at the request of the Principal Investigator.

DESIGNING FUNCTIONAL MODELS OF THE MCCA SULFITE REDUCTASE IN MYOGLOBIN AND CYTOCHROME *C* PEROXIDASE

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Sulfite reductases (SiR) are essential enzymes for sulfur metabolism. SiRs catalyze the six-electron transfer process that reduces sulfite to sulfide in a single active site that contains a siroheme coupled to an iron-sulfur cluster, a cofactor pair unique to these enzymes. Recently, a new class of Cu(I)-dependent SiR has been identified that lacks an iron-sulfur cluster and substitutes heme *c* for siroheme at the active site: the octaheme, trimeric protein MccA. The MccA SiRs display greater activity and substrate specificity than the siroheme SiRs. In order to understand the factors influencing activity and specificity in SiRs, we have engineered a Cu(I) binding site into well-understood hemoproteins, myoglobin and cytochrome *c* peroxidase, to create a functional model of MccA. The mutant proteins were designed and simulated computationally, then expressed and analyzed for Cu (I) binding.

I would like to give a very special thanks to Dr. Yi Lu and Evan Mirts for allowing me to work in the lab this summer and learn from them. I also wish to thank the National Science Foundation (NSF) and the Howard Hughes Medical Institute's Exceptional Summer Research Opportunities Program (HHMI EXROP) for funding my experience at the University of Illinois Urbana-Champaign and for all of their support.

Afternoon Poster Session
Group W – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

133. **INVESTIGATING THE TRANSCRIPTIONAL RESPONSE OF CANDIDATE PROTEINS TO AMPHOTERICIN B**

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134. **PROTEIN-RNA INTERACTIONS IN BUNYAVIRIDAE GENOME PACKAGING**

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Department of Biology, Morgan State University, 1700 E Cold Spring Lane,
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135. **PHYSICAL INTERACTION OF T-CADHERIN AND ADIPONECTIN CONFIRMS THE NOVEL ROLE OF T-CADHERIN AS A RECEPTOR**

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136. **SOX-17 CAUSES CHANGES IN EXTRACELLULAR MATRIX AND GAP JUNCTION PROTEINS**

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137. **IDENTIFYING RESIDUES ON DNAK WHICH ARE IMPORTANT FOR CO-CHAPERONE COLLABORATION**

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138. LIMITED-PROTEOLYSIS OF THE *E. COLI* GMP SYNTHATASE ATPP DOMAIN REVEALS A COMPACT DIMER CORE

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139. SYNTHESIS OF ENAMINONES AS EFFECTIVE GluR2 ANTAGONISTS

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INVESTIGATING THE TRANSCRIPTIONAL RESPONSE OF CANDIDATE PROTEINS TO AMPHOTERICIN B

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Currently, more than 30 incurable human diseases are caused by dysfunctional or missing ion channel proteins. We have demonstrated that ion channel forming small molecules, such as Amphotericin B (AmB), can imperfectly replicate the function of Trk1 and Trk2 potassium ion transporters in yeast cells, thereby acting as prostheses on the molecular scale. We hypothesize that AmB productively interfaces with networks of collaborating proteins to restore cell growth and physiology in protein deficient cells.

Several candidate proteins have been proposed to be of importance to this process of AmB-mediated rescue. We will investigate the relative changes in gene expression of these protein ion transporters in wild type versus potassium transporter deficient (trk1Δtrk2Δ) cells rescued by AmB via RT-qPCR. Based on preliminary evidence we expect to see upregulation of some candidate genes in trk1Δtrk2Δ+AmB cells compared to that in wild type. For future studies, we will further investigate expression and activity levels of candidate collaborating proteins. Gaining a better mechanistic understanding of how imperfect small molecule ion channels can restore cell growth may have significant therapeutic implications.

This project was aided by the Burke group, the Summer Research Opportunities Program, Biochemistry department, and School of Molecular and Cellular Biology at Illinois, as well as the Howard Hughes Medical Institute. In addition, this investigation was supported in part by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences.

PROTEIN-RNA INTERACTIONS IN BUNYAVIRIDAE GENOME PACKAGING

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Online access of this abstract is restricted at the request of the Principal Investigator.

PHYSICAL INTERACTION OF T-CADHERIN AND ADIPONECTIN CONFIRMS THE NOVEL ROLE OF T-CADHERIN AS A RECEPTOR

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Adiponectin (APN), a multimeric adipocyte-derived hormone encoded by ADIPOQ gene, has been shown to play an important role in protecting the vasculature from the detrimental effects of ischemia and reperfusion injury. Detailed information about the interaction of adiponectin with T-cadherin, a proposed receptor of adiponectin on cardiovascular tissues, is essential for the understanding of adiponectin effect on the injured tissues. Here we were able to produce and purify comparable amount of adiponectin and T-cadherin for structural study of the interaction surface between the two proteins. Our pull-down assay confirmed the direct physical interaction between adiponectin and T-cadherin, suggesting that T-cadherin may serve as a potential mediator for adiponectin activity on vasculature.

SOX-17 CAUSES CHANGES IN EXTRACELLULAR MATRIX AND GAP JUNCTION PROTEINS

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Online access of this abstract is restricted at the request of the Principal Investigator.

IDENTIFYING RESIDUES ON DNAK WHICH ARE IMPORTANT FOR CO-CHAPERONE COLLABORATION

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Hsp70 is an essential ATP-dependent molecular chaperone present in all organisms. DnaK, the *Escherichia coli* homolog functions in the folding, unfolding, remodeling, and activation of various client proteins. DnaK requires the activity of two co-chaperones: DnaJ, which stimulates the rate of ATP-hydrolysis by DnaK, and GrpE, a nucleotide exchange factor. *E. coli* DnaK is also known to collaborate with additional ATP-dependent chaperones, including *E. coli* Hsp90 and ClpB. To identify residues of *E. coli* DnaK that are important for collaboration with other chaperones and co-chaperones, we made amino acid substitutions in regions of interest based on structural analysis. Wild type DnaK and a DnaK mutant, DnaK D129A/Y130A, were overexpressed, purified and tested in vitro for ATPase activity and the ability to remodel substrate in collaboration with DnaJ and GrpE. Our results show that DnaK D129A/Y130A hydrolyzed ATP at a similar rate to wild type DnaK. Thus the mutant protein has wild type ATPase activity. In heat-denatured GFP reactivation assays, DnaK D129A/Y130A was defective in substrate remodeling compared to wild type DnaK. This suggests that the region of DnaK including D129A/Y130A may be important for chaperone activity of DnaK. The cause of the defect remains to be determined, but the defect is consistent with a defect in interaction with GrpE.

This research was supported by the Intramural Research Program of the NIH, NCI, Center for Cancer Research.

LIMITED-PROTEOLYSIS OF THE *E. COLI* GMP SYNTHATASE ATPP DOMAIN REVEALS A COMPACT DIMER CORE

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Online access of this abstract is restricted at the request of the Principal Investigator.

SYNTHESIS OF ENAMINONES AS EFFECTIVE GluR2 ANTAGONISTS

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Previous research by K.R. Scott has found that enaminones have the ability to be an effective GluR2 antagonist and anti-seizure medicine. Recent understanding of the detrimental effects of traumatic brain injury (TBI) has been linked to a dramatic increase in glutamate receptor (GluR2) production and a decrease in the inhibitory GABAergic proteins of excitatory interneurons. The overproduction of GluR2 increases the likelihood of uncontrolled interneuronal excitation resulting in a seizure. The controlled inhibition of the GluR2 protein can limit seizure production and the effects of TBI. This research synthesized a series of enaminones in high yields as GluR2 antagonist for the treatment of TBI. The enaminones were structurally determined using GCMS, ^{1}H and ^{13}C NMR, X-Ray crystallography and tested using a series of computational docking methods including IGemDock and Pyrx - Autodock Vina compared to a small subset of molecules that showed improved docking interaction. Future studies will incorporate these molecules into animal studies for their biological activity.

This Project was supported by The College of Natural Science and Mathematics at The Richard Stockton College of New Jersey.

Afternoon Poster Session
Group X – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

140. **STABILIZATION OF THE HIV-1 RNA GENOME 5'-UNTRANSLATED REGION (5'-UTR) MONOMER CONFORMER IN SODIUM ACETATE BUFFER**

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141. **DETERMINATION OFCUMULATIVE EFFECTS OF BACTERICIDAL AND BACTERIOSTATIC AGENTS ON *B. subtilis* AND *E. coli***

Lamarque Coke[1] ,Andrew Lippman[1], Phuong Le[1], Joulian Wilmer[1],
Ellis Benjamin[1], and Earl Benjamin[1]
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142. **MATHEMATICAL MODELING OF GLIOBLASTOMA BRAIN CANCER CELL MOTILITY AND PROLIFERATION IN 2-D CULTURE**

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143. **CHARACTERIZATION OF THE UFM1 INTERACTING MOTIF IN UBA5**

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144. **CHARACTERIZING THE EFFECTS OF PHOSPORYLATION ON THE STRUCTURAL DYNAMICS OF UBIQUITIN**

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145. **NON-IMAGE FORMING VISUAL PIGMENTS: DO THEY INTERNALIZE?**

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146. **THE EFFECT OF NEIGHBORING RESIDUES ON PEROXIDASE ACTIVITIES OF SELENOPROTEINS**

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STABILIZATION OF THE HIV-1 RNA GENOME 5'-UNTRANSLATED REGION (5'-UTR) MONOMER CONFORMER IN SODIUM ACETATE BUFFER

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The HIV/AIDS pandemic is a major health issue, with 35 million people infected worldwide and 1.5 million HIV-related deaths in 2013 alone. Human Immunodeficiency Virus (HIV) proliferates exhaustively via the viral life cycle by infecting helper T-cells in the human body. This life cycle consists of an early and a late phase. In the early phase, an HIV virion infects helper T-cells and integrates reverse-transcribed pro-viral DNA into the cell's nucleus to hijack cellular replication machinery. The late phase involves proliferation of new viral particles from the cell in which transcription of viral genomic material and translation of viral proteins are regulated by the 5'-UTR of the viral RNA. This leader RNA exists in a monomer-dimer equilibrium bridged by a structural switch. Dimerized unspliced viral RNA is packaged as new genomic material while the monomer conformer is proposed to promote translation of viral proteins, including Gag and Gag-Pol. We seek to characterize the structure of the 5'-UTR monomer conformation – this leader RNA is a highly conserved region of the HIV-1 genome and presents as an appealing drug target in clinical medicine.

The existence of a monomer-dimer equilibrium is a major obstacle in characterization of the three-dimensional monomer structure by Nuclear Magnetic Resonance (NMR) spectroscopy. At NMR concentrations, the monomer-dimer equilibrium predominantly favors the dimer conformer. 5'-UTR RNA constructs synthesized were assessed using a variety of condition-dependent electrophoretic mobility shift assays (EMSAs) to characterize the stability of the monomer in buffering environments of variable pH. A number of constructs have shown significant increases in monomer stability when in a 10 mM sodium acetate buffer, pH 4; results were quantified using ImageJ. Future efforts include the comparison of the 5'-UTR at low pH against a monomer control at biologically relevant pH using one-dimensional NMR spectroscopy to characterize structural relevance.

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DETERMINATION OFCUMULATIVE EFFECTS OF BACTERICIDAL AND BACTERIOSTATIC AGENTS ON *B. subtilis* AND *E. coli*

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Antibiotic resistance is a problem that has far reaching implications. The exponential growth of antibiotic resistance is emerging faster than new antibiotics can be discovered. Although there is ongoing research for new antibiotics, current treatments use high doses of last line antimicrobials for resistant infections. Ultimately, the target treatment should be achievement of a good clinical outcome with the least toxicity. It is generally believed that bactericidal and bacteriostatic drugs should not be combined in vivo. This study used disc diffusion assays with a series of well-established bactericidal and bacteriostatic broad spectrum antibiotics, Ampicillin (bactericidal), Chloramphenicol (bacteriostatic), Streptomycin (bactericidal), and Tetracycline(bacteriostatic), individually as well as in combination to develop models for antibiotic effectiveness on both gram positive (B. subtilis) and gram negative (E. coli) bacteria. Each effect was classified as either synergistic, additive, no effect, or antagonistic. The results found were the higher concentration of ampicillin, the higher effect of it on both E.coli and B.subtilis. Vary concentration of ampicillin does not change the effect of chloramphenicol on E.coli but reduce the effect of chloramphenicol on B.subtilis. Vary concentration of ampicillin have additive effect on the effective of tetracycline on both E.coli and B.subtilis. Vary concentration of ampicillin have no effect on the effective of streptomycin on E.coli, but have additive effect on the effective of streptomycin on B.subtilis.

MATHEMATICAL MODELING OF GLIOBLASTOMA BRAIN CANCER CELL MOTILITY AND PROLIFERATION IN 2-D CULTURE

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Glioblastoma multiforme (GBM), or high-grade glioma, is a devastating disease for which there is no cure. Its malignancy stems from a combination of cell division along with high local invasiveness within the brain. Various mathematical models have been developed to help understand the growth and behavior of GBM cells. However, many models either are extremely complicated, look at multiple interdependent phenomena, and/or use modeling software not readily available to the public. Here, we wished to develop a more simple, but useful, mathematical model to describe the motility and proliferation of GBM cells in a cell culture environment that are autocrine/paracrine stimulated by the molecule L1CAM through two different cell surface receptor systems which share a common signal transduction pathway. We chose to use an agent-based model and the publicly available NetLogo software. Here, we describe the model, its accuracy compared to experimental motility and proliferation data, and its potential usefulness for predicting future experimental outcomes. We conclude that this simple model accurately reflects much of the GBM cell motility and proliferation behavior observed *in vitro*. Furthermore, our model can be modified easily to suit the needs of cancer investigators interested in other intrinsic or extrinsic cell signaling stimuli that influence cancer cell behavior *in vitro*.

CHARACTERIZATION OF THE UFM1 INTERACTING MOTIF IN UBA5

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The UFM-ylation pathway modifies a substrate by addition of the signaling molecule Ufm1 (ubiquitin-fold modifier 1). The human ubiquitin-activating enzyme Uba5 catalyzes the first steps of this pathway that include adenylation, formation of a thioester bond between itself and Ufm1, and transfer of the thioester bond from itself to Ufc1 (the E2 in this pathway). These proteins have been shown to be upregulated in metastatic cancers and in apoptosis, but the reason why is currently unknown. Another problem is that the ubiquitin fold domain (UFD) of Uba5 prevents crystallization, so the structure of that set of residues within the full protein is unknown. In this study we investigate the structural changes that occur to Ufm1 as a result of interactions between the Uba5 UFD and Ufm1 using nuclear magnetic resonance and modeling software so that we can understand in greater detail the full structure of Uba5 and how this process is related to cancer and apoptosis. Chemical shifts were visible in the two NMR experiments in which Uba5 (residues 314-363) was added to Ufm1. There were a few residues on Ufm1 that had conserved shifts between the two experiments, which implies that this may be the area where Uba5 binds. These data suggest we have identified a new and novel Ufm1 interacting motif.

CHARACTERIZING THE EFFECTS OF PHOSPORYLATION ON THE STRUCTURAL DYNAMICS OF UBIQUITIN

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Online access of this abstract is restricted at the request of the Principal Investigator.

NON-IMAGE FORMING VISUAL PIGMENTS: DO THEY INTERNALIZE?

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G protein-coupled receptors (GPCRs), the largest family of eukaryote transmembrane receptors, respond to extracellular stimuli and trigger a response within the cell. Opsins are specialized GPCRs that are involved in the conversion of light into a biological signal. Melanopsin is an opsin that is found in intrinsically photosensitive retinal ganglion cells (ipRGCs) in the mammalian retina and regulates non-image forming functions such as circadian photoentrainment and pupillary constriction. While much of the phototransduction pathway of melanopsin remains unknown, we hypothesize that melanopsin is internalized after activation by light and deactivation by β-arrestin. To test this hypothesis, we synthesized GFP-tagged melanopsin constructs in a mammalian expression vector, PMT3, through cassette mutagenesis and expressed these constructs in Human Embryonic Kidney (HEK293) cells. We then compared localization of melanopsin within the cell through confocal microscopy. These results demonstrate that melanopsin is internalized in a heterologous expression system.

This work was funded by NSF DBI-1031420.

THE EFFECT OF NEIGHBORING RESIDUES ON PEROXIDASE ACTIVITIES OF SELENOPROTEINS

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Afternoon Poster Session
Group Y – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

147. **EN'CAS'ING THE STRESS: ENGINEERING A HUMAN CELL LINE KNOCKOUT OF HEAT SHOCK RESPONSE COORDINATOR GENES USING CRISPR CAS9 SYSTEM**

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148. **METABOLIC RESPONSES TO A HIGH-FRUCTOSE DIET IN DUAL KNOCKOUT OF THE INSULIN AND INSULIN-LIKE-GROWTH RECEPTORS (IGF) FROM THE PROXIMAL TUBULE OF MICE**

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149. **A FRAMEWORK FOR THE BEHAVIOR OF RARE VARIANT TESTS IN THE PRESENCE OF LARGE NUMBERS OF VARIANTS**

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150. **MECHANISM OF ACTION OF SCHWEINFURTHINS, A NEW CLASS OF ANTICANCER COMPOUNDS**

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151. **EFFECT OF LYSOPHOSPHATIDIC ACID ON E-CADHERIN LOCALIZATION IN MIGRATING METASTATIC BREAST CANCER CELLS**

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152. ADIPOCYTE mTORC2 KNOCKOUT DISRUPTS WHOLE-BODY GLUCOSE AND LIPID METABOLISM AND IMPAIRS THE BEIGEING OF WHITE ADIPOSE

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153. PURIFICATION AND ANALYSIS OF HUMAN LIVER FATTY ACID BINDING PROTEIN (FABP1).

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EN'CAS'ING THE STRESS: ENGINEERING A HUMAN CELL LINE KNOCKOUT OF HEAT SHOCK RESPONSE COORDINATOR GENES USING CRISPR CAS9 SYSTEM

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The Heat Shock Response (HSR) is an evolutionarily conserved response to high temperatures and other stresses that controls adaptive proteostasis, and is primarily regulated by the factor, Heat shock transcription factor 1 (HSF1). In mammalian cells, HSF1 is converted from an inactive monomeric form to an active trimer in response to heat stress. A ribonucleoprotein complex comprising of eukaryotic translation elongation factor eEF1A1, and a long noncoding RNA HSR1 are the key components of HSF1 activation. Once activated, HSF1 is recruited to Heat Shock Protein (HSP) promoter regions, upregulating chaperone activity in the cell. Along with mediating initiation of HSR, eEF1A1 is also a vital component of protein synthesis machinery. Interestingly, another isoform of eEF1A, called eEF1A2, is expressed in some specialized terminally differentiated cells of skeletal muscle, heart, pancreatic islets and motor neurons, all of which are prone to protein aggregation. The two isoforms are 92% identical and are reciprocally regulated. To better understand the role of eEF1A1 and HSF1 proteins in humans, we use a CRISPR-Cas9 nickase system to knockout HSF1 and eEF1A1 in a human cell line. We showed that the hTERT-immortalized, normal diploid foreskin fibroblast cell line, BJ-5ta, produces both eEF1A isoforms. This will allow us to perform eEF1A1 knockout in these cells. We hypothesize that HSF1 knockout cell line will survive under normal conditions but express very low thermotolerance. Conjointly, we hypothesize that the elimination of eEF1A1 may be compensated by the upregulation of eEF1A2. If viable, the eEF1A1 knockout cell line will be used for screening mutants of eEF1A2 restoring activation of HSR. Both HSF1 and eEF1A1 knockout lines will also be used for future studies to improve upon the current model of the HSR pathway and potentially reveal therapeutic targets for diseases like ALS, Alzheimer's disease, Parkinson's disease, type 2 diabetes, and amyloidosis.

This investigation was supported in part by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences. 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

METABOLIC RESPONSES TO A HIGH-FRUCTOSE DIET IN DUAL KNOCKOUT OF THE INSULIN AND INSULIN-LIKE-GROWTH RECEPTORS (IGF) FROM THE PROXIMAL TUBULE OF MICE

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Fructose is a naturally occurring monosaccharide found in fruits, table sugar and some vegetables. High consumption of fructose has been linked to diabetes, insulin resistance, obesity, increased blood pressure, increased adipose tissue, cardiovascular disease, and metabolic syndrome. Most of the absorbed fructose is metabolized by the liver with the remaining being absorbed by the kidney and adipose tissue. Fructose is able to enter the cell through the glucose transporters, GLUT5 which is expressed highly in the proximal tubule of the kidney. Fructose metabolism begins with phosphorylation by ketohexokinase (KHK), also highly expressed in the proximal tubules. We wanted to determine if changes in diet of proximal tubule-IR/IGF1R double knock-out mice and wild-type mice fed a high fructose diet would lead to changes in their metabolic response.

In this study, we hypothesized that in proximal tubule-IR/IGF1R double knock-out mice there would be changes in the metabolic response of the renal proximal tubule as a result of high levels of dietary fructose. Proximal-tubule select IR/IGF double KO mice were bred and genotyped at Georgetown University. Mice were fed control or a high-fructose diet for 4 weeks. The mice were euthanized and kidneys were collected for analysis. Western blots completed on whole kidney homogenates to assess the expression of Napi-2, KHK, GLUT5, IR, IGFR and NHE3. Across both sexes, the expression of GLUT5 was increased in fructose diet in both WT and KO mice. For WT mice, there was an increase in the expression of NHE3 for both sexes. Mice were discovered to have differences in response to fructose diet between IR/IGF KO and WT mice and male and female mice. We conclude that insulin and/or IGF1 act in the renal proximal tubule to affect the response to dietary fructose. Disruptions in this may predispose subjects to the development of type 2 diabetes.

Supported and funded by the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), APS Short-Term Research Experience for Underrepresented Persons (STEP-UP) Fellowship and the Department of Medicine at Georgetown University.

A FRAMEWORK FOR THE BEHAVIOR OF RARE VARIANT TESTS IN THE PRESENCE OF LARGE NUMBERS OF VARIANTS

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As we continue in the era of next-generation sequencing data, questions about how to most appropriately analyze genetic sequence data still remain, particularly with regard to the influence of rare genetic variants in human disease. Numerous rare variant testing methods have been proposed, but as sample size and coverage increase, along with a shift towards whole genome sequencing, the number of variants being considered continues to increase. Current gene-based rare variant testing methods have been well-tested for sets of less than 100 variants, but as the number of variants increases into the hundreds and thousands, anecdotal evidence suggests current industry standard methods may no longer perform optimally. Alternatively, two-stage methods (e.g., pathway methods) offer an alternative approach which may be better in some cases. This framework (1) classifies proposed variant set tests and explains observed differences in performance which can be used to directly connect genetic disease models with statistical power, (2) guides researchers in prospective test selection, and (3) provides the opportunity to analytically evaluate novel set-based rare variant tests.

We acknowledge that this work was funded by the National Science Foundation (MCB 1330734) and National Institutes of Health (R15-HG006915).

MECHANISM OF ACTION OF SCHWEINFURTHINS, A NEW CLASS OF ANTICANCER COMPOUNDS

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Schweinfurthins are a family of natural compounds that selectively inhibit the growth of, or possibly kill, a novel subset of NCI-60 cancer cell lines. This unusual pattern of susceptibility to treatment with the compounds suggests a unique mechanism of action. Recent reports have indicated that natural schweinfurthins act by binding to oxysterol-binding proteins to prevent cholesterol transport at the level of the trans-Golgi. However, these observations alone do not explain why some cells are susceptible and others are resistant.

We proposed schweinfurthins do not block cholesterol transport solely at the trans-Golgi. Instead, we hypothesized that schweinfurthins act at multiple sites to inhibit cholesterol transport.

We undertook live cell fluorescence imaging to establish whether schweinfurthins compromise cholesterol uptake. After twenty-four hours of schweinfurthin treatment, we found that in susceptible and resistant cell lines the classical endocytic pathway of cholesterol uptake was not functional and treatment induced cellular autophagy. Fixed cell immunofluorescence demonstrated the effect of schweinfurthins on membranes involved in moving cholesterol between organelles. These data suggested that treatment with compound abrogated the integrity of membranous organelles in a differential manner between susceptible and resistant cells.

With evidence of disruption of membrane compartments, we tested the possibility that growth and survival signaling pathways may be compromised. We discovered that EGF Receptor activation of Akt, under conditions of normal growth and following treatment with compound, was significantly different for the two cell types.

We conclude that schweinfurthins act at multiple sites to inhibit cholesterol transport. Furthermore, they induce the disintegration of cellular compartments and lead to the shutdown of growth and survival signaling pathways. Thus, schweinfurthins may represent a new class of anticancer agents that selectively inhibit the growth of cancer cells unable to tolerate the deregulation of cholesterol trafficking and oncogenic signaling.

Financial support provided by Penn State Hershey SURIP and Penn State Hershey Department of Pharmacology.

EFFECT OF LYSOPHOSPHATIDIC ACID ON E-CADHERIN LOCALIZATION IN MIGRATING METASTATIC BREAST CANCER CELLS

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As epithelial tumors progress, tumor cells invade the local environment and at a later stage travel to distant sites to form metastatic tumors. Metastasis significantly impacts patient outcome, but its mechanism is poorly understood: how the migratory phenotype of metastatic cells differs from invasive or normal epithelial cells remains unclear. Although it is assumed more malignant cells migrate more, time lapse imaging has shown that lung colony forming breast cancer cells (MCF10CA1, "M4") migrate with similar speed and over similar distances as normal breast epithelial (MCF10A, "M1") cells, while the variability of both cell speed and directionality increase in tumor cells (Weiger et al. 2013). Interestingly, lysphosphatidic acid (LPA) reduces variability of both cell speed and directionality in M4 cells, and renders the migratory phenotype similar to M1 cells. LPA treated M4 cells also express the cell-cell adhesion protein E-cadherin such that the membrane staining pattern becomes more prevalent.

It has been previously assumed that low E-cadherin levels and consequently weak cell-cell adhesions support tumor cell migration from the primary tumor towards metastatic sites: E-cadherin expression was shown to be reduced in metastatic disease. Now E-cadherin expression is also recognized as necessary to establish metastatic tumor growth upon arrival at the metastatic site.

Here, we used M4 cells to study the effect of LPA on E-cadherin expression patterns. Specifically, we determined the timeline of E-cadherin relocalization after stimulation with LPA, and whether additional microenvironmental cues like epidermal growth factor (EGF), hepatocyte growth factor (HGF), stromal cell-derived factor (SDF-1α), and transforming growth factor beta (TGF-β1) influence the effect of LPA on E-cadherin.

This work was funded through the National Cancer Institute's CCR Cancer Research Interns Program.

ADIPOCYTE mTORC2 KNOCKOUT DISRUPTS WHOLE-BODY GLUCOSE AND LIPID METABOLISM AND IMPAIRS THE BEIGEING OF WHITE ADIPOSE

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Rapamycin is an mTOR inhibitor that has been shown to increase lifespan in a range of organisms from yeast to mammals and is widely used as an antiproliferative and immunosuppressant in humans. Despite the apparent benefits of rapamycin treatment, the drug also causes dysregulation of glucose and lipid metabolism, leading to symptoms resembling type 2 diabetes. Recent findings suggest that these changes may result to some extent from rapamycin's ability to block the beigeing of white adipose tissue (WAT). Upon cold exposure or β3-adrenergic stimulation, beige fat is induced to form in WAT depots and is capable of initiating a thermogenic program similar to that characteristic of brown adipose tissue (BAT). The expression of Uncoupling Protein 1 (UCP1) in beige fat and BAT promotes energy balance by consuming excess glucose and lipids and dissipating the resulting energy as heat; therefore, the inhibition of beigeing in WAT may play a role in rapamycin's negative effects on metabolic homeostasis.

Chronic rapamycin inhibits both mTOR complex 1 (mTORC1) and the less well-characterized mTORC2. To test the hypothesis that the inhibition of mTORC2 by rapamycin contributes to the block in beigeing of WAT, mice containing an adipocyte-specific deletion of the mTORC2 subunit Rictor were first phenotyped and then exposed to a mild cold stress. The knockout mice displayed signs of metabolic dysfunction including hyperglycemia, hyperinsulinemia, and insulin resistance in addition to elevated serum cholesterol, triglycerides, phospholipids, and free fatty acids. During the three-day cold challenge, no significant differences in core body temperature, body weight, or food intake were observed between control and knockout mice. However, other results including abnormal beige fat morphology, decreased UCP1 expression, and decreased expression of thermogenesis-associated genes (UCP1, ELOVL3, and PGC-1α) in the WAT of cold-exposed knockout mice suggest that mTORC2 knockout at least partially impairs beigeing.

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PURIFICATION AND ANALYSIS OF HUMAN LIVER FATTY ACID BINDING PROTEIN (FABP1).

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Obesity is a chronic metabolic disorder affecting over one third of Americans. It is characterized by excessive accumulation of adipose tissue and is correlated with increased risk for cardiovascular disease, hepatosteatosis and Type 2 diabetes. Human Liver Fatty Acid Binding Protein (FABP1) functions to metabolize long chain fatty acids in the hepatocyte and is linked to metabolic processes such as lipid storage, oxidation, and gene expression. Whole body ablation of FABP1 in high fat fed obese C57Bl/6J mice results in reduced hepatosteatosis and increased insulin sensitivity. This suggests that small molecule inhibitors of FABP1 may be efficacious in reducing hepatic steatosis. To facilitate FABP1 inhibitor studies, the protein was expressed in E. coli and purified using nickel affinity column chromatography followed by delipidation via lipidex column chromatography. X-ray crystallographic studies have revealed that the lipid binding domain of FABP1 is found within a large interior cavity. Lipid binding was measured using the fluorescent fatty acid analogue 1,8-ANS and revealed a binding affinity of ~ 1.8 μM. Moreover, fatty acids displaced 1,8-ANS from the binding cavity thereby enabling analysis of therapeutic ligands. These studies offer the potential for identifying small molecule inhibitors of LFABP useful in the prevention of liver steatosis. Supported by the NIH R25 HL088728 and the University of Minnesota Life Science Summer Undergraduate Research Program.

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Afternoon Poster Session
Group Z – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

154. **SYNTHESIS OF INHIBITORS FOR UFM1 ACTIVATING ENZYME, UBA5**

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155. **OVEREXPRESSION OF CAH1 AND CAH8 IN CHLAMYDOMONAS REINHARDTII**

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156. **METABOLIC FLUX ANALYSIS OF TWO EXTREME THERMOPHILES: *THERMUS THERMOPHILUS* AND *RHODOTHERMUS MARINUS***

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157. **THE CHARACTERIZATION OF AN ESSENTIAL PROTEIN SPECIFIC TO TRYPANOSOME PARASITES**

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158. **CROSS-LINKING STUDIES TO INVESTIGATE STABLE DIMER FORMATION IN *E. COLI* GMPS**

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159. **TEMPORAL DYNAMICS OF CASPASE ACTIVITY IN JURKAT CELLS**

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160. **METHYLATION OF GNG7 IN HUMAN BREAST CANCER TISSUES**

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Department of Biological and Allied Health Sciences, Bloomsburg University, 400 E 2nd Street, Bloomsburg, PA 17815

161. **MOLECULAR-LEVEL ALTERATION OF SIGNALING AND METABOLIC PATHWAYS IN CANCER IMMUNOTHERAPY**

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SYNTHESIS OF INHIBITORS FOR UFM1 ACTIVATING ENZYME, UBA5

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Post-translational modifications occur on proteins after they are synthesized and play an important role in cell signaling. Ufmylation is a post-translational modification that has been associated with erythrothpoeisis, ER stress, and kinetoplastid metamorphosis; it has also been found to be up-regulated in individuals suffering from Leishmaniasis or metastatic breast cancer. This enzymatic pathway is similar to Ubiquitination in that it requires a team of three proteins (E1, E2, and E3) to tag a target protein with the eponymous protein, Ufm1. The only known E1 enzyme for this pathway is Uba5. Inhibitors for this enzyme activating step have been synthesized via Steglich reaction between AMP and Glycine, as confirmed by ^{31}P NMR. The mechanism and function of Uba5 is currently unknown; the synthesis of these inhibitors serves to aid in our understanding of the mechanism of Uba5.

OVEREXPRESSION OF CAH1 AND CAH8 IN CHLAMYDOMONAS REINHARDTII

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Lipids derived from algae hold great promise as a sustainable fuel source. However, commercial production of algal fuels is expensive in comparison with fossil fuel. Genetic manipulation of algae can make biofuel production more efficient. The photosynthetic green alga *Chlamydomonas reinhardtii* is a well-studied model organism that is easily manipulated at the molecular genetic level. In this study we are focusing on improving the carbon concentrating mechanism (CCM), a biological adaption to low carbon dioxide levels in the atmosphere. The CCM is composed of mainly three components: carbonic anhydrases, CO_2 transport proteins, and the pyrenoid. Carbonic anhydrases catalyze the interconversion of carbon dioxide and bicarbonate and thereby make inorganic carbon more accessible to the cell. The aim of this study is to increase the uptake of CO_2 in *C. reinhardtii* by overexpressing two periplasmic (between the cell wall and membrane) carbonic anhydrases, CAH1 and CAH8, and ultimately determining the effect on growth rate. *C. reinhardtii CAH1* and *CAH8* coding regions were synthesized with *C. reinhardtii* codon bias and epitope tags and the gene fragments were subcloned into expression vector pARG which contains the *ARG7* gene required for arginine biosynthesis. We transformed the *CAH1* vector into an *arg7* mutant strain and selected several ARG^+ survivors for western blot analysis to determine the expression of protein; *CAH8* lines will be generated later. We will select the best expressing lines for growth curve and dry weight analysis to determine whether the transformants overexpressing CAH1 or CAH8 are able to grow faster than the wild-type *C. reinhardtii* strain. In future both genes could be expressed together. If we are successful, the next step will be to incorporate these methods for microalgae that naturally produce higher lipid levels than *C. reinhardtii*, but are harder to manipulate, such as *Chlorella vulgaris*.

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 Science Foundation (NSF) Directorate for Engineering (ENG) Office of Emerging Frontiers in Research and Innovation (EFRI).

METABOLIC FLUX ANALYSIS OF TWO EXTREME THERMOPHILES: *THERMUS THERMOPHILUS* AND *RHODOTHERMUS MARINUS*

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Two thermophilic organisms, *Thermus thermophilus* and *Rhodothermus marinus* were investigated. A key characteristic of *Thermus thermophilus* is that it grows on multiple sugars. It was evolved for simultaneous co-utilization of glucose and xylose. The metabolism of the evolved *Thermus thermophilus* strain was researched. A key characteristic of *Rhodothermus marinus* is that it grows on glucose at high salt concentrations. Optimal growth conditions were investigated for *Rhodothermus marinus.* Parallel isotopic labeling experiments were performed on *Rhodothermus marinus* using six tracers: [1-^{13}C], [2-^{13}C], [3-^{13}C], [4-^{13}C], [5-^{13}C], and [6-^{13}C] glucose. ^{13}C-Metabolic flux analysis was used to quantify detailed intracellular metabolic fluxes on both organisms.

I would like to acknowledge the Antoniewicz group, the HHMI Science Scholar Program and the National Science Foundation MCB-1120684.

THE CHARACTERIZATION OF AN ESSENTIAL PROTEIN SPECIFIC TO TRYPANOSOME PARASITES

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Trypanosoma brucei, the single-celled eukaryotic parasite responsible for causing African Sleeping Sickness, has many unusual biochemical features and processes, including RNA Polymerase II (RNAPII)-dependent gene transcription. We have identified a nuclear, trypanosome-specific protein that was originally found associated with the basal transcription factor TFIIB, and is essential for parasite growth as assessed by RNA interference. This protein, named Essential Nuclear Factor (ENF), contains motifs associated with known transcription factors, but the protein is unique to trypanosomes. The function of this protein is unknown. We present the construction of a GST-tagged *Trypanosoma brucei* ENF (*Tb*ENF) that will be used for the identification of *Tb*ENF-interacting proteins. The identification of these proteins should shed light on *Tb*ENF function.

We gratefully acknowledge support for this work from the Villanova University Department of Chemistry.

CROSS-LINKING STUDIES TO INVESTIGATE STABLE DIMER FORMATION IN *E. COLI* GMPS

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TEMPORAL DYNAMICS OF CASPASE ACTIVITY IN JURKAT CELLS

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Apoptosis, a process in which a cell systematically triggers its own death in response to DNA damage or external stimuli, is widely utilized in the body. Malfunction of the apoptosis process can lead to serious health problems such as cancer. There are several known pathways that execute apoptosis utilizing a family of enzymes called Caspases.

The goal of this research project was to find suitable initiators of apoptosis in Jurkat T-Lymphocytes and elucidate their temporal dynamics with respect to caspase activity. Microfluidic devices were fabricated to capture cells and view this process. To identify suitable inducers of apoptosis to use, cells were exposed to several compounds and monitored over six hour time periods using Fluorescence Microscopy. Caspase activation was confirmed with the use of a caspase-specific fluorogenic probe, L-bisaspartic acid rhodamine 110.

Anti-CD95, staurosporine, and hydrogen peroxide were used as inducers of apoptosis resulting in induction rates of 8.88%, 1.66%, and 1.24% with un-induced rates of 0.59%, 1.88%, and 0.24% respectively. On a microfluidic device, cells were induced with Hydrogen Peroxide and their fluorescence intensity was measured over time. Apoptotic cells with active caspase enzymes consistently reached peak intensity two hours after caspase activation began. These results indicate that similar procedures could be used for more detailed monitoring of caspase activity in the future.

This research would not be possible without support from the Department of Chemistry at the University of Mary Washington, as well as a grant from the University of Mary Washington Summer Science Institute.

METHYLATION OF GNG7 IN HUMAN BREAST CANCER TISSUES

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Epigenetic regulation of heterotrimeric guanosine nucleotide-binding protein (G-protein) subunits has been associated with human tumors and cancer. There are three families of genes that encode each of the three subunits of the heterotrimeric G-proteins: α, β, and γ. Previous researchers have studied the epigenetic regulation of GNG7, a gene that encodes a γ-subunit, and found that the GNG7 promoter was highly methylated in head and neck cancers. New insight may be provided to the understanding of gene regulation by quantifying methylation levels near the promoter region of the gene GNG7 in human breast cancer and adjacent normal DNA. Two methods were applied in this project to quantify methylation. One method involved digesting tumor and normal DNA with methylation-specific restriction enzymes. In the second method, the DNA was treated with sodium bisulfite followed by either Sanger sequencing or methylation-specific PCR. HeLa cell DNA was used to optimize both of these procedures and as a positive control. To this date, four patients have been analyzed by the restriction enzyme method and ten patients have been analyzed by the bisulfite method out of the total sample size of ten patients. The results from the restriction enzyme digest method suggest there is no difference in methylation between tumor and normal DNA. The results from the sodium bisulfite method indicate that there is significantly more methylation in the breast cancer DNA compared to the adjacent normal DNA at two out of the four CG dinucleotide sites that were analyzed. According to our results thus far, methylation of GNG7 may play an important role in how the gene is expressed in human breast cancer.

MOLECULAR-LEVEL ALTERATION OF SIGNALING AND METABOLIC PATHWAYS IN CANCER IMMUNOTHERAPY

Sarah Pollock, Danielle Schmitt, Songon An, and Minjoung Kyoung
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Afternoon Poster Session
Group AA – Biochemical & Molecular Biology

Poster # | **Title, Author(s) & Affiliation(s)**

162. **INVESTIGATION OF REACTION CENTERS FROM RHODOBACTER SPHAEROIDES IMMOBILIZED ON GRAPHENE OXIDE**

Jessica G. Asiedu[1], Lukasz Krawiec[1], Mike Kopka[2], Arpit Patel[2], Sam L. Groveman[3], Dominic Hull[4], Robert A. Niederman[2], and Michele Vittadello[*,1]
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[4]Chemistry Department, Queensborough Community College of CUNY, Bayside, NY 11364

163. **OPTIMIZATION OF SELEX PARAMETERS FOR RNA APTAMER SELECTION**

Samuel Clark, and Randall Reif
Department of Chemistry, University of Mary Washington, 1301 College Avenue, Fredericksburg, VA 22401

164. **CHARACTERIZATION AND RATE ANALYSIS OF THE ENZYME BROMOPEROXIDASE IN DIVERSE MICROALGAE**

Amelia Harrison[1], Kevin Posman[2], Daniel Witt[2], and Steve Archer[2]
[1]Departments of Entomology and Wildlife Ecology and Marine Science and Policy, University of Delaware, Newark, DE 19716
[2]Bigelow Laboratory for Ocean Sciences, 60 Bigelow Drive, East Boothbay, ME 04544

165. **INTERACTIONS BETWEEN WATER SOLUBLE PORPHYRINS AND C-TYPE CYTOCHROMES**

C. Alex Hudson and Oleksandr Kokhan
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166. COMPARISON OF STABILITY AND KINETIC PROPERTIES OF DSZB FROM *N. ASTEROIDES* A3H1 AND *R. ERYTHROPOLIS* IGTS8

Alanna Hutchinson-Lundy, Austin Crithary, Jonathan Schmitz, and Linette Watkins
Department of Chemistry and Biochemistry, James Madison University, 800 S. Main Street, Harrisonburg, VA 22807

167. PEGLYATED BIODEGRADABLE BRAIN PENETRATING PARTICLES FOR WIDESPREAD GENE DELIVERY

Young Eun Kim[1], Panagiotis Mastorakos[2,4], Eric Song[3], Clark Zhang[2,5], Sneha Berry[2,3], Hee Won Park[1], Jong Sung Park[2,6], Seulki Lee[2,6], Jung Soo Suk[2,4,5], and Justin Hanes[1,2,4,5]
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INVESTIGATION OF REACTION CENTERS FROM RHODOBACTER SPHAEROIDES IMMOBILIZED ON GRAPHENE OXIDE

Jessica G. Asiedu[1], Lukasz Krawiec[1], Mike Kopka[2], Arpit Patel[2], Sam L. Groveman[3], Dominic Hull[4], Robert A. Niederman[2], and Michele Vittadello[*,1]
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OPTIMIZATION OF SELEX PARAMETERS FOR RNA APTAMER SELECTION

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Department of Chemistry, University of Mary Washington,
1301 College Avenue, Fredericksburg, VA 22401

The primary research goal for this project was to optimize the initial steps of a process called Systematic Evolution of Ligands by Exponential Enrichment (SELEX). SELEX is used to discover aptamers, which are oligonucleotides (RNA or ssDNA) that bind to target molecules with high affinity and specificity due to their three dimensional conformation. Aptamers have been developed for a multitude of targets including; inorganic and small organic molecules, peptides, proteins, carbohydrates, antibiotics, and cells. High affinity aptamers are isolated by introducing RNA molecules with random sequences to a specific target molecule. The RNA molecules that bind to the target are then extracted, amplified, and used in subsequent rounds of SELEX. After each round of SELEX the RNA pool shifts toward higher affinity binding to the target molecule. The DNA library was constructed, its amplification optimized, and the purification of the DNA examined. The DNA was then transcribed into RNA and purified, concluding the first half of SELEX. In the future, the RNA will be introduced to the molecular target, Streptolysin O, a hemolytic exotoxin released by *Streptococcus pyrogenes*. With the development of an aptamer for Streptolysin O, a multitude of applications are possible including exotoxin quantification, deactivation, and purification.

The authors would like to acknowledge the Summer Science Institute and the University of Mary Washington's Department of Chemistry for providing the funding for the project, Kimberly Kerns for preliminary research, and the University of Maryland, Baltimore County for the opportunity to present at this event.

CHARACTERIZATION AND RATE ANALYSIS OF THE ENZYME BROMOPEROXIDASE IN DIVERSE MICROALGAE

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Vanadium-dependent bromoperoxidases (vBPOs) are a class of enzymes that use hydrogen peroxide (H_2O_2) to oxidize bromide and iodide. Several diatoms and cyanobacteria have been shown to have vBPOs, possibly to control H_2O_2 levels. While past research has focused on identifying organisms as having vBPOs, this study addresses the gaps in knowledge concerning differences in vBPO function between organisms and compares techniques to measure activity rates. The effectiveness of a spectrophotometric thymol blue assay was compared to a spectrofluorometric aminophenyl fluorescein (APF) assay. The rates of several phytoplankton species from different environments were compared by normalizing to chlorophyll and protein content. Three different levels of sample purification were also tested: purified enzyme, crude protein extraction, and live culture. This is the first demonstration of the APF approach applied to microbial phytoplankton. It was the more sensitive and accurate assay and was ideal for live culture measurements. The thymol blue assay was only useful for more purified forms of the enzyme. All species tested showed vBPO activity. The polar species had the most activity while the warmest water species had the least.

INTERACTIONS BETWEEN WATER SOLUBLE PORPHYRINS AND C-TYPE CYTOCHROMES

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In native photosynthetic proteins, light harvesting units are coupled to chains of redox-active cofactors responsible for charge transfer and stabilization at time scales sufficient for productive photocatalysis under ambient light conditions. In an attempt to emulate the complex biological phenomena of photosynthesis and develop systems for artificial photosynthesis, we attempted to combine and study interactions of the multiheme cytochrome c_4 from *Pseudomonas stutzeri* and c-type cytochrome from the photosynthetic reaction center of *Rhodopseudomonas viridis* with several water-soluble porphyrins. These multiheme frameworks found in cytochromes are capable of storing and transporting multiple electrons, while water-soluble porphyrins serve as more stable and readily available synthetic analog of chlorophylls. We successfully found conditions for heterologous expression of cyt c4 in *E.coli* and developed a protein purification protocol. The purified protein had expected spectral and redox properties. HPLC-MS verified the purity of the isolated protein and confirmed successful covalent attachment of both hemes with a mass of 22 kD. CD measurements demonstrated the expected alpha-helical nature of the protein and revealed melting temperatures of about 70° C. Small angle X-ray scattering further confirmed the expected size of the correctly folded protein. Static fluorescence experiments revealed that several porphyrins had substantial fluorescence quenching in the presence of cyt c4 which is likely due to a complex formation and electron transfer. We are currently working on the optimization of expression conditions for *R. viridis* c-type cytochrome, as well as on time-resolved transient absorbance experiments to verify charge transfer from the excited state of porphyrins to multiheme cytochromes.

COMPARISON OF STABILITY AND KINETIC PROPERTIES OF DSZB FROM *N. ASTEROIDES* A3H1 AND *R. ERYTHROPOLIS* IGTS8

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Dibenzothiopehene (DBT) and its derivatives comprise up to 60% of the organosulfur contamination of crude oil. The enzyme 2-(2'-hydroxyphenyl) benzenesulfinate desulfinase (DszB) catalyzes the carbon-sulfur bond cleavage in the final, and rate-limiting step in the biodesulfurization of DBT. The DszB enzyme from *Nocardia asteroides* A3H1 and *Rhodococcus erythropolis* IGTS8 was overexpressed in *E. coli*, purified and characterized kinetically. Kinetic assays revealed a sigmoidal response when the velocity was plotted against [S], calling into question the previously proposed monomeric structure of the enzyme. The stability of the enzyme was measured under various storage conditions and increased stability was observed upon immobilization of the enzyme to CNBr-activated Sepharose beads. Salt and enzyme concentration studies were performed to optimize the fluorescence assay. Enzyme stability was tested with various cryoprotectants. These studies aid in the understanding of the factors that control the rate and stability of the desulfinase enzyme to learn how to make biodesulfization more economically feasible.

PEGLYATED BIODEGRADABLE BRAIN PENETRATING PARTICLES FOR WIDESPREAD GENE DELIVERY

Young Eun Kim[1], Panagiotis Mastorakos[2,4], Eric Song[3], Clark Zhang[2,5], Sneha Berry[2,3], Hee Won Park[1], Jong Sung Park[2,6], Seulki Lee[2,6], Jung Soo Suk[2,4,5], and Justin Hanes[1,2,4,5]
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Afternoon Poster Session
Group BB - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

168. ***PSEUDOMONAS* SP. REVEAL MAGNETOTACTIC BEHAVIOR ISOLATED FROM POND SEDIMENTS**

Usman Ahmad and Om V. Singh
Division of Biological and Health Sciences, University of Pittsburgh at Bradford, Bradford, PA 16701

169. **ISOLATION AND CHARACTERIZATION OF BACTERIA IN THE BUILT ENVIRONMENT: MEASURING THE EFFECT OF PERSONAL CARE PRODUCTS AND PHARMACEUTICALS ON GROWTH**

Amanda Finck, Kelly DiGeronimo, and Zakiya Whatley
Department of Biology, Gettysburg College, 300 North Washington Street, Gettysburg, PA 17325

170. **IMPACT OF ANTIBIOTICS ON HORIZONTAL GENE TRANSFER IN *VIBRIO CHOLERAE***

Swathi Penumutchu[1,2], Peter Belenky[2], and Benjamin Korry[2]
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[2]Department of Molecular Microbiology and Immunology, Brown University, 171 Meeting Street, Providence, RI 02912

171. **A STUDY ON THE LONG-TERM PERFORMANCE OF THE SAWYER PointONE™ FILTER IN THE DEVELOPING WORLD**

Holly Ross[1,3], Andrew Nevin[1,3], Daniel Yeisley[2,3], Thomas Soerens[2,3], Lawrence Mylin[1,3] and Erik Lindquist[1,3]
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172. **INTERFERON GENE EXPRESSION IN CHICKEN EMBRYO FIBROBLASTS INFECTED WITH RECOMBINANT HERPESVIRUS OF TURKEYS EXPRESSING miRNAs**

Nazanin Sarpoulaki, Michelle A. Cross, Amy S. Anderson, Erin L. Bernberg, and Robin W. Morgan
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173. **THE NOVEL ISOLATION OF *ARTHROBACTER* PHAGES WITH A ROBUST NEW METHOD**

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PSEUDOMONAS SP. REVEAL MAGNETOTACTIC BEHAVIOR ISOLATED FROM POND SEDIMENTS

Usman Ahmad and Om V. Singh
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Magnetotactic bacteria (MTB) are considered among unique and diverse group of microorganisms with the ability to orient and migrate along geomagnetic field lines. This unique property is based on specific intracellular organelles, the magnetosomes, a membrane bound crystals of magnetic iron minerals organized into chains via dedicated cytoskeleton. Due to occurrence of magnetosomes, MTB are of great interest for paleomagnetism, environmental magnetism, biomarkers in rocks, and biomineralization. The bacterial magnetites have been exploited for a variety of applications in modern biological and medical fields. We hypothesized that pond sediments would contain variety of MTB. We aimed to develop a series of racetrack methods to isolate MTB from pond sediments located in Alleghany National Park area. Total microbial flora of pond sediment was enriched in nutrient broth (NB) medium and applied to racetrack under magnetic field generated using magnetic bars of opposite pole (North—South) with respect to control (i.e. no magnet). Microorganisms moved towards each North and South pole were observed under microscope and enriched in NB medium followed by single cell isolation method on nutrient agar plates, and denoted as MTB1 and MTB2. 16S rRNA sequencing revealed both microorganisms revealed close similarity to *Pseudomonas chlororaphis* (99.39%) and *Pseudomonas koreensis* (99.89%). Microbial growth at varying temperature was characterized to explore the magnetotactic behavior among isolates. Both isolates revealed manatotactic behavior towards magnetic field under different magnetic racetrack.

ISOLATION AND CHARACTERIZATION OF BACTERIA IN THE BUILT ENVIRONMENT: MEASURING THE EFFECT OF PERSONAL CARE PRODUCTS AND PHARMACEUTICALS ON GROWTH

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This work reports the isolation and characterization of bacteria from the built environment at Gettysburg College in Gettysburg, PA. Surfaces of a water fountain on campus were swabbed and serially streaked to isolate multiple bacteria on R2A agar. Following multiple rounds of growth, the unknown microbial candidates were narrowed to two visibly-distinct organisms. Morphological characterization and phylogenetic identification based on 16S rDNA sequencing revealed that the isolates were *Chryseobactierum hispalense* and *Microbactierum maritypicum*. We report synergistic biofilm formation between *Chryseobactierum hispalense* and *Microbactierum maritypicum*. The contamination of drinking water with varying levels of personal care products and pharmaceuticals (PCPPs) is well documented. Additionally, these environmental pollutants and their derivatives affect aquatic life, as illustrated with effect of the antidepressant fluoxetine on mudsnails. To determine if previously reported contaminants affect freshwater bacteria, we assessed both planktonic growth and biofilm formation following exposure to nalidixic acid (non-fluorinated quinolone antibiotic), diphenhydramine (over-the-counter drug Benadryl), and fluoxetine (Prozac).

Funding for this work was provided by the HHMI Grant #52007540 Enhancing Cross-disciplinary Sciences at Gettysburg College.

IMPACT OF ANTIBIOTICS ON HORIZONTAL GENE TRANSFER IN *VIBRIO CHOLERAE*

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A STUDY ON THE LONG-TERM PERFORMANCE OF THE SAWYER PointONE™ FILTER IN THE DEVELOPING WORLD

Holly Ross[1,3], Andrew Nevin[1,3], Daniel Yeisley[2,3], Thomas Soerens[2,3], Lawrence Mylin[1,3] and Erik Lindquist[1,3]
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Lack of sustainable access to safe water and sanitation services dramatically impacts the health and productivity of many people living in low-income countries. The infrastructure improvements needed to provide such services are costly and slow to implement. A number of NGOs have sought instead to distribute low-cost household water filtration units in community-focused programs while providing basic training in filter use and health education.

The Sawyer PointONE™ Filter has been used in both relief and development settings; however, some NGOs have been reticent to adopt these filters, citing factors such as uncertain longevity in the field, culture/NGO-related challenges associated with filter use, and limits for repair or replacement of broken filters/filter components. In fact, one study (Murray *et al*., Journal of Water, Sanitation and Hygiene for Development, *in press*) reports serious, negative claims regarding breakage and fouling-related failure of these filters. Such claims would call into question the ability of these filters to produce adequately disinfected effluent water after two years of household use.

We conducted a two-nation study that included collection of Sawyer PointONE™ Filters that had been in use for five or more years in Cochabamba, Bolivia and Nadi, Fiji. Microbial loading and turbidity in source and effluent water from unmanipulated or chlorine-backwashed filters were tested in the field and later under controlled laboratory conditions. Field and laboratory results from quantifying total coliforms and *E. coli* loads and turbidity in source and effluent water will be presented.

While relatively few filters were located five or more years following initial distribution, a majority of those collected did reduce bacterial loads in effluent water to comply with WHO guidelines. Based on these and other results, our study will help to answer important questions about longevity for these hollow micro-fiber filters.

This study was supported in part by the Steinbrecher Endowment for Research in the Health and Life Sciences (Messiah College) and Sawyer Products, Inc.

INTERFERON GENE EXPRESSION IN CHICKEN EMBRYO FIBROBLASTS INFECTED WITH RECOMBINANT HERPESVIRUS OF TURKEYS EXPRESSING miRNAs

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Marek's disease virus serotype 1 (MDV1) is an alphaherpesvirus that causes T-cell lymphomas in chickens. Among MDV1 miRNAs, mdv1-miR-M4 is important for oncogenicity and maps upstream of the *meq* gene in a miRNA cluster. Mdv1-miR-M4 shares a seed sequence with miR-155, which is conserved across phylogeny. We inserted mdv1-miR-M4, the *meq* miRNA cluster, as well as miR-155 into the genome of herpesvirus of turkeys (HVT) in order to study the functions of these miRNAs. Expression of these miRNAs by recombinant HVTs following infection of chicken embryo fibroblasts inhibited expression of the interferon inducible genes MX-1, OAS-3, and ISG-15 compared to infection with the parent HVT. We also examined expression of IFN-alpha, IFN-beta, and IFN-lambda post infection. IFN-alpha and IFN-beta were not induced in this system under our experimental conditions (2-fold change or higher), a result that is consistent with known inhibition of these interferons by other herpesviruses. Despite the lack of type 1 interferon induction, interferon inducible genes (MX-1, OAS-3, and ISG-15) were induced suggesting that their expression might be type 1 interferon independent. Inhibition of MX-1, OAS-3, and ISG-15 induction by *meq* miRNAs was seen at 3, 6, and 48 hours after infection. Our results support the conclusion that the *meq* miRNAs, especially mdv1-miR-M4, augment inhibition of innate immune responses.

The Howard Hughes Medical Institute provided funding for this research.

THE NOVEL ISOLATION OF *ARTHROBACTER* PHAGES WITH A ROBUST NEW METHOD

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Bacteriophage isolation from environmental samples has been performed for decades using principles set forth by pioneers in microbiology. The isolation of phages infecting *Arthrobacter* hosts has been limited due to the low success rate of many previous isolation techniques. This had resulted in an underrepresented group of *Arthrobacter* phages available for study, until the recently new enrichment technique developed at Cabrini College. Unlike many others, the new method uses a filtered extract that is free of contaminating bacteria as the base for indicator bacteria growth, *Arthrobacter sp.* ATCC 21022, specifically. By first removing soil bacteria the target phages are not hindered by competition with native soil bacteria present in initial soil samples. This enrichment method has resulted in 26 unique phages from several different soil types. Different types of phages were even produced from the same enriched soil sample isolate, in some cases. The growth characteristics of these phages have been examined and they have a variety of ideal growth temperatures and cation concentrations.

We would like to thank Dr. David Dunbar of Cabrini College for his passion and being a great mentor, and Dylan Chudoff for his hard work in being the teaching assistant. We also would like to thank the SEPCHE institutions for funding the course, and Dr. Karen Snetslaar from Saint Joseph's University for taking time out of her schedule to take electron microscope pictures of the class's phages.

Afternoon Poster Session
Group CC - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

174. **IDENTIFICATION OF A NOVEL YEAST SPECIES ISOLATED FROM AN URBAN UNIVERSITY PARK**

Dylan M. Curry, and Matthew J. Farber
Department of Biology, University of the Sciences, 600 S. 43rd Street, Philadelphia, PA 19104

175. **EPIGENETIC REGULATION OF CRX BINDING TO REGULATORY ELEMENTS IN THE VERTEBRATE RETINA**

Annamarie Meinsen, Nicholas Dunham, Morgan Hedden, Courtney Stout, and Raymond Enke
Department of Biology, James Madison University,
800 South Main Street, Harrisonburg, VA 22807

176. **A GENETIC SCREEN FOR BLOOD-INDUCED PROMOTERS FROM *ASAIA BOGORENSIS*, A MIDGUT SYMBIONT OF MALARIA VECTOR MOSQUITOES**

Lianna Paul[1], Jackie Shane[1], Nicholas Bongio[1, 2], and David Lampe[1]
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[2]Department of Biology, Shenandoah University, 1460 University Drive, Winchester, VA 22601

177. **TRANSCRIPTOME ANALYSIS OF ALLIGATOR ADIPOSE**

Blair Schneider[1], Colin Kern[2], Allen Hubbard [3], Wayne Treible [4], John W. Finger Jr. [5],
Tracey Tuberville [6], Travis C. Glenn [5], Matt Hamilton [6], and Carl J. Schmidt[7]
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178. **THE ZEBRAFISH *NEUREXIN 1B*: GENOMIC ANALYSES AND cDNA CLONING**

Daniel Sergeyev, Majesta Kitts, and Cheng Huang
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179. **ANALYSIS OF DNA METHYLATION AT THE C-REGION OF IMPRINTED *RASGRF1* ACROSS MOUSE DEVELOPMENT**

Kristian Sumner and Tamara L. Davis
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IDENTIFICATION OF A NOVEL YEAST SPECIES ISOLATED FROM AN URBAN UNIVERSITY PARK

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Yeast are versatile microbes with many applications, including food production, ethanol production, and bioremediation. They are also quite diverse and abundant, with over 5.1 million species predicted and only 100,000 currently described[1]. Therefore, a survey of the local yeast in an urban university park in Philadelphia was conducted to identify population patterns and to isolate potential novel species. These studies led to the discovery of a novel, pink yeast species of the order Sporidiobolales.

Through DNA sequencing and molecular phylogenomics, it was determined that this yeast species is most closely related to *Rhodotorula colostri*. Carbon assimilation data and traditional microbiology morhphology were used to confirm the unique identity of the novel isolate.

Reference:

1. Blackwell S. (2011). The Fungi: 1, 2, 3 … 5.1 million species? *American Journal of Botany,* March 2011, vol. 98, no. 3, 426-438 http://www.amjbot.org/content/98/3/426.full

This research was supported by the Department of Biology at the University of the Sciences. We would like to thank Peter B. Berget, PhD for helpful comments and feedback during the duration of the project.

EPIGENETIC REGULATION OF CRX BINDING TO REGULATORY ELEMENTS IN THE VERTEBRATE RETINA

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The vertebrate retina is a light-sensitive, stratified layer of neuronal cells that lines the interior of the eye. Retinal neurons are responsible for converting light into electrochemical signals that ultimately allow the brain to process visual images. This specialized cellular function requires finely tuned regulation of gene expression during retinal development. CRX is a retina-specific transcription factor required for mature photoreceptor development in the retina. An epigenetic modification known as DNA methylation may play an important role in regulation of CRX-dependent retinal genes. Using a previously characterized CRX ChIP-seq data set and bisulfite pyrosequencing of genomic DNA, we demonstrate that high levels of DNA methylation in or adjacent to consensus CRX binding sites are associated with a lack of CRX binding and transcriptional silencing in the mouse retina. Interestingly, CRX binding to demethylated regulatory regions is sometimes but not always associated with active transcription. Collectively these findings suggest an important role for epigenetics in transcriptional regulation of retina-specific genes. Current studies are focused on characterizing epigenetic regulation of human homologs of murine retina-specific genes. Future studies will focus on biochemical interactions between CRX protein and methylated DNA.

This project is supported by funding from the Commonwealth Health Research Board, and JMU 4-VA. We also thank the Merbs lab at Johns Hopkins University.

A GENETIC SCREEN FOR BLOOD-INDUCED PROMOTERS FROM *ASAIA BOGORENSIS*, A MIDGUT SYMBIONT OF MALARIA VECTOR MOSQUITOES

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With over 3.4 billion people spanning 106 countries and territories at risk and an estimated 198 million cases and 500,000 deaths in 2013, malaria is a disease that poses a serious threat to global welfare. Caused by the parasite *Plasmodium*, malaria is a vector-borne disease transmitted to humans by the bite of an *Anopheles* mosquito. Its complex life cycle and ability to evolve quickly make the disease difficult to treat and control. Therefore, stopping the disease cycle within the mosquito could greatly decrease the incidence of infection.

Paratransgenesis, or the genetic modification of symbiotic bacteria to change host phenotype, offers one of the most promising preventative tactics. With *Anopheles* mosquitoes, symbiont *Asaia bogorensis,* when genetically altered, has the potential to halt the disease cycle within the host and prevent the spread of infective *Plasmodium* species. *Asaia* is a Gram-negative alphaproteobacterium that is normally found in the mosquito midgut as well as salivary glands and the ovaries of females. For this reason, the bacterium is ideal not only for disease cycle intervention but also the spread and persistence within the wild mosquito population. *Asaia* has previously been engineered with native secretion signals fused to antimalarial effector molecules. However, expressing these molecules constitutively would hinder bacterial survival in the wild.

With this in mind, we created a genetic screen to detect blood-induced promoters from *Asaia bogorensis* SF2.1. Insertion of genomic fragments into the plasmid GLR1 5' to the coding sequence of GFP allowed for visualization on iron-enhanced and deplete media, a representation of conditions preceding and following a blood meal. Isolates with possible conditionality were then given to blood-fed *Anopheles* mosquitoes and fluorescent imaging used to determine their presence within target organs. Ultimately, conditional isolates could be sequenced and tested further for potential in paratransgenesis.

All research was funded by grant 1R15AI107735-01A1 awarded to Dr. David Lampe by the NIH. Research was performed primarily during Duquesne University's Undergraduate Research Program, summer 2015.

TRANSCRIPTOME ANALYSIS OF ALLIGATOR ADIPOSE

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The crocodilians are model organisms for understanding environmental regulation of numerous biological processes including developmental biology, temperature regulation, sex determination and morphology. The recent completion of the *Alligator mississippiensis* genome provided the ability to define gene expression patterns in all tissues of this species. To better understand energy storage and mobilization, this work focuses on defining the transcriptome of alligator adipose tissue. Alligator fat body samples were provided by the Savannah River Ecology Laboratory at the University of Georgia and transcriptome libraries prepared with the following procedure: 1. Isolation of mRNA using *mir*Vana™ miRNA Kit, 2. Verification of mRNA quality and integrity by Fragment Analysis, 3. Transcriptome libraries prepared using Illumina Stranded RNA Prep Kit and sequenced at the Delaware Biotechnology Institute Core Sequencing Facility, 4. Transcription levels determined using the fRNAkenseq software, and 5. Identification of differentially expressed genes, data analysis, functional clustering and pathway analysis using JMP Pro 11 and PathRings. The University of Georgia provided additional transcriptome sequences from other tissues. To begin our analysis, we first identified transcripts enriched in adipose compared to all other alligator tissues. In addition to identifying transcripts encoding adipokines, adipose enriched transcription factors and numerous enzymes involved in processing fat, it is also clear that genes related to the immune system are enriched in adipose, specifically, genes linked to anti-inflammatory responses. Current effort focuses on comparing these results with those obtained from chicken adipose tissue to better understand the evolution of this tissue in the Archosaurs.

This material is based upon work supported by the National Science Foundation under Grant No. 1147029 and by the Agriculture and Food Research Initiative Competitive Grant No. by NIFA 2010-04233 from the USDA National Institute of Food and Agriculture.

THE ZEBRAFISH *NEUREXIN 1B*: GENOMIC ANALYSES AND cDNA CLONING

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One important goal in the field of Developmental Genetics is to identify genetic regulators of cell fate specification. Our work has identified the novel *neuer1* gene as an important regulator of blood cell fate specification in the model organism zebrafish (*Danio rerio*). The existence of a class of signaling ligands known as Neurexophilins that share a prominent protein domain with the Neuer1 protein indicates that Neuer1 may function as a novel signaling ligand. Neurexophilins bind to a family of receptors known as Neurexins, which further raises the question of which, if any, of the zebrafish Neurexins is bound by Neuer1. We reason that if we determine the expression pattern of all the zebrafish *neurexins*, the one that shares the same expression pattern with *neuer1* is likely the best candidate.

To determine the expression pattern for each of the zebrafish *neurexins* using *in situ* hybridization, we must first clone all the *neurexin* cDNAs, which can then be used to generate riboprobes that recognize each *neurexin* mRNA. However, this is an ambiguous undertaking, as there are 6 zebrafish *neurexin* genes (1a, 1b, 2a, 2b, 3a and 3b) each of which further gives rise to hundreds of mRNA isoforms due to exceptionally high levels of alternative splicing. Significantly, the intron/exon boundaries and splicing patterns are much better understood for the a forms than for the b forms. Here we report our strategy to predict the intron/exon boundaries for the b forms by using genomic analyses. Furthermore, we report the successful cloning of a *neurexin* 1b cDNA as a proof of the principle. Based on this work, we plan to further the understanding of the splicing patterns of *neurexin b* genes, as well as pursuing whether any of the Neurexin B proteins is a likely receptor for the potential ligand Neuer1.

This research project was funded by the McDaniel College Student-Faculty Collaborative Summer Research Fund and the Department of Biology.

ANALYSIS OF DNA METHYLATION AT THE C-REGION OF IMPRINTED *RASGRF1* ACROSS MOUSE DEVELOPMENT

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Genomic imprinting is an epigenetic mechanism where a small number of genes are either expressed or silenced depending on which parent the allele was inherited from: either the mother or the father. One way of understanding how imprinting occurs is through studying differential distribution of DNA methylation and histone modification on the parental alleles. These modifications help to distinguish the maternal and paternal alleles from each other and regulate the expression of these copies. In mice, *Rasgrf1* is an imprinted gene that is methylated on, and expressed from, only the paternal allele. Studying how this gene is regulated is important because if it isn't regulated properly there are defects in long-term memory formation.

It's believed that methylation of the paternal allele prevents the enhancer blocking protein, CTCF, from binding to the differentially methylated region (DMR) and thus *Rasgrf1* is expressed. The CTCF protein binds to the unmethylated maternal DMR and prohibits the enhancers from expressing *Rasgrf1*.

The size of the DMR was originally defined as 250 bp long. Recent research suggests that the *Rasgrf1* DMR is more expansive than previously thought. The sites of methylation that occur in the expanded region, the uDMR, are still being investigated and aren't as well defined as those in the original DMR. We're analyzing the C-region, which is 4 kb upstream from the originally defined DMR. By analyzing methylation patterns in this region we are learning more about how its DMR functions and how big the DMR is at different developmental stages. We are interested in understanding how and when these methylated regions change in size. We hypothesize that dynamic stage-specific methylation is an important aspect of *Rasgrf1* because it's involved in long-term memory formation. Experiments have shown that if it's not paternally methylated it's not expressed, which leads to memory formation defects.

Thank you to Dr. Tamara Davis and Rachel Shields (BMC '15) for their contributions towards the development of this project. Support for this research was provided by awards to TLD from NSF grant #1157819. Additional support for this research was provided through a HHMI science education grant awarded to Bryn Mawr College.

Afternoon Poster Session
Group DD - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

180. **IMPACT OF COAL ASH EXPOSURE ON EASTERN MUD TURTLE IMMUNE RESPONSE**

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181. **ASSESSING THE EFFECTIVENESS OF CHEMICAL AND PHYSICAL SUNSCREENS AND THEIR IMPACT ON THE AQUATIC ENVIRONMENT**

Grynyshin, K., Kamdar, H., Shupp, B., Gutowski, B., and Skokotas, A.
Biology Department, Rosemont College, 1400 Montgomery Avenue, Rosemont, PA 19010

182. **NITROGEN AND PHOSPHORUS EXCRETION BY NATIVE FRESHWATER MUSSELS AND INVASIVE CHINESE MYSTERY SNAILS IN NOVA SCOTIA**

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[2]Department of Environmental Science, Saint Mary's University, 923 Robie Street Halifax, Nova Scotia, Canada B3H-3C3

183. **SALINITY STRESS RESULTS IN DIFFERENTIAL *HSP* 70 EXPRESSION IN THE HOLOBIONT *AIPTASIA PALLIDA* AND *SYMBIODINIUM SP.***

Alana Thomas, Mitchell A. Ellison, and Susan L. Carney
Department of Biology, Hood College, 401 Rosemont Avenue, Frederick, MD 21701

184. **PURIFICATION OF VITELLOGENIN FROM NESTING LOGGERHEAD SEA TURTLES, *CARETTA CARETTA***

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IMPACT OF COAL ASH EXPOSURE ON EASTERN MUD TURTLE IMMUNE RESPONSE

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Coal-fired facilities operated on the Savannah River Site from 1951 to 2012. Coal combustion waste (coal ash) from the SRS D-Area coal facility was channeled into settling basins, which are used as habitats by a variety of organisms. As a result, these organisms are exposed to coal ash and its contents. Very little information is known about how reptiles are impacted by coal ash exposure. However, recent studies have discovered that reptiles are sub-lethally affected by coal combustion byproducts. To date, no research has been conducted on the Eastern Mud Turtle, *Kinosternon subrubrum*.

The main objective of our study was to examine the impact of coal ash exposure on the immune response of Eastern Mud Turtles. We also observed the role of size in innate immune response and compared innate immune strength of the Eastern Mud Turtle with the Yellow Bellied Slider, *Trachemys scripta.* We baited aquatic hoop net and promar traps to capture mud turtles and collected plasma samples for immune assays. We used the bacterial killing assay (BKA) to evaluate the strength of the innate immune response of turtles captured in contaminated and uncontaminated sites.

BKA results revealed that site history did not influence the innate immune response of Eastern Mud Turtles. Eastern Mud Turtles from ash and reference sites displayed and average bacterial killing efficiency of 98%. A positive correlation between larger size and killing ability was displayed by Mud Turtles. Bacterial killing ability of Eastern Mud Turtles appeared to be more effective than Yellow Bellied Sliders.

We would like to thank the National Science Foundation, Department of Energy, and Area Completion Projects for providing funding. This material is based upon work supported by the Department of Energy under Award Number (DE-FC09-07SR22506).

ASSESSING THE EFFECTIVENESS OF CHEMICAL AND PHYSICAL SUNSCREENS AND THEIR IMPACT ON THE AQUATIC ENVIRONMENT

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Ultraviolet light is nonioninzing radiation that damages cells and causes the formation of pyrimidine dimers in the DNA. In humans, exposure to UV light results in skin cancer, premature aging, and cataracts. Since UV light is also lethal to bacteria, *Escherichia coli* was used as the model organism. In this study, we address the effectiveness of chemical and physical (mineral) sunscreen products and present data on their ability to protect bacteria from cell death. Their impact on corals and their symbiotic algae will be examined.

NITROGEN AND PHOSPHORUS EXCRETION BY NATIVE FRESHWATER MUSSELS AND INVASIVE CHINESE MYSTERY SNAILS IN NOVA SCOTIA

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Native freshwater mussels, such as the Eastern Elliptio (*Elliptio complanata*), that are found in lakes in Nova Scotia, Canada, are essential for maintaining water quality and play a role in the nitrogen (N) and phosphorus (P) cycles through filter feeding and excreting N and P. Invasive Chinese mystery snails (*Cipangopaludina chinensis*) have been spreading across the Nova Scotia lakes and their N and P excretion rates may be greater than the native mussels. N and P are critical nutrients that promote algal blooms, leading to eutrophication that creates hypoxic zones that generates stress on the native mussels in the region. To measure N and P excretion rates by the mussels and snails, an experiment was conducted in Saint Mary's University's laboratory with both organisms. There were 18 five gallon aquariums with 6 different treatments; three mussel tanks, three mussel shell tanks, three snail tanks, three snail shell tanks, three tanks with combination of mussels and snails, and three blank tanks. Excretion rates of NO_3^-, NH_4^+, urea and TP of both organisms were measured every 6 hours during the 24 hour experiment. The experimental aquariums' pH (7.07-7.27) was lower than the control aquarium that contained only water from Pigott Lake (7.63) suggesting that excretion of different N and P compounds by both organisms had an effect on lowering the pH. NO_3^-, NH_4^+, urea and TP concentrations in the aquariums prior to the experiment and during sampling have yet to be analyzed to determine composition and rate of excretion. Preliminary pH data suggests that competition between the native mussels and invasive snails may have an impact on excretion rates that may alter the flow of N and P from filter feeders in Nova Scotia lake ecosystems.

I would like to thank the Gordon Brown Fund at Gallaudet University for funding my internship and research.

SALINITY STRESS RESULTS IN DIFFERENTIAL *HSP* 70 EXPRESSION IN THE HOLOBIONT *AIPTASIA PALLIDA* AND *SYMBIODINIUM SP.*

Alana Thomas, Mitchell A. Ellison, and Susan L. Carney
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Aiptasia pallida is a species of sea anemone that hosts symbiotic algae, *Symbiodinium sp.* These two organisms live together by the symbiont transferring nutrients to the host, and the host supplying shelter for the symbiont. When anemones are present in an environment with unnatural levels of temperature, light, salinity, or another stressor, they may expel some or all symbionts, a process called bleaching. Bleaching is a last resort to a stress response. During stress, heat shock proteins (Hsp) are produced in both organisms. By helping to stabilize partially unfolded proteins, Hsps aid in transporting proteins across membranes within the cell.

With salinity being the least studied stressor, the purpose of this experiment was to see if unusual salinity levels cause bleaching. These experiments were done to supplement experiments that monitored Hsp70 expression in both *Aiptasia pallida* and their symbionts to characterize cellular levels of stress. *Aiptasia pallida* clones were individually put in salinities of 27ppt, 30ppt, and 34ppt. After eight hours of exposure, the anemones tissue was homogenized and separated from the symbionts. The number of symbionts in the water and in the animals was counted on a hemocytometer and estimated using fluorescence. Hsp70 expression was measured in both organisms using quantitative real time PCR.

After the experiment algae counts were very low in the water for every anemone in all salinities, indicating little to no bleaching. Some anemones had curled tentacles, which shows that altered salinity does cause some physical stress, but it does not necessarily cause bleaching. *Aiptasia pallida* had significantly greater Hsps 70 expression at increased salinities, showing that higher salinities cause the most stress for anemones. Hsp70 expression in the symbionts did not vary significantly between salinities.

This research was funded by an award from the Hood College Summer Research Institute.

PURIFICATION OF VITELLOGENIN FROM NESTING LOGGERHEAD SEA TURTLES, *CARETTA CARETTA*

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The loggerhead sea turtle (*Caretta caretta*) is an endangered species that exists in ocean waters throughout the world. Although they nest in a variety of localities, the southeastern coast of the United States has one of the largest concentrations of loggerhead nests. Little is known about the physiology of this species, due to the fact that they spend most of their lives at sea. Knowledge of their reproductive physiology is needed to help determine how best to protect these turtles. Vitellogenin is the precursor protein to egg yolk, which provides nutrition for the developing young. Vitellogenin is produced prior to egg-laying, under the influence of estrogen. Information is sparse regarding vitellogenin levels of female loggerhead turtles. Such data can provide important insight into maternal resources, energy levels, nesting frequency, and offspring quality. We sought to purify vitellogenin from nesting female *C. caretta* for use in developing assays for this protein. Blood was collected by syringe from turtles nesting on the Atlantic coast. Plasma from the blood was subjected to protein assays and gel electrophoresis prior to vitellogenin purification. Purification involved multiple rounds of DEAE chromatography, with subsequent verification of vitellogenin's identity by gel electrophoresis and immunoassays. Presumptive purified loggerhead vitellogenin was revealed to have a molecular weight of approximately 200 kDa. It crossreacted with antibodies against vitellogenin in immunoassays, revealing that is was authentic vitellogenin. There were obvious differences in vitellogenin concentration among nesting females, indicating that assays for vitellogenin will be useful for studies of sea turtle reproductive physiology.

Afternoon Poster Session
Group EE - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

185. **SAY "BYE-O" TO FOSSIL FUELS AND HELLO TO BIOFUELS: EXPLORING THE PRODUCTION OF CELULOSIC ETHANOL IN THE INTERGRATED UNDERGRADUATE LABORATORY**

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Department of Biology, University of Delaware, 210 South College Avenue, Newark, DE 19716

186. **VALIDATION OF AN EPITOPE-TAGGED HAND1 KNOCK-IN ALLELE**

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187. **PERI- AND POST-NATAL IRON DEFICIENCY ALTERS ANXIETY-LIKE BEHAVIOR IN RATS**

Emily N. Spurlin, Timothy R. Monko, Brody J. Lipsett, Morgan N. Webb, and Erica L. Unger
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188. **JUVENILE HORMONE AND THE SPECFIICATION OF REPRODUCTIVE FATE IN PEA APHIDS**

Maho Okumura, Emily Spica, and Gregory Davis
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SAY "BYE-O" TO FOSSIL FUELS AND HELLO TO BIOFUELS: EXPLORING THE PRODUCTION OF CELULOSIC ETHANOL IN THE INTERGRATED UNDERGRADUATE LABORATORY

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This project aimed to develop two biology labs for the introductory integrated biology and chemistry classes. These labs will cover key concepts of biology (e.g. metabolism, enzyme chemistry, thermodynamics), while also providing an insight into practical applications of research by focusing on biofuels. Students will use plant-based products (e.g. cardboard, paper towels, maple leaves) as a source of cellulose (fiber) to produce sugar, glucose, which will then be fermented into ethanol. In comparison to current fuel sources, ethanol is a fuel source that releases less green house gases. After completion of this lab, students will be able to analyze the viability of ethanol as a renewable fuel source.

This project was made possible through the funding of the Howard Hughes Medical Institute.

VALIDATION OF AN EPITOPE-TAGGED HAND1 KNOCK-IN ALLELE

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The basic helix-loop-helix (bHLH) transcription factor Hand 1 (H1) is critical to proper cardiac development. H1 is robustly expressed in the left ventricle (LV). Hand factor mutations have been found in patients with congenital heart issues, such as hypoplastic left heart syndrome and ventricular septal disorders. H1 forms dimers with other bHLH transcription factors that are required for its DNA binding and can initiate changes in transcription of downstream target genes. Hand1 mRNA is first detected in embryonic day 8.5 (E8.5) mice at the base of the LV. An H1 systemic knockout causes embryonic lethality at E8.5 due to placental and embryonic abnormalities. Assessment of H1 LV function is restricted since identifying H1 transcriptional targets is limited at this developmental stage. Conditional H1 knockout in cardiomyocytes and Cre knock-in lineage analyses provide more insights, but the lack of a proficient antibody for H1 protein limits discovery of transcriptional targets to candidate gene approaches.

Alternatively, the lab engineered an epitope-tagged H1 allele (^{3XFLAG}*Hand1*) using zinc-finger nuclease technologies. The H1 sequence from gene-edited mice was PCR captured. Genotyping and sequence validation were performed to determine whether the gene was successfully combined. The desired PCR fragments were TA cloned and Sanger sequenced to confirm the presence of the three consecutive Flag epitope tag upstream and in frame with H1 codon 2. After validation, whole mount Flag-immunohistochemistry was conducted on *Hand1*$^{3XFLAG/Wt}$ E10.5 heterozygote mice to test antibody efficacy in detection of the modified ^{3XFLAG}*Hand1* allele. Comparisons of 3XFLAGHand1 flag protein immunodetection to wildtype H1 mRNA in-situ hybridizations show similar expression patterns in the pharyngeal arches and LV. Sequencing and immunohistochemistry showed successful incorporation of the triple flag tag and protein expression matches what is expected in E10.5 embryos. This work can help us to better understand the pathways and molecular basis behind heart development.

We would like to thank Indiana University-Purdue University Indianapolis and Indiana University School of Medicine Undergraduate Research for Prospective Physician-Scientists and Physician-Engineers.

PERI- AND POST-NATAL IRON DEFICIENCY ALTERS ANXIETY-LIKE BEHAVIOR IN RATS

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Iron deficiency (ID) affects an estimated 2 to 4 billion people worldwide. The presence of iron deficiency anemia in early childhood has been shown to negatively affect both cognitive and affective development. More recent evidence suggests that these behavioral outcomes may be irreversible, meaning that symptoms persist into adolescence and adulthood despite iron treatment. Behaviors associated with early ID include increased incidence of depression and anxiety and impaired memory; however, the neurobiological alterations that underlie these findings have not been clearly identified. This study in rats addressed two questions: 1) could the effects of dietary iron deficiency *in utero* or during lactation be repaired with dietary intervention; 2) is the serotonin neurotransmitter system altered due to early life iron deficiency. Iron interventions at postnatal day (P) 4, which approximates third trimester in humans in terms of overall brain growth, and at P21, which approximates human teenage years, were investigated.

Tests for novel object recognition memory, anxiety-like behavior and locomotor activity were performed. After euthanasia, the brains were harvested and dissected for quantitative analysis of proteins and neurotransmitters. There was a significant difference in anxiety-like behavior in both iron deficient treatment groups compared to the control. Novel object recognition memory and locomotor activity levels were not different between dietary treatment groups. Additionally, this study showed evidence that serotonin transporter protein levels are reduced in rats that experience early life iron deficiency. Overall, these results suggest that iron deficiency *in utero* or during lactation is associated with altered behavioral characteristics that may be associated with alterations in the serotonin neurotransmitter system.

This research was funded by the Paul Wolf Research Fund and Lebanon Valley College.

JUVENILE HORMONE AND THE SPECFIICATION OF REPRODUCTIVE FATE IN PEA APHIDS

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Pea aphids exhibit a seasonally induced polyphenism during their life cycle in which genetically identical females can be either sexual or asexual. Sexual females reproduce by laying fertilized eggs (oviparous sexual reproduction) while asexual females reproduce by allowing oocytes to complete embryogenesis within the mother without fertilization (viviparous parthenogenesis). Asexual reproduction is induced by short nights (summer) while sexual reproduction is induced by long nights (winter).

A widely held model entails an asexual-sexual switch, which is controlled by asexual-promoting juvenile hormone (JH), produced by the corpora allatum. This is supported by two pieces of evidence: JH levels and photoperiod are correlated (short nights give higher levels of JH) (Ishikawa et al., 2012) and JH is sufficient to specify asexual fate (Corbitt and Hardy, 1985). I replicated this latter result with a topical application of the JH analog, kinoprene.

Pea aphids are known to produce sexual offspring under long night conditions because their eggs are frost-resistant and able to survive the winter, and sexual reproduction is the only way to produce eggs. While some strains, such as the LSR1 (New York) strain can produce sexuals, some locations with mild climates are reported to have strains that only produce asexuals. One of those strains is the TUCSON strain from Tucson, Arizona (courtesy of Nancy Moran, U. of Texas, Austin). Previous students in the lab have confirmed that the TUCSON strain produces sexuals under extreme long nights (8L:16D) but suspecting that the TUCSON strain is only able to produce asexuals in Arizona, I attempted to prove that fact by placing them at the photoperiod of the Arizona winter solstice, 10L:14D. Identifying the difference in the responses to photoperiod will help me determine differences in sensitivity between the two strains.

My research was funded by award IOS-1051643 from the National Science Foundation. The Tucson strain was provided courtesy of Nancy Moran at the University of Texas, Austin.

Afternoon Poster Session
Group FF - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

189. **THE IMMUNOLOGICAL EFFECTS OF CESAREAN SECTIONS ON NEWBORNS IN SÃO PAULO, BRAZIL**

Isabella Gomes[1], Helio Junji Shimozako[2], Eduardo Massad[2], and C. Jessica Metcalf[3]
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190. **COMPARATIVE GENOMICS ANALYSIS OF SEVEN *DROSOPHILA* SPECIES**

Samuel Holechek and Susan Parrish
Department of Biology, McDaniel College, 2 College Hill, Westminster, MD 21157

191. **RNA-SEQ ANALYSIS OF SEXUAL- AND ASEXUAL-FATED *ACYRTHOSIPHON PISUM* EMBRYOS**

Gemma Johnson, Emily Spica, Joshua Shapiro, and Gregory Davis
Department of Biology, Bryn Mawr College, 101 N. Merion Avenue, Bryn Mawr, PA 19010

192. **SCORING SEQUENCE FOR MODELLED FOLDING CONFORMATION IN INTERACTIVE-ROSETTA USING HMMSTR**

Oluwadamilola Lawal[1,2], Christian Schenkelberg[2], Shounak Banerjee[2], Benjamin Walcott[2], and Christopher Bystroff[2]
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[2]Department of Biology, Rensselaer Polytechnic Institute, 110 8th Street, Troy, NY 12180

193. **SOCIAL MEDIA: A POTENTIAL TOOL TO UNDERSTAND AUTISM**

Gaurav Luthria[1], Jared Hawkins[2], and John Brownstein[2]
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[2]Department of Medicine Research, Boston Children's Hospital, 300 Longwood Avenue, Boston, MA 02115

194. **GENOMIC AND PROTEOMIC ANALYSIS OF CABRINIANS, A NOVEL *MYCOBACTERIUM PHLEI* BACTERIOPHAGE**

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THE IMMUNOLOGICAL EFFECTS OF CESAREAN SECTIONS ON NEWBORNS IN SÃO PAULO, BRAZIL

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While the World Health Organization recommends that national rates of Cesarean sections remain below 15%, a 2010 World Health report found that more than 6.2 million excess Cesarean sections were performed each year, with China and Brazil accounting for more than 50% of this statistic. Since then, Brazil's numbers, especially within the private sector, have pushed the country into having one of the world's highest rates of Cesarean deliveries — a procedure that, in medically unnecessary circumstances, has been associated with immunological deprivation in newborns.

Thus far, the majority of literature using Brazilian subpopulations has investigated neonatal outcomes, such as impaired intestinal bacterial colonization, as well as the development of later-in-life chronic outcomes, such as asthma and diabetes. The purpose of this research is to therefore contribute to the understanding of the more immediate immunological effects of Cesarean sections versus those of vaginal birth through a statistical analysis of newborn immunogram results.

These results were collected from the Instituto das Crianças at the Hospital das Clínicas — the University of São Paulo Medical School's central teaching hospital and the largest hospital in Latin America. By analyzing the relationship between mode of delivery and immunogram results, our findings provide statistical evidence that Cesarean sections impact the immediate immunological development of newborns, affecting neutrophil and lymphocyte counts within the first few months of life. These results are further modified by variables, such as gestational age, birthweight, mother's age at birth, previous deliveries and abnormalities during labor, such as premature membrane rupture and inadequate initial contractions.

I gratefully acknowledge the guidance and generosity of the Princeton-Brazil Global Fellows program and my research teams in the Department of Ecology and Evolutionary Biology at Princeton University and the Department of Legal Medicine at the University of São Paulo, without which the present study could not have been completed.

COMPARATIVE GENOMICS ANALYSIS OF SEVEN *DROSOPHILA* SPECIES

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The genus *Drosophila* is comprised of small flies colloquially known as fruit flies. In collaboration with the Genomics Education Partnership, we conducted a comparative genomics analysis of seven species of *Drosophila: D. melanogaster, D. yakuba, D. eugracillis, D. virilis, D. elegans, D. biarmipies and D. kikkawai.* Two adjacent genes, fuss and 4E-T, along with ~50,000 surrounding nucleotides, were analyzed. fuss is involved in the SMAD signaling protein pathway, which regulates cell growth and proliferation. 4E-T regulates mRNA degradation through poorly understood mechanisms. In the model organism *D. melanogaster*, these genes are located on chromosome 4, or the dot chromosome. Chromosome 4 in *Drosophila* is unusual as it is heterochromatic in nature, yet still contains gene rich regions. Using three lines of evidence: 1) gene prediction programs, 2) sequence conservation with *D. melanogaster*, and 3) experimental data, such as RNA-seq, both genes were annotated for exon and intron boundaries, allowing investigation of exon, intron and gene length across species. Conservation in exon length was found, while intron length was variable. The sixth intron of 4E-T in *Drosophila kikkawai* was 20-fold longer than the comparable intron in the other species. The surrounding genes of 4E-T and fuss was examined for synteny, revealing conserved gene order between 4E-T and fuss in six of the *Drosophila* species, but divergence in gene order was observed in *D. kikkawai*. Repeat analysis was performed for the seven species, revealing low GC content in those regions, as is characteristic of the dot chromosome. The larger intron in *D. kikkawai* 4E-T was shown to have a higher repeat density than other species. Further research will include building multiple sequence alignments to produce phylogenetic trees and examining conserved promoter motifs present between the species, to provide insight into the evolution of the *Drosophila* dot chromosome.

RNA-SEQ ANALYSIS OF SEXUAL- AND ASEXUAL-FATED *ACYRTHOSIPHON PISUM* EMBRYOS

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Acyrthosiphon pisum (the pea aphid) exhibits an interesting case of phenotypic plasticity: when exposed to short nights mothers produce embryos that will develop to be asexual, and when exposed to long nights they produce embryos that will develop to be sexual. The nature of the molecular switch that causes the embryos to develop as sexual or asexual is not well characterized, although juvenile hormone (JH) has been shown to be sufficient to induce asexual fate (Corbitt and Hardie 1985) and JH titers inversely correlate with night length (Ishikawa et al. 2012). While microarrays have been used to find candidate genes in late stage embryos (Gallot et al. 2010), a comprehensive study of differently aged embryos has not been undertaken. RNA-seq can be used to explore the nature of the reproductive polyphenism using an unbiased, non-candidate approach. To this end, RNA was collected from sexual or asexual fated embryos at three different stages of development: before specification, during specification, and during differentiation. An analysis pipeline using TopHat2, StringTie, and Ballgown was used to analyse the RNA sequences obtained. Since StringTie can be used to discover new transcripts, three different transcript assembly methods were used: 1) default settings, 2) more conservative settings, which restricted new transcript construction, and 3) assembling transcripts based only on annotated genes. Analysis of expression in sexual- and asexual-fated embryos in each stage and using all three transcript assembly methods shows that there are very few statistically significant differences between sexual- and asexual-fated embryos. This may be because deeper sequencing is needed for higher resolution. Additionally, many of the new transcripts assembled by StringTie may be spurious, as shown by a few individual cases.

Research was supported by Bryn Mawr College and the Howard Hughes Medical Institute.

SCORING SEQUENCE FOR MODELLED FOLDING CONFORMATION IN INTERACTIVE-ROSETTA USING HMMSTR

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Mutations introduced to an amino acid sequence during protein design may alter folding initiation sites thereby leading to folding inefficiency. I-sites (initiation sites) are short sequence-structure motifs used to predict local structures of a protein. HMMSTR is a hidden Markov model created using I-sites. Given backbone angles, HMMSTR can generate an amino acid probability profile at each position showing the propensity to maintain local structural conformations. The current study seeks to use HMMSTR in InteractiveROSETTA (a graphical user interface for ROSETTA protein modeling suite) to: 1) determine the probability for residues at each position for local protein folding, and 2) represent the folding probability for each residue as a color along a gradient.

The HMMSTR program source code was remodeled for compatibility with InteractiveROSETTA and imported as a shared object. Using Biopython, backbone atomic coordinates were extracted from the protein data bank file loaded into InteractiveROSETTA. These coordinates were passed to HMMSTR to generate an amino acid probability profile for folding using the forward-backward algorithm. These probabilities were used to calculate the log-likelihood ratio (LLR) for folding for the current residue at each position. LLR of -3 was the lowest and was represented as red while an LLR of 3 was the highest and represented as green. All LLR in between were colored along the red-green gradient.

HMMSTR in InteractiveROSETTA was tested on Green Fluorescent Protein sequence (2AWJ.pdb) that research has shown to not fold when the glycine is substituted for the tryptophan residue at position 57. A color change from green to yellow was observed at position 57 following the glycine substitution, indicating a reduction in folding probability.

This added functionality to InteractiveROSETTA will facilitate protein design by helping the user to discern and avoid non-folding (and presumably non-functional) mutations in an intuitive manner.

This project was funded by Rosetta Commons, NSF grant #1541278, and NIH grant R01 GM099827

SOCIAL MEDIA: A POTENTIAL TOOL TO UNDERSTAND AUTISM

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About one percent of the world's population has Autism Spectrum Disorder and its prevalence is increasing rapidly. Thus, there is a tremendous need to understand the heterogeneous nature of this disease. Data from social media can provide researchers with a first person, real-time perspective on an individual and can therefore offer a unique and effective solution to better understand behavioral disorders, such as autism. In this study, we evaluated whether social media platforms such as Twitter can be used as a tool to study autism. Twitter users who self-identify as having autism (test group) were compared to individuals from a control group. We saw that users from the test group had a higher friends:followers ratio and a lower lexical diversity than users from the control group. These findings are in accord with known symptoms of autism. The topic analysis using latent dirichlet allocation (lda) showed distinguishable topics between the two groups as well. Derived topics, along with other significantly different features between test and control groups were used to develop a predictive model using a linear support vector machine classification algorithm. The developed model had an accuracy of 83.5% indicating that social media is a powerful tool to help researchers understand autism. Therefore, social media provides an excellent substitute for expensive behavioral studies and can help develop therapeutic treatments for neurodevelopmental diseases such as autism.

This research was supported by the National Institute of General Medical Sciences, National Institutes of Health (NIGMS/NIH) under National Research Service Award T34 GM 008663 and Boston Children's Hospital.

GENOMIC AND PROTEOMIC ANALYSIS OF CABRINIANS, A NOVEL *MYCOBACTERIUM PHLEI* BACTERIOPHAGE

Rosendo Villafuerte-Vega , Andrew Conboy, Dylan Chudoff, and avid Dunbar
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Bacteriophages are viruses that infect and replicate within their specific host bacterium. We recently annotated the Mycobacteriophage Cabrinians genome using proteomics-based evidence. Cabrinians is the first phage isolated from the host *Mycobacterium phlei* with a genome closely related to many other Mycobacteriophage categorized as being in the F1 subcluster. Cabrinians genome is 56,669 base pairs in length containing 101 open reading frames (ORFs). Of these ORFs, we have evidence for the expression of 31 of them. Proteomics evidence also confirmed the start sites for 20 of the annotated ORFs. The most abundant expressed proteins are either virion structural or assembly proteins. Comparative proteogenomics is now being utilized to determine misannotation of homologous genes in related F1 subcluster Mycobacteriophage genomes.

We would like to thank Dr. David Dunbar of Cabrini College for his passion and being a great mentor, and Dylan Chudoff for his hard work in being the teaching assistant. We also would like to thank the SEPCHE institutions for funding the course, and Dr. Karen Snetslaar from Saint Joseph's University for taking time out of her schedule to take electron microscope pictures of the Mycobacteriophage, Cabrinians.

Afternoon Poster Session
Group GG - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

195. **ENDOCRINE CONTROL OF CARBOHYDRYTE METABOLISM IN *XENOPUS TROPICALIS*; TISSUE SPECIFIC EXPRESSION AND REGULATION OF GLUT2**

Mara Bezerko, Brooke Merchant, Yang Ding, and George Delahunty
Department of Biology, Goucher College, 1021 Dulaney Valley Road, Towson, MD 21204

196. **FUNCTIONAL ANALYSIS OF *DUF1* IN *SACCHAROMYCES CEREVISIAE***

Rachel Keuls, Brittney Lozzi and F. Les Erickson
Department of Biological Sciences, Salisbury University, 1101 Camden Avenue, Salisbury, MD 21801

197. **COPPER BIOREMEDIATION USING GENETICALLY ENGINEERED *ESCHERICHIA COLI***

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198. **APOE, IRON AND AMYLOID-β INTERACTIONS WITH BRAIN MICROGLIA**

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199. **CHARACTERIZATION OF PECTIN METHYLESTERASES FOR USE IN A SUGARBEET PULP BIOREFINERY**

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200. **THE ROLE OF NDT80 ACTIVATOR PROTEIN IN THE MEIOTIC COMMITMENT POINT OF AN IME-2 HYPERACTIVATED *S. CEREVISIAE* STRAIN**

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ENDOCRINE CONTROL OF CARBOHYDRYTE METABOLISM IN *XENOPUS TROPICALIS*; TISSUE SPECIFIC EXPRESSION AND REGULATION OF GLUT2

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These studies focus on gaining a better understanding of the endocrine control of carbohydrate metabolism, as vertebrates move from ectothermy to endothermy. In mammals, GLUT2 is a low affinity, bidirectional, glucose transporter utilized in the liver, kidney, pancreas, and intestine. Lower vertebrates may have distinct differences in carbohydrate and lipid metabolism due to their lower metabolic rate, reliance on anaerobic metabolism, and having lipid metabolism closely linked to reproductive processes. Using *Xenopus tropicalis* as an ectothermic species model, the tissue specific RNA expression of GLUT2 was characterized by RT-PCR using primers designed via the NCBI database. Expression of GLUT2 was normalized to the RPL8 gene expression levels. *Xenopus tropicalis* demonstrated GLUT2 expression similar to mammalian tissues, but additionally expressed GLUT2 in adipose tissue, stomach, and testis. Gender differences were indicated by expression of GLUT2 only in female adipose tissue, and expression of GLUT2 only in the testis, not the ovaries. Glucose injection experiments were performed on females to determine the tissue specific RNA transcript response to increased blood glucose levels. Preliminary results suggest an increase in RNA expression in the pancreas and adipose tissues.

Future research will involve studying tissue specific RNA expression in both males and females in response in elevated blood glucose and insulin levels. In addition, preliminary studies are underway to characterize GLUT2 protein expression using fluorescent immunohistochemistry studies; employing polyclonal antibodies to human GLUT2. This data will provide a comparative window to enhance our understanding of mammalian metabolic regulation, and lend further insight to today's obesity and type II diabetes epidemic.

This work is supported by the Seibert Fund of the Goucher College Summer Research Program.

FUNCTIONAL ANALYSIS OF *DUF1* IN *SACCHAROMYCES CEREVISIAE*

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Duf1 is a highly conserved, WD40-repeat protein which interacts with deubiquitinases (DUBs). DUBs carry out the process of deubiquitination, the cleavage of peptide bonds between ubiquitin and its substrate, thus orchestrating the regulatory step of the ubiquitin system. Homologues of *DUF1* have been shown to form complexes with their DUBs and have a variety of important functions. In humans, WDR48 forms a tertiary complex with UAF1 and USP1, USP12, or USP46 in order to deubiquitinate key subunits in the Fanconi anemia DNA repair pathway. In Arabidopsis LRS1 interacts with Ubp3 and Ubp4 to stimulate lateral root growth.

Research has shown that in *Saccharomyces cerevisiae*, Duf1 interacts with DUBs involved in the biosynthesis of mitochondrial subunits necessary for respiration. When grown at 37 ˚C, a stressful environment for yeast propagation, a respiratory phenotype of petite colonies was observed. Thus, we hypothesize that Duf1 enhances the yeast's ability to cope with stress. Our interest lies in determining which cellular stress pathways require the regulation that Duf1 provides for continued yeast propagation.

By exposing a *DUF1* knockout mutant strain to different stress conditions and comparing its growth to the wild type strain, a phenotype may be revealed to provide a better understanding of the cellular activities of Duf1. Cell growth experiments were performed in 96-well plates using a Spectramax i3 plate reader and the OD_{600} was plotted against time. Doubling times were computed and significance was determined using a two sample t-test assuming unequal variances with 95% confidence.

Thus far, when grown in minimal media, the knockout mutant strain experienced faster growth than the wild type strain. Inversely, when minimal media was supplemented with increasing amounts of ethanol, the wild type prevailed, leading us to believe that the involvement of Duf1 is primarily when the cell is placed under multiple stress conditions simultaneously.

We would like to thank the Henson Undergraduate Research Grant and NSF for providing funding to purchase necessary materials and equipment to conduct our research.

COPPER BIOREMEDIATION USING GENETICALLY ENGINEERED *ESCHERICHIA COLI*

Pranesh Navarathna[1], May Li[1], UMBC iGEM team[2], Cynthia Wagner[3], and Stephen Freeland[4]
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Copper is a major pollutant in a variety of freshwater ecosystems. When copper is oxidized from Cu^{+} to Cu^{2+}, it often produces a free radical known as a reactive oxygen species (ROS), which is capable of severely damaging biological molecules. *E. coli* have the ability to uptake copper, but after a certain threshold, the copper becomes toxic to the cell. Due to the toxicity of copper, *E. coli* quickly saturate and are unable to uptake more than a small amount of copper.

Our goal is to increase the efficiency of copper uptake in *E. coli* for the purpose of bioremediation in freshwater ecosystems. We engineered *E. coli* to express the yeast CUP1 gene in an attempt to increase copper tolerance. CUP1 encodes a metallothionein protein that binds 11 copper atoms, thereby preventing formation of the ROS. In addition, metallothionein detoxifies hydroxyl radicals with its cysteine groups.

Through the use of growth curves and an assay to measure copper uptake, we will present our preliminary data on *E.coli* transformed with the CUP1 gene as our initial attempt to create a bacterial strain that has a higher resistance to copper toxicity.

The UMBC iGEM Team would like to acknowledge the UMBC Student Government Association, the UMBC College of Natural and Mathematical Sciences, UMBC Biology Department and UMBC Chemistry and Biochemistry Department for their financial support without which our project would not be feasible. We would also like to acknowledge Dr. Stephen Mang for his support with copper measurement.

APOE, IRON AND AMYLOID-β INTERACTIONS WITH BRAIN MICROGLIA

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Alzheimer's disease is the most common form of dementia, affecting the lives of 5 million Americans. This neurodegenerative disease is associated with the presence of amyloid plaques in the brain. Amyloid-β (Aβ) is normally cleared by microglia cells, which become activated and phagocytose Aβ oligomers. When activated, microglia produce nitric oxide, cytokines and other pro-inflammatory mediators. Chronic microglial activation promotes neuronal cell damage and death by these mediators. Under these conditions, a decrease in Aβ receptors also occurs and plaque formation is enhanced. A role for both iron and apolipoprotein E (ApoE) in Aβ interactions with microglia has been proposed, and these interactions may help to limit their activation and alter Aβ clearance. While iron binds to Aβ, its possible interactions with ApoE remain to be characterized. To study ApoE iron-binding and Aβ-induced microglia activation, this project had three goals: 1) express and purify recombinant ApoE, 2) use isothermal calorimetry (ITC) to examine iron binding to ApoE, and 3) examine the effects of Aβ, iron and ApoE on M1 activation of immortalized microglia (IMG) cells using the Griess Assay to detect NO production. The results concluded that ApoE was successfully inserted into a bacterial plasmid, iron has an affinity to ApoE causing an exothermic reaction, and Aβ induces a pro-inflammatory response which is exacerbated by iron, but an ApoE iron complex quenches this pro-inflammatory response.

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CHARACTERIZATION OF PECTIN METHYLESTERASES FOR USE IN A SUGARBEET PULP BIOREFINERY

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Biofuels are a renewable alternative to fossil fuels and since the carbon dioxide released upon burning biofuels is recaptured during the growth of the next season's crops, they do not contribute excess carbon dioxide like fossil fuels do when burned. First generation biofuels use starch (corn) or sugar (sugarcane) to produce fuel. The use of these food commodities contributes to a rise in food prices and its own set of environmental problems. However, second generation biofuels are derived from agricultural wastes (ie. corn stover, sugar beet pulp) to extract sugars from the structural components of plants, which can eventually be fermented into ethanol and be used as fuel.

The plant cell wall that constitutes the structural component of the plant includes cellulose, hemicellulose, and pectin polymers. These cellulosic structural plant components have evolved to be resistant to break down, therefore making it difficult to get monomeric sugars from these plant components. However, various bacteria have enzymes that are capable of breaking down these components. Our study compares the properties of different bacterial pectin methylesterases (PMEs), which catalyze the hydrolysis of methylesters from pectin making subsequent digestion of this key polymer by pectate lyases and hydrolases much more efficient. Specifically we cloned, expressed and purified the PMEs from *Serratia sp. 39006*, *Yersinia enterolitica subsp. enterolitica*, *Pectobacterium atrosepticum* and *Pectobacterium wasabiae* and compared the properties of these enzymes to the well characterized pmeA from *Dickeya dadantii 3937*. The temperature range, pH range and specific activities of all of the enzymes were determined. Interestingly, despite having the same protein fold and close sequence similarity to some of the other proteins, the PME from *P.atrosepticum* is the only one to refold to an active conformation following heat denaturation. Further characterization will provide insight into which PMEs have characteristics best-suited for use in biofuel production.

THE ROLE OF NDT80 ACTIVATOR PROTEIN IN THE MEIOTIC COMMITMENT POINT OF AN IME-2 HYPERACTIVATED *S. CEREVISIAE* STRAIN

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In yeast, glucose starvation is the key signal that causes diploid cells to enter meiosis resulting in spore formation. The Sum1 repressor is displaced in part due to phosphorylation by IME-2 kinase. Subsequently, Ndt80 activator induces exit from prophase I resulting in meiotic development. If nutrients are added prior to the completion of prophase I, cells will exit the meiotic program and return to mitotic growth. This is controlled by Gpa2p, a G-binding protein that binds to the carboxyl terminal end of IME-2 kinase and inhibits sporulation. In this study, we use an IME-2 hyperactivated strain with a truncated C-terminal regulatory domain. In addition, we use an estradiol inducible promoter to regulate Ndt80 expression and trap cells in meiotic prophase. Here, we examine the effect of simultaneous addition of glucose and estradiol at the commitment point. The results indicate that cells return to mitotic growth and that the IME2 hyperactivated strain is unable to override inhibition of meiosis by glucose.

Afternoon Poster Session
Group HH - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

201. **REGULATION OF RECOMBINATION ACTIVATING GENE 2 (RAG2) IN RESPONSE TO DNA DAMAGE**

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202. **EFFECT OF DIETARY SELENIUM AND THE 15KDA SELENOPROTEIN IN A MODEL OF INFLAMMATORY COLITIS**

Katie Garrett[1], Kristin Peters[1], Janelle Hartman[1], Jessica Canter[1], Bradley Carlson[2], Vadim Gladyshev[3], Yunkai Yu[4], Liang Cao[4], Cindy Davis[5], Dolph Hatfield[2], and Petra Tsuji[1]
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203. **TARGETING MITOCHONDRIAL BIOGENESIS TO OVERCOME INTRINSIC AND ACQUIRED DRUG RESISTANCE TO MAPK PATHWAY INHIBITORS**

Omotayo Ope,[1,6] Gao Zhang,[1] Lawrence Wu,[1] Dennie T. Frederick,[2] Zhi Wei,[3] Young Chan Chae,[1] Xiaowei Xu,[4] Clemens Krepler,[1] Gordon B. Mills,[5] Dario C. Altieri,[1] Keith T. Flaherty,[2] and Meenhard Herlyn[1]
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204. **DICHOTOMY IN THE EPIGENETIC MARK LYSINE ACETYLATION IS CRITICAL FOR THE PROLIFERATION OF PROSTATE CANCER CELLS**

Zimuzoh Orakuae[1], Ambreka Benson[1], Marc Philizaire[1], Ravi Pathak[2] and Shiraz Mujtaba[1]
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205. **GLYCOCHENODEOXYCHOLATE, A CANDIDATE PROGNOSTIC MARKER OF BREAST CANCER PATIENT SURVIVAL, INHIBITS BREAST CANCER CELL PROLIFERATION**

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206. **OBESITY AND YOUNG ONSET COLORECTAL CANCER: A REVIEW**

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REGULATION OF RECOMBINATION ACTIVATING GENE 2 (RAG2) IN RESPONSE TO DNA DAMAGE

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Adaptive immunity requires the production of many unique lymphocyte antigen receptors that are highly specific, yet robust in aggregate coverage. The recombination-activating gene (RAG) protein complex, a heterotetramer of RAG1 and RAG2, creates pairs of DNA double strand breaks (DSBs) at designated sites in antigen receptor loci of B cells and T cells. Once cleaved, widely-dispersed antigen receptor gene segments are recombined and assembled to generate complete antigen receptor genes. RAG is thus critical for conferring adaptive immunity against a diverse array of antigens. Regulation of RAG activity is important, as aberrant recombination can result in lymphomagenic translocations. In the presence of RAG-independent DSBs, the chance of translocations involving RAG-induced DSBs increases. We aim to further understand how RAG activity is affected by DNA damage, within the context of DSBs induced by ionizing radiation (IR) in mouse primary pre-B cells. Preliminary results show that DNA damage causes decreased RAG1 transcription and protein levels. In contrast, while RAG2 transcription is lost, protein levels remain steady. This study seeks to determine the mechanism(s) of RAG2 protein persistence. First, I found the half-life of endogenous RAG2 to be longer than four hours, a result which has not been reported in primary lymphoid cells. I also investigated the phosphorylation state of RAG2 following IR through immunoblotting and co-immunoprecipitation, though the data were inconclusive.

I would like to acknowledge the University of Pennsylvania Perelman School of Medicine Biomedical Graduate Studies for funding my research project.

EFFECT OF DIETARY SELENIUM AND THE 15KDA SELENOPROTEIN IN A MODEL OF INFLAMMATORY COLITIS

Katie Garrett[1], Kristin Peters[1], Janelle Hartman[1], Jessica Canter[1], Bradley Carlson[2], Vadim Gladyshev[3], Yunkai Yu[4], Liang Cao[4], Cindy Davis[5], Dolph Hatfield[2], and Petra Tsuji[1]
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Selenium is a trace element that is nutritionally essential to mammals and is incorporated into selenoproteins via selenocysteine. Selenoproteins are involved in cell homeostasis, including DNA repair and apoptosis. Previous research has shown that mice lacking the 15 kDa selenoprotein (Sep15) are protected against preneoplastic colon lesions and have an increased expression of genes related to interferon-γ-regulated inflammation.

Sep15 knockout mice and littermate controls were administered a selenium-deficient (0.04 ppm) or selenium-adequate (0.1 ppm) diet in the form of selenite during the course of the study. Additionally, the animals were either exposed to 2% dextran sulfate sodium in the drinking water for one week to induce inflammatory colitis, or maintained on water as untreated reference groups. Colon epithelia, serum, and other tissues were collected from each animal to examine the role of dietary selenium and Sep15 in the inflammatory response. The mRNA expression of genes known to be involved in immune and pro-inflammatory responses in both Sep15 knockout and control mice were compared by means of quantitative RT-PCR and microarray analyses. Preliminary results suggest that selenium-deficiency may contribute to increased expression of interferon-γ-regulated gene expression, as expected. Mice lacking Sep15 appear to have an enhanced expression of interferon-γ-regulated genes than littermate controls, independent from the selenium provided in the diet. Pro-inflammatory serum cytokines are currently being analyzed using a Mesoscale multiplex assay to further elucidate the contribution of Sep15 on the expression of pro-inflammatory genes.

Supported by the NIH Office of Dietary Supplements, Towson University's Fisher College of Science and Mathematics, and Jess and Mildred Fisher Endowed Chair funds (P. Tsuji).

TARGETING MITOCHONDRIAL BIOGENESIS TO OVERCOME INTRINSIC AND ACQUIRED DRUG RESISTANCE TO MAPK PATHWAY INHIBITORS

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Targeting multiple components of the mitogen-activated protein kinase pathway can prolong the survival of $BRAF^{V600E}$ melanoma patients, but this approach is not curative. Some $BRAF^{V600E}$ melanoma cells are intrinsically resistant to MAPK pathway inhibitors, impeding the efficacy of targeted therapies. At the systemic level, our knowledge of how signaling pathways underlie both intrinsic and acquired drug resistance needs to be expanded.

Acquired drug resistant melanoma cells were established in vitro after the prolonged drug exposure. cDNA samples and FFPE slides were derived from paired pre-treatment, early on-treatment and progressive tumor biopsies from patients with metastatic melanoma who received the treatment of MAPK pathway inhibitors. Real-time oxygen consumption rates were measured by using Seahorse Bioscience XF24 Extracellular Flux Analyzers. Time-course gene expression microarray and reverse phase protein array (RPPA) experiments were conducted using intrinsically drug resistant melanoma cells. 1205Lu melanoma xenografts were treated with the BRAF inhibitor, PLX4720, tumor metabolism inhibitors, 2,4-DNP or Gamitrinib or the combined use of these inhibitors.

These intrinsically drug resistant cells exploited an integrated stress response, exhibited an increase in mitochondrial DNA content and required oxidative phosphorylation to meet their bioenergetic needs. Genetically silencing TFAM or TRAP-1 or pharmacologically targeting TRAP-1 with Gamitrinib significantly augmented the targeted therapy response by inducing mitochondrial dysfunction, which was mediated by the increase in reactive oxygen species (ROS). Our study establishes mitochondrial biogenesis coupled with aberrant tumor bioenergetics not only as a novel therapy escape mechanism but also a tractable therapeutic target.. Therefore, our study paves the way for a rationale-based combinatorial strategy to overcome both intrinsic and acquired drug resistance.

DICHOTOMY IN THE EPIGENETIC MARK LYSINE ACETYLATION IS CRITICAL FOR THE PROLIFERATION OF PROSTATE CANCER CELLS

Zimuzoh Orakuae[1], Ambreka Benson[1], Marc Philizaire[1], Ravi Pathak[2] and Shiraz Mujtaba[1]
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The dynamics of lysine acetylation serve as a major epigenetic mark, which regulates cellular response to inflammation, DNA damage and hormonal changes. Microarray assays reveal changes in gene expression, but cannot predict regulation of a protein function by epigenetic modifications. The present study employs computational tools to inclusively analyze microarray data to understand the potential role of acetylation during development of androgen-independent PCa. The data revealed that the androgen receptor interacts with 333 proteins, out of which at least 92 proteins were acetylated. Notably, the number of cellular proteins undergoing acetylation in the androgen-dependent PCa was more as compared to the androgen-independent PCa. Specifically, the 32 lysine-acetylated proteins in the cellular models of androgen-dependent PCa were mainly involved in regulating stability as well as pre- and post-processing of mRNA. Collectively, the data demonstrate that protein lysine acetylation plays a crucial role during the transition of androgen-dependent to -independent PCa, which importantly, could also serve as a functional axis to unravel new therapeutic targets.

GLYCOCHENODEOXYCHOLATE, A CANDIDATE PROGNOSTIC MARKER OF BREAST CANCER PATIENT SURVIVAL, PROMOTES BREAST CANCER CELL PROLIFERATION.

Sandra Reyes[1], Wei Tang[2], Tiffany Dorsey[2] and, Stefan Ambs[2]
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Otto Warburg first observed cellular metabolism aberrations in cancer. Since his discoveries, multiple studies established a link between oncogenic pathways that drive tumorigenesis and alterations in tumor metabolism that are induced by these pathways. Breast tumors acquire changes in metabolism during disease development and progression. We conducted an analysis into the relationship between metabolite abundances in breast tumors and patient survival, and linked increased abundance of glycochenodeoxycholate (GCDC), a bile acid, in breast tumors to improved survival in our cohort. We further performed a fluorometric assay to access the effect of GCDC on breast cancer cell viability and proliferation. A reduction of cancer cell viability was observed with Tamoxifen (Tam) treatment, which we used as a positive control. Next, we found that GCDC also promotes cell growth and proliferation. Based on our findings, we propose that GCDC promotes breast cancer growth and its tissue level is a prognostic metabolite for the disease.

This was supported and funded by Cancer Research Interns Summer Program, NCI at NIH in Bethesda, and Laboratory of Human Carcinogenesis.

OBESITY AND YOUNG ONSET COLORECTAL CANCER: A REVIEW

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Colorectal cancer is the third most common cancer and the third leading cause of cancer death in both men and women in the United States, according to the American Cancer Society. Incidence and mortality have been decreasing in individuals 50 years of age and older, largely due to screening. However, incidence and mortality among individuals under age 50 has increased approximately 2% per year over the past two decades and is predicted to climb. The majority of cases are sporadic, suggesting behavioral and environmental factors may play a role. A well-established risk factor for colorectal cancer is obesity, defined as having body mass index of 30 kg/m^2 or greater. In the past three decades, childhood and adolescent obesity has drastically increased. Data explaining a potential association between obesity and young onset colorectal cancer are limited. To evaluate this relationship, we conducted a literature review of obesity and subsequent development of colorectal cancer in individuals under 50 using the PubMed and Medline databases. Our final analysis included 12 articles, which studied a wide range of age groups, with only one focusing exclusively on those under 50. A positive association was described in 11 articles, while one article reported a negative association. In addition, we collected data from the Behavioral Risk Factor Surveillance System and the New York State Cancer Registry to analyze trends in New York State. We concluded that early life obesity may be associated with increased risk of colorectal cancer in individuals under age 50. However, data are limited in this age group. In addition, we found that New York State trends parallel national trends, but they differ when stratified by race/ethnicity. This information is the first step in establishing obesity as a potential contributor to the national increase in young onset colorectal cancer incidence.

I would like to acknowledge the 2015 Columbia Summer Institute for Training in Biostatistics, funded by NIH grant 5 T15 HL117444-03. Research relating to this abstract was also funded by a National Cancer Institute R25T CA094061-12 Cancer Epidemiology Training Grant (PI: Alfred Neugut).

Afternoon Poster Session
Group II - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

207. **DIFFERENTIATION OF EMBRYONIC STEM CELLS CULTURED ON EXTRACELLULAR MATRIX COATD SURFACES**

Crystal Abrams, Ijaz Ahmed and Alam Nur-E-Kamal
Department of Biology, Medgar Evers College of the City University of New York, 1638 Bedford Avenue, Brooklyn, NY 11225

208. **THE FUNCTION OF RED1 AND MMI1 IN *S. POMBE* MEIOTIC GENE SILENCING**

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209. **CYTOSKELETON ASSOCIATED PROTEINS INVOLVED IN LENS FIBER CELL ELONGATION**

Troy Rubenstein, Dylan Audette, and Melinda Duncan
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210. **CHARACTERIZATION OF THE MUTATIONAL LANDSCAPE OF BREAST CANCER LUNG METASTASES BY NEXT GENERATION SEQUENCING**

Jack Sanford[1,2], Karol Szczepanek[1], Ngoc-Han Ha[1], and Kent W. Hunter[1]
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211. **BONE PAIN FROM METASTATIC PROSTATE CANCER MAY BE MEDIATED THROUGH PURINERGIC SIGNALING**

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DIFFERENTIATION OF EMBRYONIC STEM CELLS CULTURED ON EXTRACELLULAR MATRIX COATD SURFACES

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Mouse embryonic stem (ES) cells are permanent cell lines derived from the inner cell mass of mammalian blastocysts. Mouse ES cells possess the ability to grow in cell culture condition and differentiate into all types of body cells under appropriate conditions. However, a chemically defined condition for the growth and differentiation of ES cells needs to be developed. Previously, we have studied the ability of synthetic polyamide nanofibers to support the growth of mouse ES cells. The polyamide nanofibers mimic the three-dimensional nanofibrillar topology afforded by the extracellular matrix (ECM). Therefore, they may provide a more physiologically relevant substrate for cell growth than the two-dimensional glass or plastic surfaces normally employed to culture cells. We found that mouse ES cells adhere and grow on both glass and nanofiber surfaces. In this study we demonstrated an enhanced rate of proliferation on nanofibers. We also found that Rac GTPase-PI3 kinase is involved in proliferation of ES cells on nanofibrillar surfaces. We have covalently attached growth factors and ECM-derived peptides with 3D nanofibers and found increased proliferation. We also found induction of differentiation into neuronal cells. Both GFAP and nestin gene expression were induced as determined by Western blotting, immunocytochemistry and RT-PCR. The results suggest that growth and differentiation of stem cells could be directed by growth factors and/or ECM derived peptides.

THE FUNCTION OF RED1 AND MMI1 IN *S. POMBE* MEIOTIC GENE SILENCING

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Online access of this abstract is restricted at the request of the Principal Investigator.

CYTOSKELETON ASSOCIATED PROTEINS INVOLVED IN LENS FIBER CELL ELONGATION

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The lens is comprised of two cell types; lens epithelial cells cap the anterior of the lens and differentiate at the equator into elongated lens fibers which fill the center of the lens. While the process that drives fiber cell elongation is not completely understood, prior studies demonstrate that the cytoskeleton is vital for this event. Our prior work analyzed gene expression in a fiber cell elongation deficient mouse model as compared to wild type, and identified several cytoskeletal associated genes whose expression was down regulated. Among these were the actin nucleation factor Spire1, the atypical microtubule subunit Tubb6, the end binding protein Mapre3, and the lens specific kinesin, Kif1a. We characterized the expression of these four genes in wild type lenses using a fluorescent or immunohistochemistry stain. The results show that in wild type lens, the proteins are expressed within the elongating fiber cells, and in the fiber cell deficient model there is a marked decrease in expression. This data supports the hypothesis that these genes are involved in the morphological differentiation of lens fiber cells. We plan on furthering our investigation to demonstrate that these cytoskeletal associated genes are essential for fiber cell elongation by knocking inhibiting cytoskeletal dynamics and observing if fiber cell differentiation still occurs.

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CHARACTERIZATION OF THE MUTATIONAL LANDSCAPE OF BREAST CANCER LUNG METASTASES BY NEXT GENERATION SEQUENCING

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Breast cancer is one of the leading causes of cancer-related mortality of women in the United States. The majority of cancer deaths are due to metastases rather than the primary tumor; thus, a better understanding of the mechanisms that lead to metastatic disease is critical in reducing mortality rates. The current model of metastasis suggests that cancer is a selective process. This means that the majority of cancer cells that leave primary tumor are either removed by immune system or fail to survive in distant organs. However, some cancer cells acquire molecular features that allow them to survive and proliferate in secondary sites, such as lungs, liver, brain, and bones.

The goal of this project is to characterize the genomic and transcriptomic landscapes of breast cancer metastases from lungs to identify potential genes that drive the metastasis process (metastasis promoters) or inhibit it (metastasis suppressors). This was done by isolating DNA and RNA from the primary breast tumor, background lung tissue, and lung metastases from dissected Polyomavirus middle-T antigen (PyMT) mice. This animal models spontaneous and highly aggressive human breast cancer. Deep sequencing of these DNA and RNA samples should reveal genes that are highly expressed in metastases and enable cancer cells to metastasize. These genes can then be explored for the development of targeted therapies.

BONE PAIN FROM METASTATIC PROSTATE CANCER MAY BE MEDIATED THROUGH PURINERGIC SIGNALING

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Prostate cancer (PCa) is the second leading cause of cancer-related deaths in men in the US. Bone is the primary site of PCa metastasis, occurring in over 60% of PCa patients. These patients experience severe cancer-induced bone pain (CIBP) that significantly reduces the quality of life of the patient. Current therapeutics for CIBP are minimally effective as the exact mechanism behind this pain is not well understood. PCa metastatic tumors release high levels of ATP that we postulate is increased with mechanical loading of bone to stimulate nociceptive neurons through activation of the P2X3 purinergic receptor.

C4-2B4, a highly-metastatic PCa cell line, was mechanically stimulated via hypotonic swelling and the resulting conditioned media (CM) collected immediately or at 5 or 10 minutes post-hypotonic challenge. The CM was then applied to primary dorsal root ganglia (DRG) neurons and changes in intracellular calcium ($[Ca^{2+}]_i$) measured using the fluorescent calcium chelator, Fluo 4. This change in $[Ca^{2+}]_i$ was compared to changes in $[Ca^{2+}]_i$ in response to exogenous addition of 0.1, 1, 5, and 10μM ATP. Addition of control media to DRG failed to increase $[Ca^{2+}]_i$. However, CM from hypotonically swelled C4-2B4 cells produced an increase in $[Ca^{2+}]_i$ similar to the response to 5 to 10 μM ATP. This could be blocked when apyrase was added to the CM to hydrolyze ATP. To determine if P2X3 was the main regulator of DRG response to CM from PCa cells, DRG were pretreated with the P2X3 competitive inhibitor, TNP-ATP. TNP-ATP inhibition failed to significantly block the response of DRG to CM from C4-2B4 cancer cells.

These results suggest that ATP released from PCa cells during loading can affect nerve signaling. However, the ineffectiveness of P2X3 block suggests another P2 receptor is involved. These data point to a potential therapeutic target to reduce CIBP in PCa patients.

Afternoon Poster Session
Group JJ - Biological Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

212. **FUNCTIONAL CHARACTERIZATION OF A NOVEL BTB/POZ DOMAIN ZINC FINGER TRANSCRIPTION FACTOR ZBTB8B IN MAMMALIAN LENS DEVELOPMENT**

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213. **EFFECT OF ECCENTRIC EXERCISE ON FASL IN PBMCS**

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214. **PREDICTING MAXIMALLY INFORMATIVE FUTURE EXPERIMENTS FROM EXISTING REPOSITORIES OF GENE EXPRESSION**

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215. **USING THE AMBORELLA TRICHOPODA EXPANSIN SUPERFAMILY TO ELUCIDATE THE HISTORY OF ANGIOSPERM EXPANSINS**

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216. **A COMPARISON OF PLANKTONIC COMMUNITIES AT THE MURPHY'S BOTTOM ECOLOGICAL SITE**

Youstina Seliman, Caroline Kirby and Brady Porter
Department of Biological Sciences, Duquesne University, Pittsburgh PA 15282

217. **PATTERNS OF TORSOLIKE AND ACTIVATED MAP KINASE DURING OVIPAROUS DEVELOPMENT IN THE PEA APHID**

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FUNCTIONAL CHARACTERIZATION OF A NOVEL BTB/POZ DOMAIN ZINC FINGER TRANSCRIPTION FACTOR ZBTB8B IN MAMMALIAN LENS DEVELOPMENT

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Mutations in genes that function in early eye development are known to cause human birth defects such as congenital cataracts. Therefore, we aimed to apply a bioinformatics eye gene discovery tool called *iSyTE* along with an animal model to identify a new regulatory gene in early eye development. *iSyTE* predicted a novel zinc finger transcription factor gene *Zbtb8b* that exhibits enriched expression in eye tissue beginning from an early developmental stage at mouse embryonic (E) day 9.5. Whole mount and section *in situ* analyses confirmed highly enriched *Zbtb8b* mRNA expression in mouse lens development. *Zbtb8b* mRNA is expressed in the presumptive eye region in E9.5 mouse embryos and becomes progressively lens specific in later stages, being enriched in the anterior epithelium of the lens by E12.0. Immunofluorescence using an anti-Zbtb8b polyclonal antibody demonstrated that Zbtb8b protein is expressed in the lens placode at E9.5 prior to being localized to the anterior epithelium of the lens by E13.0.To investigate its function in lens development, *Zbtb8b* was transiently knocked down (KD) using siRNA in the mouse lens epithelial cell line 21EM15. RNA was extracted from scrambled siRNA transfected control and *Zbtb8b*-KD cells, and transcript levels of important lens genes were analyzed by reverse transcriptase quantitative PCR (RT-qPCR). RT-qPCR analysis demonstrated that several eye defect-linked genes such as *Mab21l1*, *Cdh1*, *Pknox1* and *Meis1* are down-regulated in *Zbtb8b*-KD cells. In sum, we identify a new transcription factor Zbtb8b that functions in lens development by controlling key early genes associated with mammalian eye defects.

EFFECT OF ECCENTRIC EXERCISE ON FASL IN PBMCS

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Regular physical activity increases exercise capacity, endurance, and skeletal muscle strength. In addition, regular physical activity prevents the onset of cardiovascular disease. It is suggested that individuals should engage in at least 30 minutes of moderate-intensity exercise on most days of the week (1). Eccentric physical activity is more damaging than concentric physical activity (2), and recent findings suggest that apoptosis plays an important role following eccentric exercise (3). The focus of this study was on the FasL gene which has a crucial role in cell death or apoptosis via binding and clustering to Fas (CD95). Apoptosis through Fas is important for maintaining peripheral immune tolerance and inflammatory responses (4).

The aim of this study was to test the effects of an acute bout of eccentric exercise on FasL expression and compare FasL expression in Peripheral Blood Mononuclear Cells (PBMCs) before and after exercise. The hypothesis was that downhill running would increase FasL mRNA; therefore, resulting in an upregulation of the FasL gene after a 30 minute bout of eccentric exercise. To determine changes in gene expression, young and healthy participants (n=3) completed 30 minutes of downhill running at a 15% decline. This acute bout of exercise was done at a speed that elicited 70% of the participant's previously determined VO_2max. Venous blood samples were taken immediately before and after exercise. RNA was extracted from PBMCs and RT-quantitative PCR was performed to determine FasL expression before and after exercise. FasL mRNA was not upregulated when comparing gene expression before (0.456 ± 0.0221 fold) and after exercise (0.452 ± 0.0296 fold) after normalizing for 18s expression; there was no significant difference ($p = 0.280$). Perhaps apoptosis does not play an important role in the inflammatory response after muscle damage caused by eccentric exercise.

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PREDICTING MAXIMALLY INFORMATIVE FUTURE EXPERIMENTS FROM EXISTING REPOSITORIES OF GENE EXPRESSION

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When considering large databases of genome-wide gene expression data, we have noted that many experiments show similar patterns of gene expression, such that many newly added experiments tend to be limited in their ability to provide substantially new insights into genome-wide metabolic and regulatory behavior. We have recently developed a method to predict experimental conditions which will be substantially different than existing gene expression data, and, thus, have the potential to provide maximal amounts of information about the regulatory and metabolic behaviors of hypothetically annotated genes and poorly understood pathways. In our method, we first utilize a metric to determine which genes have consistently low expression values across all experimental conditions, suggesting the genes are rarely, if ever, activated in the current set of experiments. We then utilize integrated regulatory and metabolic models to predict what type of experiments will activate these genes. We will present results from application of our method to large repositories of gene expression for *E. coli* and *S. oneidensis*.

We acknowledge that this work was funded by the National Science Foundation (MCB 1330734) and National Institutes of Health (R15-HG006915).

USING THE AMBORELLA TRICHOPODA EXPANSIN SUPERFAMILY TO ELUCIDATE THE HISTORY OF ANGIOSPERM EXPANSINS

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Expansins are a superfamily of proteins found in plants that assist in cell wall loosening during growth and development. The superfamily is divided into four families: EXPA, EXPB, EXLA, and EXLB (Sampedro and Cosgrove 2005). Previous work on *Arabidopsis thaliana*, rice, and *Populus trichocarpa* has clarified the evolutionary history of expansins in angiosperms (Sampedro et al. 2005). *Amborella trichopoda* is a very early diverging flowering plant. Thus, it is a sister lineage to all other extant angiosperms (Amborella Genome Project 2013). Because of this relationship, comparing the *A. trichopoda* expansin superfamily to those of other flowering plants can suggest which expansin genes were present in the last common ancestor of all angiosperms. The *Amborella* expansin superfamily was assembled from the *A. trichopoda* genome by using BLAST searches with angiosperm expansin queries. The results of these BLAST searches were analyzed and annotated to isolate the complete *A. trichopoda* expansin superfamily. This superfamily is smaller than other angiosperm expansin superfamilies. This is probably due to an absence of genome duplication events in *A. trichopoda's* history(Amborella Genome Project 2013). Phylogenetic and syntenic analyses of *A. trichopoda* expansins have improved our understating of the evolutionary history of expansins in angiosperms. It was possible to place nearly all of the *A. trichopoda* expansins into an existing Arabidopsis-rice expansin clade with high confidence. The results of phylogeny and synteny analyses allow us to estimate the number of expansins found in the last common ancestor of all angiosperms at 11 EXPA genes, 2 EXPB genes, 1 EXLA gene, and 2 EXLB genes.

This work was funded by Lebanon Valley College through an Arnold Student-Faculty Research Grant and the Wolf Fund. These sequence data were produced by the Amborella Genome Project in collaboration with the user community.

A COMPARISON OF PLANKTONIC COMMUNITIES AT THE MURPHY'S BOTTOM ECOLOGICAL SITE

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Murphy's Bottom is an ecological research site located along the Allegheny River in Armstrong County, PA. Due to mining activities in the mid- 1980's, a 25 acre lake and small annex were created adjacent to the Allegheny River. The goal of this project is to obtain baseline data on planktonic communities to predict the possible ecosystem changes that might result from connecting the lake to the river to form a backwater wetland. We hypothesized that the shallow and deep lake's planktonic communities should be highly similar due to their current connection. The shallow lake and the annex are of similar depth and connect seasonally; possibly leading to similar planktonic communities. Furthermore, all three sites are predicted to be very different from the isolated river. To test these predictions, we analyzed the plankton composition of water samples taken in 2008 and preserved in Lugol's solution. Aliquots were concentrated through a 0.45 μm filter which was examined using light microscopy. Similarity indexes compare species composition across all four sample sites and seasonal changes within sites. In mid June, the dinoflagellate *Ceratium* dominated the annex and the shallow lake while cyanobacteria and diatoms thrived in the deep lake and the river respectively. Our prediction of connectivity-based similarity was partially supported. As predicted, the lake sites and annex differed from the river in planktonic composition. The shallow lake was more similar to the annex than the deep lake, revealing the importance of water depth in shaping planktonic communities.

We would like to thank Dr. John Stolz and Dr. Alan Seadler for providing the necessary materials, training and protocols for this project and Benjamin Latoche for performing the sample collections. Funding for this project was provided by the Pennsylvania Environmental Protection Agency through the Murphy's Bottom Ecological Project.

PATTERNS OF TORSOLIKE AND ACTIVATED MAP KINASE DURING OVIPAROUS DEVELOPMENT IN THE PEA APHID

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The Terminal System is a network of genes and gene products that specifies the anterior and posterior poles of developing *Drosophila* embryos. One of these genes is *torso-like* and the system works through activation of MAP kinase. Aphids undergo two forms of reproduction, sexual oviparous reproduction and asexual viviparous reproduction. Previous work has shown that the pea aphid homolog of *torso-like* is expressed differently during these two modes of development. Here we focus on whether *torso-like* and activated MAP kinase are expressed and distributed in a manner consistent with terminal system-like function during oviparous development in the pea aphid. In particular, we investigated the possible relationship between *torso-like* and activated MAP kinase during oviparous development by in-situ hybridization and antibody staining, respectively. Adjacent distributions of *torso-like* mRNA and activated MAP kinase were observed, consistent with the hypothesis that Torso-like plays a role in activating MAP kinase. Further testing, such as double antibody and in-situ labels or RNA interference will be required to demonstrate the relationship between *torso-like* and MAP kinase and their involvement in specifying posterior fate during oviparous development in aphids.

Afternoon Poster Session
Group KK – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

218. **ANALYSIS OF THE EFFICACY OF THE ANNVILLE WASTE WATER TREATMENT PLANT FOR REMOVAL OF NSAIDS**

Megan Blauch and Owen A. Moe
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219. **LEAD DETERMINATIONS IN SOIL FROM A RECREATIONAL SHOOTING RANGE BUILT ON A RECLAIMED STRIP MINE: EFFECTS OF OXIDANT FLOW RATE ON PB MEASUREMENTS OBTAINED BY FAAS, AND OTHER CONSIDERATIONS**

Luke J. Metzler and Mark T. Stauffer
Chemistry Department, Division of Natural Sciences, Mathematics, and Engineering,
University of Pittsburgh at Greensburg, Greensburg, PA 15601

220. **EXTRACTING AND TESTING CHITOSAN FOR USE IN REVERSIBLE CARBON CAPTURE**

Haneef Muhammad, Benjamin Barnes, Preeti Sharma and Victoria V. Volkis
Department of Natural Sciences, University of Maryland Eastern Shore, Princess Anne, MD 21853

221. **USING GEOGRAPHIC INFORMATION SYSTEM (ArcGIS) ON LAKES IN NORTH-CENTRAL MINNESOTA**

Jeronimo Ocampos[1], Kenton Montgomery[2] and Daniel Lundberg[1]
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800 Florida Avenue NE, Washington DC 20002
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222. **DESIGN OF A HIGHLY EFFICIENT, COST EFFECTIVE ANODE FOR CHLORINE EVOLUTION AND WASTEWATER TREATMENT**

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ANALYSIS OF THE EFFICACY OF THE ANNVILLE WASTE WATER TREATMENT PLANT FOR REMOVAL OF NSAIDS

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Pharmaceuticals play an important role in maintaining the health of humans and animals. Due to the design of many pharmaceuticals to be toxic for infections or to be interactive with receptors within an organism, if they are introduced into the environment certain animal and plant species may be negatively affected. Multiple sources for the introduction of pharmaceuticals into the environment have been pinpointed but waste water treatment plants have been identified as one of the greatest sources. Our research focuses on the Annville Pennsylvania Waste Water Treatment Plant's process for waste water treatment, specifically its ability to reduce the levels of three non-steroidal anti-inflammatory drugs (NSAIDs). We have determined the levels of ibuprofen, naproxen, and salicylic acid using a process that involved pre-concentration by solid phase extraction (SPE), followed by derivatization using BF_3/MEOH, and analysis of the resulting methyl esters by gas chromatography-mass spectrometry (GC/MS). Although we did see appreciable scatter in our data, our results are consistent with the reduction of naproxen and salicylic acid concentrations through the treatment process. The fate of ibuprofen was less conclusive. Our future work will involve the use of LC/MS as an alternative approach to the analysis of these NSAIDs.

LEAD DETERMINATIONS IN SOIL FROM A RECREATIONAL SHOOTING RANGE BUILT ON A RECLAIMED STRIP MINE: EFFECTS OF OXIDANT FLOW RATE ON PB MEASUREMENTS OBTAINED BY FAAS, AND OTHER CONSIDERATIONS

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This investigation is a continuation of ongoing research into leaching of lead through soil at a recreational shooting range built on a reclaimed strip mine. The focus of the current investigation is to determine the optimum analysis conditions, particularly the oxidant flow rate used for determination of lead in soil from the shooting range by flame atomic absorption spectrometry (FAAS). Lower-than-expected Pb recoveries on spiked soil samples have generated high interest in determination of an optimum oxidant flow rate for the FAAS absorbance measurements for Pb. A preliminary experiment on a set of Pb-spiked soil samples yielded lower recoveries for Pb. It was hypothesized that not all of the ionic Pb present in the analysis solutions undergoes reduction to neutral Pb in the air-acetylene flame. Adjustment of oxidant flow rate to a value yielding a richer flame produced Pb results for spiked samples that indicated essentially complete Pb recoveries. The current objective is to study the effect of oxidant flow rate on the slopes of calibration curves used for Pb determinations, and subsequently on the results obtained for Pb in soil. In addition to the oxidant flow rate study, water from the man-made lake downhill from the shooting range will be analyzed for lead to determine if any migration actually occurs.

Results of the aforementioned investigation, along with details of sample collection, preparation, analytical methods, discussion of factors that might hinder Pb migration through the soil, and future directions for this research, will be presented and discussed.

EXTRACTING AND TESTING CHITOSAN FOR USE IN REVERSIBLE CARBON CAPTURE

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The increasing dependence of the modern world on fossil fuels for generating electricity, industrial products, and transportation has resulted in an unprecedented increases in the levels of greenhouse gasses in Earth's atmosphere. These gasses serve as in insulating layer, blocking the escape of certain wavelengths on radiation from the earth, and reemitting it thus leading to a gradual increase in average global temperatures over long periods of time. A variety of materials have been proposed for capturing carbon dioxide from high concentration point sources but so far, the most successful techniques have captured the carbon via an irreversible pathway, meaning that captured carbon dioxide must be stored geologically or subjected to oceanic release. Both of these release procedures cause damage to the environment in the long term, and are also expensive and not immediately re-warding for society in the short term. There are materials capable of capturing carbon dioxide via a reversible mechanism, however, which would allow a successive controlled release of the gas into developing green chemical processes, or into algal farms to create biomass for fuel. The materials proposed for this process are chitin and chitosan, polysaccharides that can be extracted from mollusk and arthropod body components.

USING GEOGRAPHIC INFORMATION SYSTEM (ArcGIS) ON LAKES IN NORTH-CENTRAL MINNESOTA

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Geographic Information System (ArcGIS) is a designed software system for storing, adding, and manipulating geographical data and layers. ArcGIS can be used for city planning, politics, and environmental research to name a few. This summer, I learned how to use ArcGIS to increase understanding of Brainerd, MN area lakes and their watersheds.

A watershed represents the area of land where water, that is under it or drains off of it, goes into a lake. The lake's health can be greatly affected by what occurs in the watershed, especially from human influence. Impenetrable surfaces like roads and development do not absorb the water as well as forested landscapes, which causes more runoff into the lake. Runoff can carry nutrients and pollutants from the watershed into the lake, changing the lake's water quality. ArcGIS can determine the watershed boundary and sub-watersheds can then be analyzed after pour points on the lake are identified.

Digital elevation media helps determine the pour-points for each lake. A pour-point represents a location where a sub-watershed drains water directly into the lake. In each sub-watershed, land use can be determined. The program lists seven different types of land use, such as brushland, forests, development, and agriculture. With this knowledge, possible sources of nutrient loading and pollution can be identified, leading to recommendations to the lake association on how to manage these possible problems.

ArcGIS also helped to determine the lake's water volume, littoral zones, bathymetry, and create three-dimension images of the watershed (using topography) and the lake (using bathymetry). The wealth of images generated by the program helps people better understand the land use, watershed, and lake. These images were excellent tools in describing how watersheds are directly associated with the lakes' health to lake associations, which helps them maintain/improve their lakes' health.

I would like to thank the anonymous donor, contributing through the Gallaudet University Development Office, for providing financial support to the internship. And to the Gordon Brown Fund, administered by the Gallaudet University Career Center, for my stipend and lodging.

DESIGN OF A HIGHLY EFFICIENT, COST EFFECTIVE ANODE FOR CHLORINE EVOLUTION AND WASTEWATER TREATMENT

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Nearly 20% of the manufacturing expense of the Hoffmann group's self-contained, photovoltaic-powered toilet and wastewater treatment system is contributed by the cost of the electrode array. This high expenditure is due largely to the current design of the IrO_2 anode used within the wastewater electrolysis cell. In order to reduce this cost, an alternative anode design was proposed that replaces the costly IrO_2 interlayer with a more affordable substance: RuO_2. The anodes were prepared by repetitively brush coating Ru precursor onto pretreated conductive Ti sheets and annealing until a desired molar loading was achieved. Then an aqueous titanium glycolate complex was thermally decomposed onto the surface to provide stability. These anodes were then coupled with a stainless steel cathode and AgCl reference electrode in a single compartment electrolysis cell and electrochemically analyzed with potentiostatic methods for current efficiency, chlorine evolution rate, and reaction onset potential. Next, each anode was subjected to an accelerated life test operated at a high current in order to determine the stability. The most promising samples were then employed in wastewater electrolysis tests to observe their chemical oxygen demand (COD) and ammonium removal capabilities. Overall, the best performing electrode design was comprised of $1.56\ \mu mol/cm^2$ RuO_2 and $3.76\ \mu mol/cm^2$ TiO_2. Finally, a mechanistic study was performed in order to examine the pathway by which wastewater treatment is achieved. These tests showed that ClO^- formation is negligible in the electrolysis process while $\cdot OH$ and $\cdot Cl_2^-$ are produced in significant concentrations. The results indicate that the cost-effective RuO_2 electrode is even more efficient than the previous IrO_2 design in terms of chlorine evolution and COD removal. Additionally, the previous mechanism that attributed free chlorine as the most important agent in COD reduction was challenged and alternate compounds of interest were identified.

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Afternoon Poster Session
Group LL – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

223. **CHARACTERIZATION OF TRANSITION METAL BOUND 1, 4, 7-TRITHIACYCLONONANE**

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224. **THE SYNTHESIS AND CHARACTERIZATION OF POLYMERIZED COBALT SELENIDE CLUSTERS WITH PHOTOVOLTAIC APPLICATIONS**

Daniel A. Corbin[1], Devon M. Shircliff[1], Brian J. Reeves[2], and Brycelyn M. Boardman[1]
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225. **SYNTHESIS, STABILITY, AND DNA-BINDING AND CYTOTOXICITY STUDIES OF ORGANORHENIUM COMPLEXES OF MEFENAMIC ACID**

Tiara Hinton and Santosh K. Mandal
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226. **SYNTHESIS OF HYPERVALENT BISIODONIUM SALTS AND A POLYFLUOROALKYLATION INVESTIGATION**

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227. **THE POLYMERIZATION AND CHARACTERIZATION OF FUNCTIONALIZED PALLADIUM COMPLEXES WITH BENZOTHIADIAZOLE CO-MONOMERS**

Alexandra M. Moore, Jessica L. Shott, and Brycelyn M. Boardman
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228. **SYNTHESIS, STABILITY, DNA-BINDING AND CYTOTOXICITY STUDIES OF ORGANORHENIUM COMPLEXES OF FLUFENAMIC ACID**

Sabreea Parnell and Santosh K Mandal
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229. **NOVEL IMIDAZOLIUM BASED ARCHITECTURES THAT EXHIBIT EXCITED STATE INTRAMOLECULAR PROTON TRANSFER**

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230. **SYNTHESIS AND ION-BINDING STUDIES OF RUTHENIUM(II) BIPYRIDINE/POLYETHYLENE OLIGOMER COMPLEXES**

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CHARACTERIZATION OF TRANSITION METAL BOUND 1, 4, 7-TRITHIACYCLONONANE

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Online access of this abstract is restricted at the request of the Principal Investigator.

THE SYNTHESIS AND CHARACTERIZATION OF POLYMERIZED COBALT SELENIDE CLUSTERS WITH PHOTOVOLTAIC APPLICATIONS

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Hybrid organic-inorganic bulk heterojunction (BHJ) photovoltaic devices have recently gained interest within the scientific community for their potential to offer higher efficiency solar cells at a lower cost than those in the current market; however, the orthogonal properties of organic and inorganic materials cause the two to interact poorly within the devices. Since the BHJ architecture relies on good interaction between the donor (organic) and acceptor (inorganic) materials, this has proven to be a large obstacle to developing a more ideal hybrid solar cell.

To overcome this issue, we synthesized a new series of polymers via Stille coupling with a constant amount of $(Me_3Sn)_2(C_4H_2S)$ and varying ratios of $Co_6Se_8(Br(C_4H_2S)P(Ph)_2)$ (**1**) to $Br_2(C_4HS)(CH_2)_5CH_3$; these polymers have been named poly(cluster-co-thiophene-co-hexylthiophene)a-d (PCLTHTa-d). All of the polymers were studied through the use of UV-Visible, Nuclear Magnetic Resonance, and Fluorescence Spectroscopy, as well as Atomic Force Microscopy. Not only do PCLTHTa-d exhibit a wide range of solubility, a problem that has long plagued inorganic photovoltaics, but they also show increased charge transfer efficiency compared to representative simple mixtures of **1** and poly(thiophene-co-hexylthiophene).

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SYNTHESIS, STABILITY, AND DNA-BINDING AND CYTOTOXICITY STUDIES OF ORGANORHENIUM COMPLEXES OF MEFENAMIC ACID

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Numerous transition-metal complexes of nonsteroidal anti-inflammatory drugs (NSAIDs) are known and many of them possess interesting biological properties. Surprisingly, the examples of organometallic complexes of NSAIDs are scarce. The only known complexes are organocobalt complexes of aspirin derivatives. We have synthesized a series of organorhenium complexes of mefenamic acid, a NSAID, from the reactions of corresponding pentylcarbanonato complexes with mefenamic acid as shown in eq. 1:

$$\textit{fac}\text{-}(CO)_3(N\text{-}N')ReOC(O)OC_5H_{11} + \text{Mefen-H} \longrightarrow \textit{fac}\text{-}(CO)_3(N\text{-}N')Re(\text{Mefen}) + C_5H_{11}OH + CO_2 \quad (1)$$

The mefenamato complexes were characterized spectroscopically and in some cases through X-ray crystal structure determinations. The stabilities and the DNA-binding studies of the complexes at *p*H 7.2 were studied using UV-Vis spectrophotometry. Preliminary experiments demonstrate that the mefenamato complexes are cytotoxic against U-937 lymphoma cell lines.

SYNTHESIS OF HYPERVALENT BISIODONIUM SALTS AND A POLYFLUOROALKYLATION INVESTIGATION

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The installation of fluorine and fluorous groups into organic molecules provides many advantageous changes to their physical, chemical, and biological properties. Such advantageous effects include enhancements in metabolic stability and lipophilicity, which makes the optimization of fluorination methods an active field in drug discovery. An efficient fluoroalkylation is observed with the trifluoroethylation of indole derivatives *via* C-H bond activation through the use of hypervalent iodonium salts, such as 2,2,2-trifluoroethy(mesityl)-iodonium triflate. Due to the high reactivity of the mono-aryliodonium salt, we were interested in synthesizing bis-aryliodonium salts with the expectation that they would display similar polyfluoroalkylating abilities. This poster describes our progress on the syntheses of hypervalent bisiodonium salt species and their ability as polyfluoroalkylating agents.

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THE POLYMERIZATION AND CHARACTERIZATION OF FUNCTIONALIZED PALLADIUM COMPLEXES WITH BENZOTHIADIAZOLE CO-MONOMERS

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In the field of photovoltaics, inorganic silicon-based structures have offered power conversion efficiencies of up to 46%. Despite this, constructing such materials requires expensive processing conditions. Therefore, these technologies are not cost-effective enough to compete with existing energy systems. Hybrid organic-inorganic materials have recently been explored in this respect, incorporating nanoparticle-polymer blends into the active layer. These architectures offer improved efficiencies, although phase separation between the organic and inorganic components may lead to poor charge separation and charge recombination. A potential solution to this is to covalently link cobalt-chalcogenide clusters to conjugated polymers via functionalized phosphine ligands. However, this process yields complex structures that are difficult to characterize. To better understand the interactions between the organic and inorganic components, a model complex capable of polymerization will be investigated. This model consists of a palladium metal center covalently linked to functionalized phosphine ligands.

The synthesis of functionalized phosphine ligands was performed to isolate palladium complexes capable of polymerization. The ligands were prepared via lithium halogen exchange with n-butyllithium followed by the addition of chlorodiphenylphosphine. The reactions of 2,5-dibromothiophene and 4,7-dibromobenzo[c]-1,2,5-thiadiazole under these conditions produced 2-bromo-5-diphenylphosphinothiophene (**1**) and 4-bromobenzo-7-diphenylphosphino-1,2,5-thiadiazole (**2**) respectively. Compounds **1** and **2** were purified via column chromatography and then allowed to react with dichloro(1,5-cyclooctadiene)palladium(II). The polymerizable metal complex bis(2-bromo-5-diphenylphosphinothienyl)dichloropalladium(II) (**3**) results from **1** while no palladium complex was isolated from the reaction with **2**. Compound **3** was then allowed to react with 2,5-bis(trimethylstannyl)thiophene, 2,5-dibromo-3-hexylthiophene, and 4,7-dibromobenzo[c]-1,2,5-thiadiazole in various ratios to produce three sets of co-polymers. Characterization of the co-polymers using ^{1}H and ^{31}P NMR, UV-Visible spectroscopy, and fluorescence spectroscopy indicates covalent attachment of **3** into the polymer backbone. Manipulation of the co-monomer ratios also results in the ability to fine tune the optical properties of the co-polymers.

Research Corporation Single Investigator Cottrell College Science Award 22628 and NSF REU CHE-1461175 supported this work.

SYNTHESIS, STABILITY, DNA-BINDING AND CYTOTOXICITY STUDIES OF ORGANORHENIUM COMPLEXES OF FLUFENAMIC ACID

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Many metal complexes of non-steroidal anti-inflammatory drugs (NSAIDs) are known to exhibit interesting biological properties. Very few examples of organometallic complexes of NSAIDs are known. Recently a few organocobalt aspirin derivatives have been synthesized and found to be highly effective against MCF-7 breast cancer cell lines. We have synthesized a series of organorhenium polypyridyl (N~N) complexes of flufenamic acid (FNA-H), an NSAID, from the reactions of the corresponding pentylcarbanato complexes with FNA-H as shown in eq.1:

$$fac\text{-}(CO)_3(N\sim N)ReOC(O)OC_5H_{11} + FNA\text{-}H \rightarrow fac\text{-}(CO)_3(N\sim N)Re(FNA) + C_5H_{11}OH + CO_2 \quad (1)$$

The organorhenium flufenamato complexes were characterized through FT-IR and FT-NMR spectroscopic techniques and in some cases through X-ray crystal structure determinations. The stabilities and DNA-binding properties of the flufenamato complexes were determined using UV/Vis spectrophotometry. Preliminary experiments suggest that the compounds presumably bind to DNA intercalatively and are cytotoxic against U-937 lymphoma cell lines.

NOVEL IMIDAZOLIUM BASED ARCHITECTURES THAT EXHIBIT EXCITED STATE INTRAMOLECULAR PROTON TRANSFER

Nicholas Pompetti, Gabriel Andrade, and Joel Rosenthal
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Online access of this abstract is restricted at the request of the Principal Investigator.

SYNTHESIS AND ION-BINDING STUDIES OF RUTHENIUM(II) BIPYRIDINE/POLYETHYLENE OLIGOMER COMPLEXES

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The Harris lab focuses on the synthesis and ion-binding studies of novel ruthenium host guest complexes. Host guest complexes are supramolecular complexes that consist of a "host" molecule that binds a "guest" ion or molecule through non-covalent interactions. These complexes have many biological and environmental applications such as chemical sensing, drug delivery, and the removal of ions from waste products. Our goal is to develop fluorescent host complexes that selectively bind or "sense" specific cationic or anionic guests. The ion-binding affinities of ruthenium(II) bipyridine/polyethylene oxide oligomer host complexes were investigated by fluorescence spectroscopy to determine the equilibrium constant (K_s) and stoichiometry (n) with a library of alkali, alkaline earth, and transition metal cation guests.

I would like to acknowledge the LVC Department of Chemistry and the Neidig Endowed Research Fund for the continued support of this project.

Afternoon Poster Session
Group MM – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

231. **THEORETICAL STUDY OF THE METALLA-DIELS-ALDER CYCLIZATION: AN EXAMINATION OF THE EFFECT OF RHODIUM, COBALT, AND OSMIUM REPLACEMENT OF C-H IN 1,3-BUTADIENE VIA THE ISOLOBAL ANALOGY**

Anastasiya Badziai and Elena Votto, Edyta Greer
Department of Natural Sciences, Baruch College, 55 Lexington Avenue, New York, NY 10010

232. **PREPARATION AND CHARACTERIZATION OF METAL/LIGAND COMPLEXES USING SUBSTITUTED N-TRIAZOLYLPROPANAMIDE LIGANDS**

Julia E. Beiro, Donna S. Amenta, and John W. Gilje
Department of Chemistry and Biochemistry, James Madison University, 800 S. Main Street, Harrisonburg, VA 22807

233. **EXPLORATION OF NANOMAGNETISM PRESENTED IN A FAMILY OF $[Ln^{III}_4Mn^{III}_4]$ (Ln^{III} = Y^{III}, Dy^{III}, Ho^{III}, Er^{III}) COMPOUNDS**

Andrew H. Davis[1], Curtis Zaleski[2], Jeff W. Kampf[3],
Vincent L. Pecoraro[3], and Thaddeus T. Boron[1]
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[3] Department of Chemistry, University of Michigan, 930 N. University Avenue,Ann Arbor, MI 48108

234. **aSYNTHESIS OF QUATERANARY HETEROCYCLIC SALTS**

Gbeminiy Jesutimi and Angela Winstead
Department of Chemistry, Morgan State University, 1700 E. Cold Spring Lane, Baltimore, MD 21251

235. **PREPARATION OF RUTHENIUM COMPLEXES OF N-TRIAZOLYLPROPANAMIDE DERIVATIVES**

Robert Sherman[1] Cristian Hrib[2], Frank Edelmann[2], John Gilje[1] and Donna Amenta.[1]
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800 S Main Street, Harrisonburg, VA 22807
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236. **RUTHENIUM TRIS-BIPYRIDINE CAGE COMPLEXES AS HOST SYSTEMS FOR ALKALI AND ALKALINE EARTH GUESTS**

Alyssa Smale, Adam Thomas, Marc Harris
Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

THEORETICAL STUDY OF THE METALLA-DIELS-ALDER CYCLIZATION: AN EXAMINATION OF THE EFFECT OF RHODIUM, COBALT, AND OSMIUM REPLACEMENT OF C-H IN 1,3-BUTADIENE VIA THE ISOLOBAL ANALOGY

Anastasiya Badziai and Elena Votto, Edyta Greer
Department of Natural Sciences, Baruch College, 55 Lexington Avenue, New York, NY 10010

This study presents an *in silico* study on the replacement of carbon fragments in the parent Diels-Alder cycloaddition reaction involving 1,3-butadiene and ethylene. The overall goal is to determine mechanistic preferences and reaction energetics for hypothetical derivatives of 1,3-butadiene where carbons are replaced by metal fragments based on the principle of isolobal analogy. In previous works, substituting a carbon fragment with its isolobal metal fragments led to reductions in the activation energies of Bergman cyclization and Cope rearranagement. Based on these findings, we hypothesized that a similar reduction in activation energy could be achieved for metalla-Diels-Alder cycloaddition. =CH– fragment in *cis*-butadiene was replaced by its isolobal metal fragments, $=Rh(PH_3)_3–$, $=Co(PH_3)_3–$, and $=Os(PH_3)_3H–$.

Interestingly, the cases of involving 2-rhodobutadiene and 2-cobaltobutadiene did not undergo the expected Diels-Alder mechanism, but rather, cyclopropanation. The activation energies were 25.47 kcal/mol and 24.89 kcal/mol, respectively. Contrary to the expected energetic influence of the metal fragments, the activation energies were higher than the experimental activation energy of Diels-Alder cycloaddition, which was 23.4 kcal/mol. Unlike rhodium and cobalt fragments, 2-osmobutadiene underwent Diels-Alder cycloaddition to give rise to osmacyclohexene. However, the activation energy of this reaction was 36.78 kcal/mol, which was higher than that of the parent Diels-Alder system.

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PREPARATION AND CHARACTERIZATION OF METAL/LIGAND COMPLEXES USING SUBSTITUTED N-TRIAZOLYLPROPANAMIDE LIGANDS

Julia E. Beiro, Donna S. Amenta, and John W. Gilje
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The synthesis and characterization of previously unreported substituted N-triazolylpropanamide ligands N,N-dimethyl-(N-benzotriazole)-propanamide, **1**, 3-(N-(5-methylbenzotriazole))-propanamide, **2**, and 2-methyl-(N-(1,2,4-triazole))-propanamide, **3**, have been accomplished. Each synthesis produced multiple isomers in varying amounts. Isomers of **1** were synthesized from the reaction of benzotriazole and N,N-dimethylacrylamide in the presence of triton B. The reaction of ruthenium(II)chloridetristriphenylphosphine with isomeric mixtures of **1** resulted in predominately one isomer. We propose that the predominate isomer contains a chelating ligand. Isomeric mixtures of **1** were also allowed to react with ruthenium(II)chloridetristriphenylphosphine in a 2 to 1, metal to ligand, molar ratio resulting in a bridged diruthenium complex. The reaction of isomeric mixtures of **1** with manganese(II)chloridetetrahydrate is currently under investigation. Isomers of **2** were synthesized from the reaction of 5-methylbenzotriazole and N,N-dimethylacrylamide in the presence of triton B. The reaction of ruthenium(II)chloridetristriphenylphosphine with isomeric mixtures of **2** resulted in three new ruthenium complexes. Isomers of **3** were synthesized from the reaction of 1,2,4-triazole and methacrylamide in the presence of triton B. The reaction of ruthenium(II)chloridetristriphenylphosphine with isomeric mixtures of **3** resulted in two new ruthenium complexes.

I would like to acknowledge the National Science Foundation (NSF REU CHE-1461175) and the James Madison University Department of Chemistry and Biochemistry for providing funding for this research.

EXPLORATION OF NANOMAGNETISM PRESENTED IN A FAMILY OF $[Ln^{III}_4Mn^{III}_4]$ (Ln^{III} = Y^{III}, Dy^{III}, Ho^{III}, Er^{III}) COMPOUNDS

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SYNTHESIS OF QUATERANARY HETEROCYCLIC SALTS

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PREPARATION OF RUTHENIUM COMPLEXES OF N-TRIAZOLYLPROPANAMIDE DERIVATIVES

Robert Sherman[1] Cristian Hrib[2], Frank Edelmann[2], John Gilje[1] and Donna Amenta.[1]
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2-Methyl-3-[N-benzotriazole]-propanamide was successfully synthesized from the reaction of 1,2,3-benzotriazole and methacrylamide in the presence of Triton B. Two isomers formed, 2-methyl-3-[1N-benzotriazole]-propanamide (**1**) and 2-methyl-3-[2N-benzotriazole]-propanamide (**2**), and were separated by fractional recrystallization. Isomer **1** has been characterized by NMR and IR spectroscopy and x-ray crystallography. Isomer **2** has been characterized by NMR and IR spectroscopy. Single crystals of **2** have been obtained and are awaiting x-ray analysis. Separate reactions of both **1** and **2** with excess tris(triphenylphosphine) ruthenium(II) chloride **(4)** appear to yield mainly bridged diruthenium complexes, which are being characterized.

I would like to thank the National Science Foundation REU for the funding grant CHE-1461175.

RUTHENIUM TRIS-BIPYRIDINE CAGE COMPLEXES AS HOST SYSTEMS FOR ALKALI AND ALKALINE EARTH GUESTS

Alyssa Smale, Adam Thomas, Marc Harris
Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

Bipyridine tripodal ligands were synthesized in order to form ruthenium cage complexes, which function as host molecules for cationic guests (Na^+, Cs^+, Mg^{2+}, Ba^{2+}). The host properties of the ruthenium cage were investigated using liquid-liquid membrane ion transport studies and UV-visible and fluorescence titration studies, which were performed in order to determine the binding efficiency and selectivity of the hosts for the various cationic guests.

This project was supported in-part by Lebanon Valley College and grants from the Endowed Neidig Chemistry Research Fund.

Afternoon Poster Session
Group NN – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

237. **CHARACTERIZATION OF FCP1 HELICAL STRUCTURE**

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[2]Department of Chemistry, Pennsylvania State University, 104 Chemistry Building, University Park, PA 16802

238. **CHEMICAL AND CHEMOENZYMATIC SYNTHESIS OF UDP-N-ACETYL GLUCOSAMINE BACTERIAL CELL WALL PROBES**

Tyler Heiss, Kristen DeMeester, Hai Liang, Borja Barbero, and Catherine Leimkuhler Grimes
Department of Chemistry and Biochemistry, University of Delaware, 163 The Green, Newark, DE 19716

239. **INFLUENCE OF SALTS AND D_2O ON THE SECONDARY STRUCTURE AND STABILITY OF PROTEINS**

Elijah T. Johnson and Gina MacDonald
Department of Chemistry and Biochemistry, James Madison University, Harrisonburg, VA 22807

240. **MULTI-STEP SMALL-MOLECULAR EXTRACTION FOR ENHANCING THE DETECTABLE COVERAGE OF THE METABOLOME IN SINGLE EMBRYONIC CELLS USING CE-ESI-MS**

David O. Plotnick, Rosemary M. Onjiko, and Peter Nemes
Department of Chemistry, The George Washington University, 800 22nd Street, NW, Suite 4000, Washington DC 20052

241. **THE TRANSITION FROM B TO Z-DNA USING TRIS(ETHYLENEDIAMINE)COBALT(III) TRANSITION METAL COMPLEX AND NEUTRAL ALCOHOL OSMOLYTES**

Kelsey Polak, Aloise Diedrich, and Richard Preisler
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242. **USING FLUORINATED AMINO ACIDS AS PROBES IN PROTEIN- PROTEIN INTERACTIONS**

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CHARACTERIZATION OF FCP1 HELICAL STRUCTURE

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Intrinsically disordered proteins (IDPs) are proteins that lack a temporally stable three dimensional structure. This flexibility imbues IDPs with the functional versatility to participate in essential cellular processes, such as signal transduction and transcription. Previous studies have shown that when some IDPs bind to cooperatively folded proteins they form more orderly secondary structures, thus modulating their function in biological pathways. Yet the mechanisms involved in these interactions remain poorly understood. Our system of interest is the FCP1: Rap74 complex, where upon binding FCP1 folds into a more orderly alpha helix. The goal of our project is to characterize the interactions responsible for FCP1: Rap 74 complex formation. Our hypothesis is that stabilization of FCP1's helical content will increase the binding affinity that drives complex formation. We tested this using nuclear magnetic resonance to monitor the helical character of mutant FCP1 strains. Future studies into understanding the structure of biologically significant IDPs could give further insight into signaling pathways and diseases.

This investigation was supported in part by a MARC Undergraduate Student Training in Academic Research (U-STAR) National Research Service Award (NRSA) Institutional Research Training Grant (2 T34 GM008663) from the National Institutes of Health, National Institute for General Medical Sciences and NSF MCB Grant # 0953918.

CHEMICAL AND CHEMOENZYMATIC SYNTHESIS OF UDP-N-ACETYL GLUCOSAMINE BACTERIAL CELL WALL PROBES

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Online access of this abstract is restricted at the request of the Principal Investigator.

INFLUENCE OF SALTS AND D_2O ON THE SECONDARY STRUCTURE AND STABILITY OF PROTEINS

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The secondary structure of the model protein, myoglobin, has been studied using circular dichroism (CD) spectroscopy. Previous work has shown that the presence of ions in the Hofmeister series can alter the solvation of these proteins. Previous experiments have shown that Hofmeister salts alter myoglobin solvation and stabilize the protein in the predicted Hofmeister order. These studies have shown that myoglobin's thermal stability can be affected by the presence of ions. The previous studies also suggested that the anions affected the interaction of water molecules and that deuterium oxide influenced the effects of some anions. Earlier studies led to speculation that protein solvation may be affected differently in water and deuterium oxide. Additional, more complete studies that could help discern the role of cations was also of interest. The present work expands on previous studies. Myoglobin unfolding was studied with cationic Hofmeister ions, NH_4^+, Cs^+, Rb^+, K^+, Na^+, Li^+, Ca^{2+}, Mg^{2+}, Ba^{2+}, with a constant anion, Cl^-, at temperatures ranging from 25-105°C. The experiments were performed in water and deuterium oxide at pH 7.5. Our data shows significant cation-dependent changes in the thermal stability and aggregation of myoglobin in water and deuterium oxide.

MULTI-STEP SMALL-MOLECULAR EXTRACTION FOR ENHANCING THE DETECTABLE COVERAGE OF THE METABOLOME IN SINGLE EMBRYONIC CELLS USING CE-ESI-MS

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Knowledge of all metabolites (the metabolome) produced in embryonic cells (blastomeres) promises to help better understand basic biochemical processes that drive normal development. As a downstream result of genomic, transcriptomic, and proteomic processes, the metabolome is considered to be a powerful descriptor of a cell's phenotype. However, metabolites are difficult to characterize analytically because they are dynamic, have a wide concentration range, and encompass molecular classes that range from highly polar to highly apolar. This challenge is further compounded by the limited amount of material available in single blastomeres. To characterize the metabolome of single blastomeres, new protocols are needed that are able to collect single blastomeres and to efficiently extract their metabolites for analysis by high-resolution mass spectrometry, the analytical method of choice for the detection and quantitation of small molecules.

Our goal was to develop a multi-step approach to enhance the detectable coverage of the metabolome in single blastomeres. Blastomeres were isolated from the 8-cell embryo of the South African clawed frog (*Xenopus laevis*), a popular model for cell and developmental studies, and different solvents were used to extract the polar, moderately apolar, and apolar portions of the single-cell metabolomes. The resulting extracts were characterized using a custom-built single-cell capillary electrophoresis electrospray ionization mass spectrometer that separated, detected, and identified metabolites with high confidence. Relative comparison of metabolite abundances revealed complementary performance using the multi-step extraction approach, essentially increasing the coverage of the single-blastomere metabolome than what would be detectable using the traditional one-step extraction. A more comprehensive coverage of the metabolome will enable us to derive a deeper understanding of biological pathways underlying early embryonic development.

This work was supported by the GW Start-Up Funds (to P.N.), the National Institutes of Health Grant R21 GM114854 (to P.N.), and the Cosmos Club Foundation (to R.M.O.).

THE TRANSITION FROM B TO Z-DNA USING TRIS(ETHYLENEDIAMINE)COBALT(III) TRANSITION METAL COMPLEX AND NEUTRAL ALCOHOL OSMOLYTES

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The transition from B to Z-DNA is influenced by the presence of salt cations, transition metal cations, and hydration. Highly charged octahedral transition metal complexes have been shown to stabilize the Z-conformer, including tris(ethylenediamine)cobalt(III).[1] Previous research has shown that increasing osmotic stress through the presence of osmolytes (solutes that remove water from the vicinity of the DNA) decreases the concentration of the transition metal complex necessary to induce the transition from B to Z-DNA.[2,3] We have created a hydrophobicity trend that expands on and is consistent with those determined by Rau, et al.[2] and Chaze, et al.[3] using neutral diol solutes and tris(ethylenediamine)cobalt(III). In the series of alcohols and diols we examined recently, as hydrophobicity of the osmolyte increases, the concentration of tris(ethylenediamine)cobalt(III) required for the transition decreases.[4] We also determined that as the concentration of the osmolyte increases, the amount of the complex to stabilize Z-DNA decreases. Overall, it has been concluded that the amount of transition metal complex required for the transition from B to Z- DNA is affected by the nature and the concentration of the osmolyte. Most recently, the (+) and (-) enantiomers of tris(ethylenediamine)cobalt(III) were separated via a resolution experiment in order to explore the extent of stereospecific interactions component of the stabilization of Z-DNA. Preliminary data indicates that the (+) enantiomer may stabilize the Z conformation to a greater extent than does the (-) enantiomer. However, inconsistencies between DNA samples and a limited amount data prevent us from drawing any firm conclusions. Future experiments will continue to determine the relevance the stereochemistry of the cobalt complex in the transition from B to Z-DNA.

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3. Preisler, R. S. and Chaze, C. (2013), Biophysical Society 57th Annual Meeting poster.

We thank Dr. Alan Pribula of Towson University for preparation of coordination compounds and Dr. Donald C. Rau for useful discussions. This research was supported by Undergraduate Research Grants (to M. Cisse, A. Diedrich and D. Rey-Ardila) and by the Department of Chemistry, Towson University.

USING FLUORINATED AMINO ACIDS AS PROBES IN PROTEIN-PROTEIN INTERACTIONS

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^{19}Fluorine is a 100% naturally abundant isotope and is not naturally found in biological systems. It has a nuclear spin of ½, making it highly responsive to NMR measurements. I am using fluorinated amino acids to study protein-protein interactions. Protein-protein interactions control activities in cells, such as cell growth, gene expression, intercellular communication, and nutrient absorption. The activities of the cell are regulated by outside stimuli, which activate proteins through these PPI, which in turn code for responses within the cell. Diseases and cancers occur because of defects in signal transduction. Research interest lies with targeting these active PPI binding domains with alternative proteins that will have better binding than the protein causing the defect.

To study protein-protein interactions, I have synthesized the unnatural amino acid, perfluoro-*tert*-butyl homoserine, in six steps on a multigram scale. This amino acid has a ^{19}Fluorine nucleus that will be used to detect the interaction by ^{19}F NMR. The perfluoro-*tert*-butyl moiety contains nine equivalent fluorines which adds special sensitivity of a 81 fold increase over a single fluorine. This group is a sharp singlet by ^{19}F NMR. Using the sensitivity of the perfluoro-tert-butyl group will allow for detection of subtle changes in the local environment and can provide information on tertiary structure and interactions of fluorine labeled proteins and peptides. This unnatural amino acid was incorporated as a probe into three novel peptides ligands to determine the effect of replacing natural amino acids of a native peptide sequence, to test whether the replacement increases affinity and/or specificity within SH3 domains, and to determine the effectiveness of the perfluoro-*tert*-butyl moiety as a probe in PPI. The peptides have a PXXP motif because of the hydrophopic SH3 domains. SH3 domains are found in signaling proteins. I will use three proteins containing SH3 domains: Src, Crk, and Grb.

Thank you to NSF for funding this project.

Afternoon Poster Session
Group OO – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

243. **SYNTHESIS OF CDSE AND AU NANOPARTICLES ASSEMBLIES TO STUDY THE OPTICAL PROPERTIES OF NEW HYBRID NANOMATERIALS**

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Department of Chemistry and Biochemistry, UMBC, 1000 Hilltop Circle, Baltimore, MD 21250

244. **GROWTH, CHARACTERIZATION X EXFOLIATION OF MOLYBDENUM SULFIDE NANOMATERIALS**

Mandy Houghton, Henry D. Snyder, and Paul Sabila
Biology, Chemistry & Physics, Department of Math, Science, & Technology,
Gallaudet University, 800 Florida Avenue NE, Washington DC, 20002

245. **A CATALYTIC AND SURFACE ANALYSIS OF THE HYDROGENATION OF PHENOL USING PALLADIUM NANOPARTICLES**

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Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

246. **TETRA-AZA LIGANDS FOR An(III)\Ln(III) SEPARATIONS**

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Department of Chemistry, Salisbury University, 1101 Camden Avenue, Salisbury, MD 21801

247. **THE DIRECT BORYLATION OF ALKENES, A BORYL-HECK REACTION**

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SYNTHESIS OF CDSE AND AU NANOPARTICLES ASSEMBLIES TO STUDY THE OPTICAL PROPERTIES OF NEW HYBRID NANOMATERIALS

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It is predicted that when the coupling of cadmium selenide quantum dots and gold nanorods occurs, the properties of the resulting system will be qualitatively different from the properties of the isolated particles. These properties can further be controlled by exciting the system with a short laser pulse. This project is important because it will show that the nanoparticles assemblies can serve as a key enabling technology for future optical information processing at high speeds and low powers, including quantum-mechanical information processing at the single-photon level (i.e. quantum computers). This also has potential applications in providing a more efficient conversion of sunlight into electricity and in enabling highly efficient displays and ultra-small lasers. The goal of this project is to couple cadmium selenide quantum dots to gold nanorods in order to study the optical properties of this new type of hybrid nanomaterial.

Currently, we have synthesized both the quantum dots and gold nanorods. They have been characterized using UV-Vis Spectrophotometry, Fluorometry, and Transmission Electron Microscopy. The next step is to couple the quantum dots with the gold nanorods. We will approach this by reacting the ligands coating the particles.

This work was funded through an Undergraduate Research Award from the UMBC Office of Undergraduate Education.

GROWTH, CHARACTERIZATION X EXFOLIATION OF MOLYBDENUM SULFIDE NANOMATERIALS

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Two-dimensional nanomaterials show to be promising through the existence of quantum confinement and the absence of interlayer interactions. When a large material reduces its dimension to under 100 nm, substantial changes in its properties can follow.

Molybdenum disulfide (MoS_2) is a transition metal dichalcogenide (TMD) and has promising electronic and optoelectronic applications. In 1963, bulk MoS_2 was exfoliated successfully. In this account, we describe the chemical exfoliation of MoS_2 layers using n-BuLi and the deposition of MoS_2 on silicon wafers. The properties and evidence for deposition of MoS_2 on silicon wafers are summarized and discussed. These products are analyzed using Raman microscope, Scanning Electron Microscope (SEM), Electron Dispersive X-Ray Spectroscopy (EDS), optical microscope, and Octave software.

This work was done under Partnership for Reduced Dimensional Materials (PRDM) supported by grants from the National Science Foundation (NSF #1205608). We thank Howard University, Howard Nanofabrication Facility (HNF), Howard University Chemistry Department, Gallaudet University, and Dr. Tito Huber's Research Laser Facility for their support.

A CATALYTIC AND SURFACE ANALYSIS OF THE HYDROGENATION OF PHENOL USING PALLADIUM NANOPARTICLES

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The hydrogenation of phenol is an important industrial reaction in that it produces cyclohexanone, a key intermediate in the manufacture of nylon polymers. In our work phenol was hydrogenated in a bench top reactor using polyvinylpyrrolidone (PVP) capped palladium nanoparticles in water. A gas chromatograph/mass spectrometer (GC/MS) was used to monitor the reaction every 20 minutes for two hours. A selected ion monitoring (SIM) method on the GC/MS was programmed to find only the expected products of cyclohexanone and cyclohexanol, along with the starting material phenol. Additional reactions were performed with a commercial 5% palladium on silica catalyst and PVP-capped palladium nanoparticles immobilized on silica microspheres. For each catalyst the turnover number and turnover frequency were determined and the turnover frequency was used to compare the different types of catalysts for their catalytic activity. Furthermore, reaction selectivity for cyclohexanone with each catalyst was also found.

The surface of the palladium nanocatalysts was analyzed to determine the number of surface palladium atoms that are available for binding. Attenuated total reflectance fourier transform infrared (ATR FTIR) spectroscopy was used to monitor the adsorption of carbon monoxide on the nanoparticle surface. Modified Beer's law plots were used to find the number of surface binding sites, which in turn was used to calculate the dispersion factor, or ratio of active surface sites to total palladium atoms. The dispersion factor is then used to "correct" the turnover frequencies. Through varying reaction conditions of temperature and reactant concentrations, as well as nanocatalyst particle size and PVP capping agent molar mass, we aim to elucidate the factors that influence the atomic scale design of greener and more efficient catalytic materials.

Acknowledgment is made to the Donors of the American Chemical Society Petroleum Research Fund for support of this research.

TETRA-AZA LIGANDS FOR An(III)\Ln(III) SEPARATIONS

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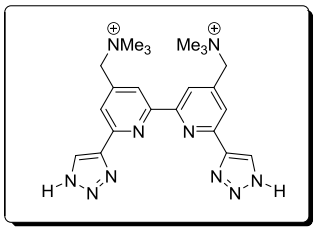
Nuclear waste generally consists of 94% uranium, 4%-5% fission product, 1% plutonium, 0.1% minor actinides (americium and curium). Currently, there is no clear policy for spent nuclear fuel recycling nor for nuclear waste disposal in the United States.[1] Leaving nuclear waste unprocessed is neither energy efficient nor environmental friendly due to its high radiotoxicity. Ideally, purified uranium from the nuclear waste could reenter the reactor to generate more energy.

In addition to reusing the uranium, finding ways to efficiently reduce the transplutonium elements that contribute to the radiotoxicity of nuclear waste is an important consideration when finding practical ways to overcome the problems that this waste causes. Integral to these considerations are finding separation methods that use ligands that have binding affinities to An(III) that are greater than that for the Ln(III). Although 4f and 5f elements have nearly identical chemical behavior (e.g. charge, size), separation methods that use ligands with soft N-donors have been shown to have a greater binding affinity to An(III) than that for Ln(III).

A new class of tetra-aza ligands, bistriazolylbipyridines, have been shown to exhibit physical and chemical properties that might be conducive to nuclear waste processing conditions. A major concern is to make these N-donor ligands soluble in sufficient concentration to allow for practical separation on an industrial scale. Previous attempts have been made to make these ligands soluble in nonpolar solvents, however with limited success. To circumvent this issue, we propose ligands that are water-soluble as they should still maintain their extraction efficiencies while removing long R groups that would be necessary for organic solubility. As a consequence, making the ligands water-soluble instead might provide a more practical method to carry out these separations.



THE DIRECT BORYLATION OF ALKENES, A BORYL-HECK REACTION

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Online access of this abstract is restricted at the request of the Principal Investigator.

Afternoon Poster Session
Group PP – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

248. **ISOLATION, BIOASSAY, AND FTIR ANALYSIS OF ACETYLEUGENOL AND EUGENOL FROM CLOVES**

Danielle Brown[1], Jerica Wilson[2], Christopher Stromberg[3], Perry Wood[2] and Debra Ellis[2]
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249. **FLUORINATED BROMOTHYMOL BLUE: A CERENKOV-ABSORBING MOLECULAR PROBE FOR IN-VIVO PH MEASUREMENT OF TUMOR MICROENVIRONMENT**

Nikaela W. Bryan[1, 2], Alejandro D. Arroyo[3], Alexander V. Kachur[3],
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250. **USING POLYMERIC RESINS FOR THE EXTRACTION OF PURE ANTHOCYANIN FROM ARONIA MITSCHURINII BERRIES**

Adaobi Egwuagu[1], Heather Goldsborough[1], Taiwo Ola[1], Andrew Ristvey[2], and Victoria Volkis[1]
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251. **DETERMINATION OF COCOA LIQUOR PROVENANCE USING FATTY ACID AND TRACE ELEMENTAL SIGNATURES**

Sara Maloney, Paige Sudol, Ravleenkaur Khalsa, Shannon E. Stitzel, and Ryan E. Sours
Department of Chemistry, Towson University, 8000 York Road, Towson, MD 21252

252. **CATION EFFECTS ON THERMODYNAMICS OF CAFFEINE PARTITIONING BETWEEN AQUEOUS AND CYCLOHEXANE PHASES**

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ISOLATION, BIOASSAY, AND FTIR ANALYSIS OF ACETYLEUGENOL AND EUGENOL FROM CLOVES

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Opportunities that integrate hands-on experience with research-grade instrumentation and interdisciplinary techniques are crucial for engaged learning. Experiments that have real-life applications are particularly effective at increasing students' interest. The partnership between organic chemistry and microbiology for the pharmaceutical industry is a good example of a real-life interdisciplinary application. The extraction and bioassay of naturally occurring antibiotics in cloves intertwine three fields of science: organic chemistry, microbiology, and instrumentation.

Multiple variations of the distillation procedure for extracting the antibiotics from cloves were performed to determine the optimum procedure for the organic chemistry lab at Frederick Community College (FCC). The challenge with using the standard whole cloves procedure is the long time required to perform the distillation needed to obtain sufficient oil. Cloves were prepared in the following alternative ways to identify the shortest procedure with the highest yield: soaking overnight in water, grinding with a mortar and pestle ground, and grinding in a coffee grinder. None of the alternative methods offered a time or yield advantage. Portions of the distilled clove oil are subsequently extracted to obtain the natural antibiotics eugenol and acetyleugenol. Students are able to check the identity and purity of the clove oil, eugenol and acetyleugenol extracts on the newly acquired, state-of-the-art FTIR.

In addition, a bioassay was designed specifically for use in the FCC organic chemistry lab, given that some students will have no microbiology experience. The clove oil, eugenol and acetyleugenol extracts were bioassayed for their antibiotic properties against two microorganisms; *Escherichia coli* (*E. coli*) and *Bacillus cereus* (*B. cereus*). Overall, the Gram negative bacterium *E. coli* exhibited a greater resistance to the oil and extracts compared to the Gram positive bacterium *B. cereus*. Acetyleugenol was more effective at inhibiting *B. cereus* growth, and eugenol was slightly more effective at inhibiting the growth of *E. coli*.

This project was supported by Hood College, Frederick Community College, and Mount St. Mary's University and funded through the National Science Foundation's Improving Undergraduate STEM Education program (DUE-1431522).

FLUORINATED BROMOTHYMOL BLUE: A CERENKOV-ABSORBING MOLECULAR PROBE FOR IN-VIVO PH MEASUREMENT OF TUMOR MICROENVIRONMENT

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Cerenkov radiation is a multispectral photonic emission that occurs when charged particles travel faster than the speed of the light through a medium. It can be detected and quantified using standard optical imaging equipment. This factor makes Cerenkov-specific molecules a powerful detection method for biological processes and environments *in vitro* and *in vivo*. We report here a probe that selectively attenuates Cerenkov emissions in response to varying pH. ^{18}F labeled derivatives of bromothymol blue were synthesized by electrophilic fluorination. The presence of fluorinated bromothymol blue products was confirmed and their structures were characterized by pKa studies, UV-vis absorbance spectra, and analysis by NMR and Liquid Chromatography/Mass Spectrometry. Fluorination of the compounds was determined to be mainly through bromine- fluorine exchange. Attenuation of Cerenkov photon emission was measured across a range of wavelengths as a function of pH. We observed a pH dependent attenuation of Cerenkov radiation at wavelengths closest to the molecule's lambda max. Cytotoxicity of the fluorinated products was tested in prostate cells using the MTT assay. Neither fluorinated product was determined to be harmful to the survival of prostate cells in vitro. We plan to test the distribution of the fluorinated products via tail vein injections in mice. With the delivery of this probe to tumors, potential measurement of pH in the tumor microenvironment will be possible.

USING POLYMERIC RESINS FOR THE EXTRACTION OF PURE ANTHOCYANIN FROM ARONIA MITSCHURINII BERRIES

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Aronia Mitschurinii is a berry commonly known as black chokeberry. Belonging in the Rosacea family, it is a fruiting woody shrub local towards the East Coast of the U.S and normally found in swampy areas. The berries thrive mostly in full sun areas and woodlands, and one of its species is naturalized in European countries. Chokeberries are usually cultivated for ornamental and consumption purposes due to their fleshy and purple colored skin but frequently, the berries are processed to extract their antioxidant contents. The extracted antioxidants contain high quantities of polyphenolic compounds like anthocyanins, which is present in both the berry juice and the pulp, and is accountable for the deep-pigmented purple color of the berries. The anthocyanins are considered beneficial in solving health problems and studies have shown that rising interest in human healthy feeding, food processing, coloring and cell culture studies has brought about an increase in the worldwide cultivation of the berries.

This project aims to test current resin techniques, such as those used with grapes, on the extraction of anthocyanin from both aronia juice and pulp. The resin beads are soaked in aronia juice or a pumice extract and alcohol solution. Once the anthocyanin molecules adsorb to the vast internal surface area of the beads, the beads are removed from the solution and exposed to a solvent where anthocyanin is released from the resin. The solvent is then evaporated off, leaving antioxidant behind. Antioxidant profile before and after the resin treatment, comparison of different resins, as well as purity analysis of isolated products will be presented.

Authors would also like to thank the UMES Honors Program as well as the American Chemical Society for support.

DETERMINATION OF COCOA LIQUOR PROVENANCE USING FATTY ACID AND TRACE ELEMENTAL SIGNATURES

Sara Maloney, Paige Sudol, Ravleenkaur Khalsa, Shannon E. Stitzel, and Ryan E. Sours
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In a country concerned with quality, flavor, and ethical practices, the traceability of cocoa and chocolate is very important. Although traceability of cocoa exists in some markets, the majority of consumers currently purchase chocolate without a viable way to trace the country of origin for major ingredients such as cocoa. This project investigated whether fatty acid signatures and trace elemental signatures could be correlated to the country of origin for single-origin cocoa liquors. Fatty acid methyl esters (FAMEs) were analyzed by GC-MS, and elemental analysis was performed using ICP-MS. The resulting data from both approaches were analyzed by discriminant analysis. Three main questions were investigated: 1) Can cocoa liquor samples from four or more countries be discriminated from each other based on fatty acid signatures and/or trace elemental signatures? 2) Do samples from the same country but with different sample characteristics have the same elemental/fatty acid signatures? 3) Which fatty acids/elements are most useful in this determination?

Initial results indicate that single-origin cocoa liquors from four countries can be discriminated from each other based on their elemental and fatty acid signatures. In addition, the cocoa liquor samples with different characteristics, such as temperature and roast time, had some differences in their signatures, but they were still correctly classified by country of origin. Future work will examine additional single-origin cocoa liquors and will investigate which elements/fatty acids are the most useful for the determination of provenance.

The authors would like to thank the Towson University Fisher College of Science and Mathematics and the Towson University Office of Undergraduate Research for their financial support. The authors would also like to thank Dr. Ed Seguine for the donation of the cocoa liquor samples and for many helpful discussions about cocoa and chocolate production.

CATION EFFECTS ON THERMODYNAMICS OF CAFFEINE PARTITIONING BETWEEN AQUEOUS AND CYCLOHEXANE PHASES

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Online access of this abstract is restricted at the request of the Principal Investigator.

Afternoon Poster Session
Group QQ – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

253. **AN NMR STUDY ON INTERACTIONS OF HOFMEISTER IONS WITH CAFFEINE**

Nicolas Johnson and Yanjie Zhang
Department of Chemistry and Biochemistry, James Madison University,
901 Carrier Drive, MSC 4501, Harrisonburg, VA 22807

254. **NITROGEN VARIATIONS AND POSSIBLE IMPACTS ON CHARDONNAY AND ALBARINO WINES**

Matthew E. Meyers and Stephen Robertson
Department of Chemistry, McDaniel College, 2 College Hill, Westminster, MD 21157

255. **SELF-POWERED ENZYMATIC BIOSENSOR FOR SIMULTANEOUS DETECTION OF TWO BIOMARKERS OF PARKINSON'S**

Gaige Vandezande, Jasmine Olvany, Julia Rutherford, and Michelle Rasmussen
Department of Chemistry, Lebanon Valley College, 101 North College Avenue, Annville, PA 17003

256. **CHEMICAL CHARACTERIZATION OF OSMIUM FAMILY ESSENTIAL OILS**

So Jin Park, Diamond Nwaeze, and Victoria V. Volkis
Department of Natural Science, University of Maryland Eastern Shore, One Backbone Road,
Princess Anne, MD 21853

257. **IDENTIFICATION OF COMPONENTS IN AN ANALGESIC TABLET USING INFRARED SPECTROSCOPY**

Jerica Wilson[1], Danielle Brown[3], Christopher Stromberg[2], Perry Wood[1] and Debra Ellis[1]
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AN NMR STUDY ON INTERACTIONS OF HOFMEISTER IONS WITH CAFFEINE

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NITROGEN VARIATIONS AND POSSIBLE IMPACTS ON CHARDONNAY AND ALBARINO WINES

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Varying levels of nitrogen, sugar and environmental factors contribute heavily to the overall chemical composition of a wine grape and its final destination, wine. With that said, should grape growers only worry about the sugar content for harvest, or should there be a heavier emphasis on the overall chemical composition of the grape? This question was answered through the use of a refractometer to measure grape sugar content and through the use of UV-vis spectroscopy to measure the nitrogen available for yeast consumption. The methods were conducted with two varieties of white grape, each assessed over a 4-week span leading to grape harvest and crush. The methods showed variation of available nitrogen and a gradual rise in sugar concentrations leading up to harvest.

Thank you to the McDaniel College Undergraduate Research Fund for funding my research and thank you to Old Westminster Winery for providing the grape samples and support for my research.

SELF-POWERED ENZYMATIC BIOSENSOR FOR SIMULTANEOUS DETECTION OF TWO BIOMARKERS OF PARKINSON'S

Gaige Vandezande, Jasmine Olvany, Julia Rutherford, and Michelle Rasmussen
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Parkinson's disease is a chronic neurodegenerative disorder which affects 1% of the world population over 60 years of age. There is currently no definitive test to detect Parkinson's disease in patients, thus it is diagnosed through symptoms and patient history. The purpose of this study is to fabricate and analyze self-powered enzymatic biosensors that have the ability detect biomarkers of Parkinson's disease prior to an onset of symptoms. A specific range of uric acid and glutathione levels in the plasma are the two biomarkers that indicate the possible presence of the condition. It has been found that in a patient with Parkinson's disease, uric acid levels are lower than normal while glutathione levels are higher.

Biosensors function by registering the amount of electrons donated or consumed through an electric current, produced by redox reaction that occurs directly on, or near the sensor. In order to detect the relative concentrations of the desired substances in the blood, two biosensors must be developed that are either enzymatically hindered or exacerbated by the presence of the biomarkers. This study utilized amperometry and cyclic voltammetry to explore the effectiveness of a laccase cathode and an uricase anode, which both respond, to the presence of glutathione and uric acid, respectively, by decreasing in current output. The changes in current can then be translated into relative concentrations of the substances in the blood, allowing the tested plasma to be compared to both healthy and diseased blood.

This project was conducted under the aid of Lebanon Valley College Endowed Research fund.

CHEMICAL CHARACTERIZATION OF OSMIUM FAMILY ESSENTIAL OILS

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Essential oils from plants are known for antibacterial and antiseptic properties. Many of plants along with these oils has a high antioxidant content. Essential oils are found to be effective at preventing microorganism activity, whereas antioxidants are encouraging the eradication of free radicals which, in turn, can aid in the prevention and eradication of cancer and other diseases. Antioxidants are compounds with highly conjugated systems that are able to inhibit the process of oxidation by eliminating free radicals within the environment.

In this project we study variety of herbs belong to the Ocium (basil) family: *Ocium Basilicumm, Ocimum Gratissimum, Tulsi Rama, Tulsi Kapoor* and *Ocimum Sanctum.* All of these hetbs are lovally grown by medical herbs specializing Habanera Farm in Maryland, whereas the origin and the most popular growing locations for these herbs are in India, Malaysia and part of Tibet. The essential oil of *Ocimum Gratissimu*, and *Ocimum Sanctum* grown in India and Malaysia was already found to contain antimicrobial and antioxidant compounds.

The essential oils from different Holy basil samples were obtained through wet distillation and the molecular components of the essential oils were studied by using gas chromatography-mass spectrometry (GC-MS). It was determined that the essential oil from various Holy Basil samples contained Eugenol, Ursoic acid, Rosmarinic acid, Ocimarin, Apigenin, and Ocimunoside A and B. Then locally grown samples were compared with literature data about same herbs from India and Malaysia.

IDENTIFICATION OF COMPONENTS IN AN ANALGESIC TABLET USING INFRARED SPECTROSCOPY

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Knowledge of and experience with instrumentation is crucial to success within a STEM field of study. Community college students gain theoretical knowledge in the classroom but often find themselves at a disadvantage with the lack of experience with state-of-the-art equipment. Our objective was to modify existing undergraduate organic chemistry laboratory experiments to incorporate a Fourier Transform Infrared Spectrometer (FTIR).

The isolation of components within an analgesic tablet is an excellent lab for demonstrating the use of a combination of separation and extraction techniques. The goal of the experiment is to successfully separate caffeine, acetaminophen, and acetylsalicylic acid from an Excedrin tablet. Prior to receiving the FTIR, there was not a definitive method for determining if the separations and extractions performed were successful. Several student samples were tested on the FTIR, but the correct products were not being obtained, even for those students with a sufficient yield. The incorporation of the new instrumentation allowed us to identify flaws within the experiment, scrutinize why the issues were occurring, and modify the protocol. The integration of the FTIR not only gives students access to important technology, but it has elevated the experiments being performed and holds the students accountable for their work in the laboratory.

This project was supported by Hood College, Frederick Community College, and Mount St. Mary's University and funded through the National Science Foundation's Improving Undergraduate STEM Education program (DUE-1431522).

Afternoon Poster Session
Group RR – Chemical Sciences

Poster # | **Title, Author(s) & Affiliation(s)**

258. **DETERMINATION OF THE KINECTICS OF BIODIESEL FORMATION USING ^{1}H NMR**

Antoinette H. Issis and Anderson L. Marsh
Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

259. **EFFECTS OF SOLVENT ON THE IONIC LIQUID MEDIATED ELECTROCHEMICAL CONVERSION OF CO_2 TO CO AT A BISMUTH-BASED ELECTRODE**

Thomas P. Keane, John L. DiMeglio, and Joel Rosenthal
Department of Chemistry and Biochemistry, University of Delaware, Brown Lab, 162 Academy Street, Newark, DE 19716

260. **SYNTHETIC ROUTES TO $OS_3(CO)_{10}(\mu\text{-}OET)_2$ & ALKOXIDE SUBSTITUTION REACTIONS WITH 1,5-PENTANEDIOL**

Robert Sommerhalter, Katie Marak, and Mary-Ann Pearsall
Department of Chemistry, Drew University, 36 Madison Avenue, Madison, NJ 07940

261. **EXAMINING THE EFFECTS OF LIGAND STRUCTURE ON THE STEREOISOMERS OF RUTHENIUM (II) DICHLORIDE COMPLEXES WITH CHIRAL, TETRADENTATE AMINSOSULFOXIDE LIGANDS**

Cassidy Stout and Tim Brunker
Department of Chemistry, Towson University, 8000 York Road, Towson, MD 21252

262. **PHOTOCHEMICAL O-ATOM EXCHANGE AND OXIDATION OF NITROSAMINES WITH MOLECULAR OXYGEN**

Radhika Viswanathan[1], Ashwini A. Ghogare[1], Marilene Silva Oliveira[1,2], Inna Abramova[1], Edyta M. Greer[3], Fernanda Manso Prado[2], Paolo Di Mascio[2], and Alexander Greer[1]
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DETERMINATION OF THE KINECTICS OF BIODIESEL FORMATION USING ^{1}H NMR

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Department of Chemistry, Lebanon Valley College, 101 N. College Avenue, Annville, PA 17003

Declining petroleum reserves has spurned interest into the development of alternative transportation and heating fuels. Biodiesel is composed of fatty acid methyl esters (FAMES) derived from vegetable oil through a transesterification reaction with methanol. The most common catalyst used is a homogeneous hydroxide, either potassium or sodium. Heterogeneous catalysts offer the benefits of being able to be separated from the glycerol layer that forms and to be recycled for further use. In order to develop a more efficient catalytic process the kinetics of the reaction must be understood.

The purpose of this project was to determine the kinetics of biodiesel under various conditions and to determine how varying elements of the transeterification reaction will affect the kinetics of biodiesel. Biodiesel production was monitored at 25 ^{0}C, 35 ^{0}C, and 45 ^{0}C at various starting volumes of oil with sodium hydroxide as the alkaline catalyst. Aliquots were extracted at selected times during the reaction, quenched to inhibit further reaction, and then analyzed using proton nuclear magnetic resonance (NMR) spectroscopy. Peak integrations from NMR spectra were used to calculate the concentrations of the reactant oil and methyl ester products, the rate of product to methyl ester, and the rate of oil conversion to methyl ester.

The results above will be applied to analyze the kinetics of biodiesel production via a recyclable nanocatalyst. Magnetic nanoparticles were prepared by co-precipitation using iron salts. The magnetic nanoparticles were subsequently coated with silica. The silica coated nanoparticles were subsequently functionalized with quaternary ammonium hydroxide functional groups on the surface. Future work will involve using this catalyst in the transesterification of vegetable oil to form biodiesel and monitoring the kinetics with proton NMR spectroscopy.

Acknowledgment is made to the Donors of the Lebanon Valley College Chemistry Endowed Research Fund for support of this research.

EFFECTS OF SOLVENT ON THE IONIC LIQUID MEDIATED ELECTROCHEMICAL CONVERSION OF CO_2 TO CO AT A BISMUTH-BASED ELECTRODE

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Rising atmospheric CO_2 concentrations, and the reality of anthropogenic climate change currently drive a desire to shift the global energy portfolio from reliance on fossil fuels towards more sustainable sources, such as wind and solar power. However, a major obstacle to accomplishing this goal is the need for a reliable, compact mode of storage and transport for the energy generated by these sources. One solution to this problem involves the storage of this energy in chemical bonds, via the electrochemically catalyzed conversion of CO_2 to CO. CO produced in this process can be converted to liquid fuels through Fischer-Tropsch chemistry in order to yield carbon neutral fuels which can be readily integrated into the already-present global infrastructure.

A bismuth-based, heterogeneous catalyst was recently developed by our lab which, in the presence of imidazolium-based ionic liquids, can drive the electrochemical reduction of CO_2 to CO with high current density at a low overpotential. The function of this electrode in several different solvents was studied in order to gain insight into the nature of the catalyst's function. Correlations were sought between solvent properties and both reaction current density and selectivity for CO production. The goals of this project were twofold: First, to improve the function of the catalyst system; and second, to gain insight into the mechanism behind the reaction. Progress along both these lines will be discussed.

SYNTHETIC ROUTES TO $OS_3(CO)_{10}(\mu\text{-}OET)_2$ & ALKOXIDE SUBSTITUTION REACTIONS WITH 1,5-PENTANEDIOL

Robert Sommerhalter, Katie Marak, and Mary-Ann Pearsall
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With the goal of developing systematic syntheses of linked triosmium carbonyl clusters, we report the investigations of new synthetic pathways to the starting compound $Os_3(CO)_{10}(\mu^2\text{-}OEt)_2$ (**1**), and the substitution reaction of **1** with 1,5-pentanediol. Synthetic approaches to **1** starting from known clusters $Os_3(CO)_{12}X_2$ (X=Cl (**2**), Br (**3**), I (**4**)) and $Os_3(CO)_{10}(\mu\text{-}X)_2$ (X=Br (**5**), I (**6**)) are reported. Viable pathways to **1** are observed from **2**, **5** and **6**, with pathway **2** being the most favored. Reaction of **1** with 1,5-pentanediol affords the cluster $Os_3(CO)_{10}(\mu\text{-}O(CH_2)_5OH)_2$ (**7**) as the major product. Over extended reaction periods, quantitative conversion to the cluster $H_4Os_4(CO)_{12}$ (**8**) is observed. Linking of two triosmium carbonyl clusters via the 1,5-pentanediol bridging ligands will be discussed.

EXAMINING THE EFFECTS OF LIGAND STRUCTURE ON THE STEREOISOMERS OF RUTHENIUM (II) DICHLORIDE COMPLEXES WITH CHIRAL, TETRADENTATE AMINSOSULFOXIDE LIGANDS

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PHOTOCHEMICAL O-ATOM EXCHANGE AND OXIDATION OF NITROSAMINES WITH MOLECULAR OXYGEN

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We are interested in photooxygen atom transfer and oxidation processes of *N*-nitrosamines. We have provided evidence for an O-atom exchange process of nitrosamines with molecular oxygen through acyclic and cyclic peroxy intermediates called *O*-nitrooxide, 1,2,3,4-trioxazetidine, and 1,2,3,5,6,7-hexaoxadiazocane. The photolysis of four nitrosamines (*N*-nitrosodiphenylaniline **1**, *N*-nitroso-*N*-methylaniline **2**, *N*-butyl-*N*-(4-hydroxybutyl)nitrosamine **3**, and *N*-nitrosodiethylamine **4**) with ultraviolet light was examined in an $^{18}O_2$-enriched atmosphere in solution. HPLC/MS and HPLC-MS/MS data show that ^{18}O-labeled nitrosamines were generated for **1** and **2**. In contrast, nitrosamines **3** and **4** do not exchange the ^{18}O label and instead decomposed to amines and/or imines under the conditions. A density functional theory study of the structures and energetics of peroxy intermediates arising from reaction of nitrosamines with O_2 is also presented. A reversible head-to-tail dimerization of the nitrooxide to the hexaoxadiazocane (30 kcal/mol barrier) with extrusion of O=^{18}O accounts for exchange of the oxygen atom label. The unimolecular cyclization of *O*-nitrooxide to trioxazetidine (46 kcal/mol barrier) followed by a retro [2 + 2] reaction is an alternative, but higher energy process. Both pathways would require the photoexcitation of the nitrooxide.

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Morning Poster Session - STEM BUILD at UMBC

263. **CORRELATION BETWEEN THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA POPULATION SIZE**

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264. **CORRELATION BETWEEN AGE AND THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA**

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265. **THE IMPACT OF GEOGRAPHIC LOCATION ON MICROBIOME DIVERSITY**

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266. **THE DIVERSITY OF THE HUMAN MICROBIOTA IS AFFECTED BY THE PROXIMITY TO NATURAL WATER SOURCES**

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267. **INVESTIGATION OF THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA IN GEOGRAPHICALLY PROXIMAL LOCATIONS**

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268. **THE DIVERSITY OF BELLYBUTTON MICROBIOTA IN RELATION TO HUMAN DEVELOPMENT**

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269. **INVESTIGATING COMMON BELLY BUTTON MICROBIOTA BY GENUS CLASSIFICATION IN U.S. REGIONS**

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CORRELATION BETWEEN THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA POPULATION SIZE

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For every cell in the human body, there are ten microbial cells. Previous studies have shown that an individual's microbiota contributes to personal health. Through a metagenomics approach, we can investigate how microbes are involved in diseases, such as obesity, diabetes and heart disease. The purpose of this study was to investigate if there was a correlation between city population size and the level of microbial diversity found in individual's belly buttons. Given that individuals from a larger city would have a greater opportunity of human contact, we hypothesized that individuals from a more populated city, Los Angeles (3.884 million), would have a greater diversity of microbes in their belly buttons compared to individuals from a less populated city, Manhattan Beach (.036 million). The data analyzed in this study was from a previous study investigating the biodiversity of the belly button microbiome. A metagenomics approach using 16s rRNA sequencing was used and data was analyzed using Phinch.org software. Our data reveals that Manhattan Beach had a greater diversity of microbes when investigating the species taxa compared to Los Angeles. Manhattan Beach had twenty-nine total microbial species while Los Angeles had eleven. Interestingly, when comparing the kingdom taxa, Manhattan Beach showed two total taxa: one being bacteria and the other proposed as archaea. Los Angeles only had one kingdom, bacteria. Based on our results, our hypothesis was refuted because we observed a greater diversity in belly button microbiota in the Manhattan Beach sample when compared to the Los Angeles. This may be explained by the closer proximity of Manhattan Beach to the Pacific Ocean compared to Los Angeles. Future goals of this research will investigate whether the diversity of the belly button microbiota is affected by proximity to seawater and investigate the correlation between microbiota and diseases associated with populated areas.

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CORRELATION BETWEEN AGE AND THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA

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The microbiome is the genetic material isolated from a microbial community and is used to study the human microbiota, or the microbes on or within an individual. Human microbiota is of interest to scientists and clinicians, as various health factors and diseases are linked to the presence of microbes. Microbial DNA isolated from an individual can be studied using the process of metagenomics analysis, which is the study of genetic material from a specific environment. Using previously acquired data from a metagenomics study, we analyzed the belly button microbiota to determine the diversity of the bacteria and how it correlates to age. We hypothesized that children between 0 and 10 years of age have the most diverse microbiome because of their body's immature immune response. To test our hypothesis, we grouped individuals based on years of age: 0 and 10 years (children), 20 and 30 years (young adults), and 40 and 50 years (middle-aged adults) and analyzed data using Phinch software investigating the family taxonomic level. Our data revealed that certain age groups contained specific microbiota that were linked to certain diseases. For example, young adults (20-30 years) had a high prevalence of bacteria in the *Enterobacteriacae* family, of which *Salmonella*, a common form of food poisoning, is a member. Young adults (20-30 years) were also found to have a high prevalence of bacteria in the *Staphylococcacae family*, which contains the species responsible for staph infections. Our collected evidence suggests that young adults may have the most diverse belly button microbiota, which refutes our hypothesis. A possible explanation is that young adults have a greater interaction with others, with further investigation needed to identify how social interacts affect microbiota diversity.

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THE IMPACT OF GEOGRAPHIC LOCATION ON MICROBIOME DIVERSITY

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Microbes are extremely important to human health, as they play a role in diseases, such as diabetes and certain types of cancer. External factors, such as exposure to ultraviolet light, proximity to aquatic environments, or temperature, could potentially affect the microbiota, or the microbial community present in or on a person. This study focused on the microbiota of human bellybuttons; more specifically, it investigated whether the geographic location that an individual lives in affects the diversity of microbes found in belly buttons. It was hypothesized that individuals residing in warmer climates near aquatic environments have a more diverse belly button microbiota. To test this hypothesis we compared the belly button microbiota of individuals in Minnesota to that of individuals in Florida. A metagenomic approach was used to address the hypothesis using previously acquired 16S ribosomal RNA sequencing data. Phinch software was used to analyze the dataset to reveal microbiome diversity on the species taxonomic level. Three individuals from Florida and three individuals from Minnesota were analyzed for this study. It was observed that the average number of bacterial species in the belly button microbiome of individuals residing in Florida was 69 with a range of 17 species to 104 species. In contrast, the average number of bacterial species in Minnesota was 43 with a range of 22 species to 70 species. Although our sample size was too small to achieve statistical significance, the data shows a trend that supports our hypothesis. To further support our hypothesis it would be necessary to obtain a larger sample size and control other factors, including travel, age, wash frequency, and belly button morphology. For future research, we could explore the correlation between geographical region, belly button microbiota, and disease incidence.

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INVESTIGATION OF THE DIVERSITY OF THE BELLY BUTTON MICROBIOTA IN GEOGRAPHICALLY PROXIMAL LOCATIONS

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Metagenomics is the study of genetic material from environmental samples in order to identify the microbes present. Knowing information on specific microbes can help scientists analyze bacteria present on an individual to determine how the bacteria contributes to human health and disease. In this study, we used a metagenomics approach to investigate whether the diversity of the microbes present in individuals' belly buttons was different in proximal geographic regions of the United States. We hypothesized that similar proportions of the same types of bacteria, using the taxonomic level of order, are found in geographically proximal cities. We analyzed a previously acquired belly button microbiome, or DNA isolated from bacterial community, data set for our study. Our analysis focused on six cities in Texas that were geographically proximal: Austin, Corpus Christi, Houston, Lufkin, Manor and San Marcos. The cities in Texas were selected because of their cultural diversity and fast-growing population size. We used Phinch software to analyze the data and specifically used donut partitions to investigate the taxonomic level of order. Our results revealed that there were three bacterial order taxa, *Actinomycetales*, *Bacillales*, and *Bacteroidales*, commonly found in the six cities. Within these cities, San Marcos and Manor, which are forty-seven miles apart, share the same predominant bacterial order, *Actinomycetales*. Alternatively, *Bacillales* and *Bacteroidales* were the most prevalent bacterial order in Austin, Corpus Christi, Houston, and Lufkin. Based on our data, we conclude that our hypothesis was correct. With this knowledge, we can better understand the human microbiota and how microbiota-associated diseases may affect individuals from specific geographic locations. Future plans include exploring alternative geographically proximal locations in the US and internationally.

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THE DIVERSITY OF THE HUMAN MICROBIOTA IS AFFECTED BY THE PROXIMITY TO NATURAL WATER SOURCES

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An individual's microbiota is comprised of microscopic organisms that are found on the skin, mouth, and intestines. The human microbiota is influenced by internal health factors, and a correlation has been found between the existence of certain microbes and the development of various diseases. Moreover, external factors, such as climate change and geographic location, also affect the diversity of the microbiota found on individuals. In this study, we focused on the external factors that influenced the diversity of the belly button microbiota. Specifically, we investigated whether the distance from an ocean affects the microbiome diversity in the belly button using a previously collected microbiome data set. To accomplish this, we used a metagenomics approach investigating 16S ribosomal RNA isolated from individuals living in coastal cities in California, Texas, Florida, and Massachusetts and compared the data to individuals living in inland areas in Ohio and Massachusetts. In our analysis, we focused on the phylum taxonomic level and hypothesized that the proximity to a natural water source affects the microbial diversity in the navel. Our data focused on the presence of two bacterial phyla: *Firmicutes* and *Actinobacteria.* Our results demonstrate that there is a greater prevalence of *Firmicutes* in coastal cities (average 56.6%) compared to inland cities (average 19.3%). In comparison, we observed a greater presence of *Actinobacteri*a in inland areas (average 61%) compared to coastal cities (average 27%). Thus, our results suggest that geographic proximity to the ocean affects the microbial presence in the human belly button. Given that *Firmicutes* has been associated with obesity, future research on whether the obesity rates in coastal areas are due to a high *Firmicute* presence should be investigated. Additionally, since tuberculosis is caused by microbes in the *Actinobacteria* phylum, future research can also investigate its presence in inland areas.

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THE DIVERSITY OF BELLYBUTTON MICROBIOTA IN RELATION TO HUMAN DEVELOPMENT

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The human microbiota is made up of an estimated 100 trillion different microbes that live on or within us. In comparison to the human body, microbes outnumber human cells 10:1. Understanding our microbiota and how it changes as we develop can help researchers better understand how microbes play a role in human health and disease. In this study, previously collected belly button microbiome data was used to investigate whether there were differences in the diversity of bacteria found in the naval cavity. We analyzed metadata from individuals at various stages of human development, with a specific focus on the phylum taxonomic level. We focused on individuals who were 0-10, 11-20, and 21-30 years of age, which contained 35, 38, 55 samples respectively. We hypothesized that the distribution of bacteria within the ages of 0-10 would show the greatest and most diverse number of bacterial phyla among the age groups investigated. The data revealed that a majority of the same phyla were shared within all age groups, with little variation between them. Some of these phyla included *Firmicutes*, *Proteobacteria*, *Cyanobacteria*, *Bacteroidetes*, and *Actinobacteria*. Ages 11-20 featured two additional phyla not found in age groups 0-10 and 21-30 that were associated with bacteria found in the intestines and groundwater. Our preliminary data did not support our hypothesis and instead suggested that the 11-20 age group had the greatest belly button microbiota diversity. This may be due to greater social interactions in this age group, providing evidence that social interactions contribute to the diversity of an individual's microbiota. Future directions of this work include following individuals from birth to the age of 30 and tracking the diversity of their bellybutton microbiomes as they develop and interact with their environment.

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INVESTIGATING COMMON BELLY BUTTON MICROBIOTA BY GENUS CLASSIFICATION IN U.S. REGIONS

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The microbiota is the community of bacteria that live in and on humans. These bacteria influence human health in numerous ways. However, most of the human microbiota has been unidentified. A recent study attempted to classify the bellybutton microbiota by sequencing bacterial DNA isolated from an individual, or the microbiome. This study revealed immense diversity in individual microbiota but also noted a trend of "predictability", meaning the most abundant bacteria tended to remain prominent among different human populations. The previous study identified five common phylum of bacteria. The purpose of this study was to investigate the consistency of belly button microbiome diversity at the genus taxonomic level. We hypothesized that the most abundant bacteria found in different regions of the United States will be dominated by the same genus. We used previously collected belly button microbiome data and stratified it by its state of origin. Then we classified each state into five geographical regions: Northeast, Southeast, Midwest, Southwest, and Western. Finally, we examined bubble charts to identify the top genus in each region. Our results revealed that *Corynebacterium* and *Staphylococcus* were universally expressed as the top two genus of bacteria across every region. However, genus three through five varied among the regions. *Alicyclobacillus* was only found in Southern regions. In addition, *Streptococcus* and *Pseudomonas* only appeared in the Midwest while *Campylobacters* was only found in the West. Since the top two genus of bacteria were consistent among regions, our data implies that human populations from different geographic locations have similar microbiota. Nevertheless, our results partially refuted the hypothesis since only two of the genus remained consistent in each region. Future directions of this research are to perform advanced statistical analysis to accurately confirm the geographic distribution of the microbiota.

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