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## THIRTEENTH ANNUAL SUMMER RESEARCH SYMPOSIUM TRINITY COLLEGE

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## **BIOLOGY**

#### 1.

### SPECIALIZATIONS FOR YOLK CELLULARIZATION IN THE LIZARD SCLEROPORUS UNDULATUS

Madeline Barnes '20, Luisa Lestz '19 Faculty Sponsor: Daniel Blackburn

This summer we devoted our research to better understanding the methods by which the lizard *Sceloporus undulatus* processes yolk in the egg to provide nutrients for the growing embryo. By examining yolk from various stages of embryonic development under the scanning electron microscope (SEM), we have been able to notice a pattern in yolk cellularization. In early stages, 28-34, the yolk consists of free yolk spheres with few nutrient-rich, endodermal cells. As development continues, (in stages 34-37) the endodermal cells proliferate, phagocytosing large yolk spheres and possibly breaking them down into smaller droplets within the cell. In this stage, cells appear to maintain thin cellular connections. Finally, late stage eggs, 37-40, show the proliferation of blood vessels which are surrounded by endodermal cells filled with yolk droplets. These "spaghetti strands" provide the growing lizard embryo an efficient means to receive nutrients from the yolk. This elaborate pattern of yolk uptake was also discovered in several snake species. In the future, we hope to further our studies by examining a few turtle species to ascertain if this pattern is ancestral for all reptiles.

#### 2.

## BUILDING A SEGMENTED BODY: THE CELL DYNAMICS OF SEGMENTATION AND ELONGATION

Nicole Duan '18 Faculty Sponsor: Terri Williams

Segmentation is a key feature of arthropod diversity. Most arthropods add segments during development from a posterior region called "the growth zone", which is the site of elongation and segment patterning. While segment patterning is studied in diverse taxa, the cell behaviors underlying elongation are less well known. The prediction for a growth zone is that there is a posterior region of undifferentiated cells dividing continually to provide the tissue required for new segments. We tested this model by examining cell division patterns in the posterior growth zone in a crustacean, Thamnocephalus platyurus, that adds segments after hatching. Our findings do not support this model of growth zone elongation: by looking at cells undergoing either S phase or M phase, we find that the rate of mitosis in the growth zone was surprisingly low. Interestingly, these data show that DNA synthesis is spatially organized in the growth zone, with distinct anterior and posterior domains of cell cycling. Cells in the anterior growth zone undergo an apparent synchronization resulting in all cells of the newly specified segment being in S-phase. Cell cycle domains in the growth zone are correlated with expression of Wnts and pharmacological knockdown of Wnts disrupts cell orientation and morphology in the growth zone. Additionally, cell cycle regulators, string and cyclins, also map to discrete growth zone domains in patterns that suggests a modified cell cycle, with a shortened or lost G1 phase, and coincide with boundaries of key segmentation genes. Overall in the growth zone, we find low numbers of cells in mitosis and cell cycle regulation tightly correlated to segmental patterning.

#### 3.

#### A NEW SPECIES OF *DIGENEA* (RHODOMELACEAE, CERAMIALES) BASED UPON A MOLECULAR ASSESSMENT AND MORPHOLOGICAL OBSERVATIONS OF PLANTS HISTORICALLY KNOWN AS *D. SIMPLEX* IN BERMUDA

Bilal Hamzeh '19 Faculty Sponsors: Craig W. Schneider; Gary W. Saunders

Using mitochondrial COI-5P and plastid *rbc*L genetic markers, the red algal species previously identified as *Digenea simplex* in Bermuda is found to be distinct from this species found in the Adriatic (type locality) and Mediterranean seas and other localities worldwide. This finding warranted a comparative study of the morphological characteristics of Bermuda specimens and *D. simplex* from the type locality, as well as other congeners from tropical seas. Our data show the Bermudian specimens to be morphologically, as well as with genetically, distinct from *D. simplex* necessitating the description of *D. arenahauriens* sp. nov. for plants found in the islands. It represents the first species partitioned from the pantropical *D. simplex* and we present genetic evidence of additional undescribed isolates in the species complex requiring further study.

#### 4.

# **REPLACING A 65 AMINO ACID SERRATE SEQUENCE WITH THE VERTEBRATE HOMOLOG JAGGED TO INVESTIGATE EFFECTS ON NOTCH RECEPTOR ACTIVATION**

Tyler Seckar '18 Faculty Sponsor: Robert Fleming

The gene Serrate encodes a ligand that can control the activity of the Notch cell-to-cell signaling pathway. Previous studies in the Fleming lab have demonstrated that nearly half of the Serrate protein coding regions can be deleted without significant loss of Serrate activity when compared to wild type Serrate in the Drosophila wing. In addition to this, the lab has found that a previously uncharacterized 65 amino acid segment of the juxtamembrane domain of the Serrate protein is necessary for Serrate to activate the Notch receptor. In an effort to determine if the 65 amino acid sequence contains certain properties or architectural patterns intrinsic to the activation of the receptor, the 65 amino acid segment was replaced with the comparable portion of the vertebrate Serrate homolog known as Jagged. Due to many difficulties in obtaining this construct, a series of troubleshooting experiments was conducted to determine if the techniques being employed were at fault or if the construct had inherent complications with transformation into competent bacterial cells. The ultimate objective of this work is to discover if the overall architecture of the receptor is different between the normal and the vertebrate homolog and if similarities are tied to the receptor's function and ability to activate the pathway.

#### 5. GENETIC SCREENING FOR GENES INTERACTING WITH SERRATE DURING NOTCH SIGNALING IN *DROSOPHILA MELANOGASTER* Fabiola Yun '18

Faculty Sponsor: Robert Fleming

Notch signals play a critical role during organismal development by promoting or suppressing cell proliferation, cell death, acquisition of specific cell fates, or activation of differentiation programs. In *Drosophila melanogaster*, the Notch ligand Serrate extends through the cell membrane and contacts the Notch receptor to mediate changes in gene expression. The objective of this study was to screen for potential genes that have the ability to modify the interaction of Serrate with Notch and affect the Notch signaling pathway. A modified Serrate gene called NCleve was inserted into the *Drosophila* genome and driven under a Serrate enhancer (H<sub>2</sub>) to generate a wing vein phenotype. Male wild type fruit flies were mutagenized with ethyl methanesulfonate and mated to the H<sub>2</sub>NCleve homozygous female fruit flies. In total, 6118 flies among the offspring were screened for mutations that altered the expected H<sub>2</sub>NCleve phenotype, and one mutation was isolated. It is believed that this mutation is an enhancer of Notch signaling. This mutation, and others from additional screens, will be mapped and further studied to determine the mechanism of its effects on the Notch signaling pathway.

## **CHEMISTRY**

#### 6.

**TRITYL CATIONS AS TUNABLE CATALYSTS IN DOUBLE ADDITION REACTIONS** Maria Boucher '20

Faculty Sponsor: Cheyenne Brindle

Catalysts are often used in chemistry to speed up the rate of reactions. The ability to tune these catalysts to make them more or less reactive allows us to select the most efficient catalyst for a given reaction. Since trityl compounds are easily tuned, they have promise to be effective catalysts. Due to this, trityl compounds were used as catalysts in double addition reactions. Catalysts tuned to optimize each double addition reaction were used to react 1.1 equivalents of aldehyde with 2 equivalents of indole. To determine the scope of these reactions, the catalyst and aldehyde used in the addition reactions were varied. Previous results showed that the reaction of most aldehydes could be catalyzed effectively with malachite green, a fairly weak trityl catalyst. However, some substrates such as isobutyraldehyde and acetophenone gave low yield even after long reaction times with this catalyst. To improve the reaction, more reactive, pre-catalysts were used. By tuning the trityl cations to be more reactive, the rate of these reactions could be increased. Although many of these reactions resulted in low yields, some substrates were found to give improved results. Others, like acetophenone, did not improve with catalyst tuning. Isolation of an indole-catalyst adduct suggests that the catalyst reacts with the indole rather than activating the ketone in these cases.

#### 7. REDUCING DICTYOSTELIUM DISCOIDEUM CELL ADHESION IN MICROFLUIDIC DEVICES BY USING SUPPORTED BILAYER COATINGS Julia R. Clapis '18

Faculty Sponsor: Michelle L. Kovarik

The research goal is to understand how the purity and composition of a supported lipid bilayer coating affects its performance in a microfluidic device. Due to the small channel size, microfluidic devices are particularly susceptible to fouling by cells or biomolecules when they are used for biological applications. Supported lipid bilayers are a convenient solution to this problem because they spontaneously form inside channels and serve to reduce fouling. Phosphatidylcholine (PC) is a common component of cell membranes and is often used in supported lipid bilayers due to its zwitterionic head group. In one study, phosphatidylcholine was extracted from boiled chicken eggs, and small unilamellar vesicles were made from the lipid before and after column purification. The effect of lipid purity was studied by comparing the electroosmotic flow in channels coated with in house lipids to that of commercial lipid coatings. Column purification of the extracted lipid increases the time until failure of the microfluidic device by around 20 minutes. Although the run times of the devices with commercial and unpurified lipids were similar, the commercial lipids afforded slightly faster and more efficient separations. In the second study, the composition of the bilayer was altered to study the effect of a net negatively charged bilayer on reducing adhesion of *Dictyostelium discoideum* cells. The bilayer composition varied from 0 to 30 mole % phosphatidylglycerol, a negatively charged lipid, with the remainder consisting of phosphatidylcholine. Cell adhesion was measured by allowing cells to adhere to the channels for 15 minutes, then counting the cells before and after rinsing the channels. Although it was originally suspected that an increased negative character would reduce cell adhesion, preliminary data suggests that the opposite is in fact true. Around 90% of cells washed away has been observed consistently with the 0% PG containing bilayer, while greater cell adhesion and higher variability were observed as % PG increased. Due to this unexpected trend, the effect of a net positively charged bilayer will be studied in the future. By better understanding how lipid purity and lipid composition influence failure by fouling, microfluidic devices can be optimized for single cell analysis.

#### 8.

## SINGLE CELL ANALYSIS OF REACTIVE OXYGEN SPECIES IN *DICTYOSTELIUM DISCOIDEUM* USING CHEMICAL CYTOMETRY

Jessica Duong '19

Faculty Sponsor: Michelle L. Kovarik

Cells have heterogeneous responses to stress despite their genetic uniformity. In order to report on individual heterogeneity, cells must be analyzed singularly. The purpose of this study is to determine the heterogeneity of oxidative stress responses in *Dictyostelium discoideum*. Hydrogen peroxide was applied exogenously to the cells to increase the level of reactive oxygen species (ROS). Dichlorodihydrofluorescein diacetate (DCFH<sub>2</sub>-DA) was loaded into cells to indicate the presence of ROS and carboxyfluorescein diacetate was loaded as an internal standard. DCFH<sub>2</sub>-DA was loaded at 200  $\mu$ M to compete with the level of native antioxidants in the cell. The cells were lysed on a microchip with a laser, releasing the dyes which were then electrophoretically separated and detected by fluorescence. Results suggest that cells respond more heterogeneously with hydrogen peroxide exposure. Future work is directed towards testing a range of peroxide concentrations from sublethal to lethal.

9.

## SYNTHESIS OF TURBOMYCIN ANALOUGES FOR THE DEVELOPMENT OF NEW ANTIBIOTICS: MODIFICATION OF THE PHENYL REGION

Matthew Epstein '19

Faculty Sponsor: Cheyenne Brindle, American Chemical Society, CO-ADD

Overuse of antibiotics in recent years has resulted in the emergence of numerous strains of resistant bacteria. As the development of novel antibacterial agents is not economically fruitful for pharmaceutical companies, the demand for new antibiotics has not been met. In a 2002 metagenomic study of soil bacteria, Gillepse et al. isolated turbomycin A and B, and determined that these two triaryl cations have broad spectrum antibiotic properties. Our research goal is to structurally modify these structures by synthesizing analogues to determine the structural features of the molecule that are important for biological activity. Previous work focused on modification of the indole component of the molecule. This work indicated that isopropylation of the indole nitrogen maximizes antibacterial activity without deleterious off target activity toward human cells. This summer, our research focused on both the steric and electronic modification of the phenyl region of the molecule, yielding analogues containing both a modified phenyl region and an isopropylated indole component. Two different protocols were utilized for this, to varying levels of success, and the analogues were sent to an independent lab for testing of their biological activity. Future work involves the optimization of a protocol for the synthesis of analogues that contain a modification to the phenyl component, but with only one of the indole components.

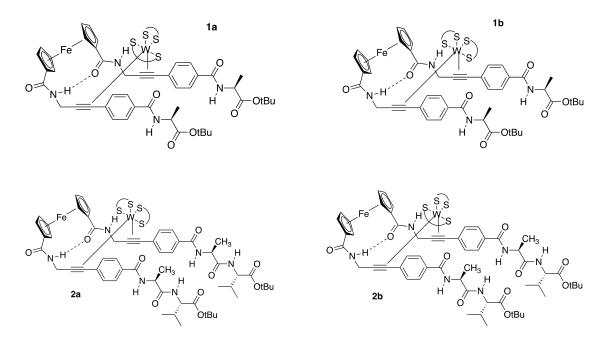
10.

## SYNTHESIS AND CHARACTERIZATION OF PEPTIDE DERIVATIVES OF A CONSTRAINED BIMETALLIC RING SYSTEM

Josephine Frempong '18 Faculty Sponsor: Timothy P. Curran

The main goal of this project was to investigate whether amino acid amide NH protons of peptides linked to a rigid, bimetallic ring system will engage in intramolecular hydrogen bonding. A second goal was to determine if the rigid, bimetallic ring system will be altered as the peptide chains are made longer. The bimetallic ring system, previously discovered in the Curran lab, is made of a ferrocene-dialkyne system coordinated to a tungsten complex  $[W(dmtc)_2(CO)_3]$  such that the two alkynes are held in the syn orientation by the tungsten complex. Thus, peptides attached to this ring system via the two alkynes are hypothesized to be in close proximity and have a possibility of intramolecular hydrogen bonding interactions. To study intramolecular

hydrogen bonding interactions in peptides, an alanine dipeptide ferrocene-phenylacetylene complex (**1a-b**) was successfully synthesized by attaching para-substituted alanine amino acid esters to a bimetallic ring system and coordinating a tungsten complex to the two alkyne ligands. A benzene spacer was used to link the peptides to the alkynes in the ring system. A tetrapeptide derivative (**2a-b**) of **1a-b** was also synthesized by attaching alanine and valine amino acids to the bimetallic ring system and coupling it to the tungsten complex. An inseparable mixture of diastereomers (**1a-b** and **2a-b**) was obtained in each case. Intramolecular hydrogen bonding was observed between the amide NH protons in the bimetallic ring system but no other intramolecular hydrogen bonding interactions were observed between the peptide strands. These products were purified using flash chromatography and characterized using <sup>1</sup>H-NMR, <sup>13</sup>C-NMR and ES-MS. The purity of the products was ascertained using HPLC and TLC and the conformations of the products were confirmed by DFT calculations from Hope College. The detailed synthesis and characterization procedures will be shown in the poster.



#### 11. EXTRACTION AND RECOVERY OF ALDEHYDES AND HIGHLY REACTIVE KETONES FROM MIXTURES

Max Furigay '19 Faculty Sponsor: Cheyenne Brindle

Purification techniques such as distillation, recrystallization, or column chromatography can be time consuming, wasteful in terms of atom efficiency, or complicated to perform, whereas extraction is relatively quick and easy. By reacting certain aldehydes with saturated sodium bisulfite, it is possible to extract these carbonyl compounds into an aqueous solvent layer, thus purifying the mixture. However, some carbonyl compounds, such certain ketones, do not react with bisulfite, allowing them to remain in an organic layer and allowing a separation of these fairly similar carbonyl compounds through extraction. The procedure can be modified for imine purification by performing a solid sodium bisulfite filter. This avoids the need for water, which is not compatible with imines due to hydrolysis. Additionally, conditions were found to easily recover the reactive carbonyl compound by reversing the bisulfite reaction through basification and re-extraction, which results in pure aldehyde recovery. An enolizable aldehyde tested with this method was found to decompose, so future work will aim to address this problem.

#### 12.

## SYNTHESIS AND CHARACTERIZATION OF CYCLIC ORGANOMETALLIC PEPTIDES

Cosmic Gober '18 Faculty Sponsor: Timothy P. Curran

The objective of this project is to synthesize and characterize organometallic peptides that have the potential to adopt  $\beta$ -sheet conformations. The desired organometallic peptide contains a bimetallic ring system that has a ferrocene unit at one end, a tungsten bis-alkyne at the other end, and two peptide chains attached to the two alkynes. To synthesize this complex, we needed first to form a ferrocene diamide and an alanine derivative acylated with 4-iodoaniline. These two molecules are coupled together through a Sonogashira reaction. Finally, the resulting dialkynylpeptide is reacted with W(CO)<sub>3</sub>(dmtc)<sub>2</sub> to produce the bimetallic ring system. The intermediate and final compounds were characterized for purity using TLC and HPLC. The identities of the intermediate and final compounds were established by <sup>1</sup>H NMR, <sup>13</sup>C NMR and MS. Whether the final product adopts a  $\beta$ -sheet conformation can be determined using NMR, Xray crystallography, and density functional theory (DFT) calculations. Details regarding this work, which is funded by an NSF grant, will be presented.

#### 13.

#### SYNTHESIS OF METALLACYCLIC DIPEPTIDES IN AN ANTI-PARALLEL β-SHEET CONFORMATION THROUGH TUNGSTEN COORDINATION TO TWO ALKYNES Paul Handali '18

Faculty Sponsor: Timothy P. Curran

Tungsten is a transition metal with the capacity to form air stable complexes with alkyne ligands. It potentially can be coordinated to dialkynyl peptide ligands to form metallacyclic peptides in a  $\beta$ -sheet conformation. Prior work has shown that most of these metallacyclic peptides are not constrained to a single conformation, but rather a multitude of possible conformational isomers. Recently in the lab, a novel constrained molecule that features both ferrocene and a tungsten bisalkyne complex was found to adopt only one solution conformation. Since then, further work has examined whether peptides appended to the two alkyne ligands will associate with each other as do the two peptide strands in a  $\beta$ -sheet. Two derivatives that have been successfully synthesized are alanine and glycine derivatives. Details of their synthesis and characterization, which was supported by a grant from the National Science Foundation, will be presented on this poster.

#### 14. KETOGENIC DIET INDUCED METABOLIC CHANGES INFLUENCING NEUROPROTECTION IN SH-SY5Y CELLS Lucy Honeycutt '18

Faculty Sponsor: William H. Church

The ketogenic diet is a high-fat, low-carb diet that has been used widely and successfully for the treatment of seizures and has recently gained recognition for its neuroprotective effects against neurotoxins. However, the mechanism by which the ketogenic diet provides neuroprotection is still unknown, though it is thought to be a result of metabolic changes such as increased ATP stores and decreased reactive oxygen species (ROS) production. Previous work on this project has resulted in the development of a ketogenic diet-type cell culture media. The present work will investigate whether this media provides neuroprotection from rotenone toxicity, and, if so, what metabolic changes have occurred within the cells that may be influencing this neuroprotection. Undifferentiated SH-SY5Y human neuroblastoma cells were cultured in either normal media (LSFM), low glucose media (LG), or ketogenic diet media (LG KD). Following 48 hours of media treatment, cells were stressed with 500 nM and 1 µM rotenone for 24 and 48 hours before being analyzed for ATP and superoxide (ROS) content. After 24 hours, cells treated with KD and stressed with 500 nM and 1 µM rotenone were found to have significantly lower ATP levels than LSFM cells (p < 0.001). Cells treated with KD and stressed for 24 hours with 500 nM rotenone were also found to have significantly lower superoxide levels than cells treated with LSFM (p < 0.01). No significant differences in ATP content or superoxide levels were found after 48 hours of treatment (p > 0.05). Future work on this thesis project will investigate metabolic differences between undifferentiated and differentiated cells, as well as differences in cell death between treatment groups.

#### 15.

#### **TRITYL CATION-CATALYZED COMPLEX MOLECULE SYNTHESIS** Vanessa Jones '19 Faculty Sponsor: Cheyenne Brindle

The use of multiple single addition reactions to produce a single large product is an important technique for building molecular complexity. This allows the product to be formed piece by piece, greatly diminishing the possibility of creating multiple products, rather than the intended product. We create large products from the sequential addition of two different indoles to an imine, using triarylmethyl cation catalysis. This requires the synthesis of imines from aldehydes. The imines are produced by adding different types of aldehydes to paranitroaniline. The solid produced from this reaction were then recrystallized to ensure purity. This step was then tested using nuclear magnetic resonance. The imine was then reacted with indole to produce a single addition product. This technique allows the complicated product to be produced that would otherwise be nearly impossible to synthesize. These first few steps still have to be perfected, the most successful temperature and concentration must be found for each reaction performed, in

order to continue on. Future work will include sequential addition of indoles, but this can only be possible after the first step of the reaction is optimized.

#### 16.

#### DETERMINING HOW LOADING METHOD AFFECTS THE METABOLISM OF EXAGENOUS MOLECULES IN INTACT CELLS OF *DICTYOSTELIUM DISCOIDEUM*: COMPARISON OF PINOCYTOSIS, ELECTROPORATION AND MYRISTOYLATION

Grigorii Kalminskii '20, Rahuljeet Chadha '20, Allison J. Tierney '17 Faculty Sponsor: Michelle L. Kovarik

By loading molecules into living cells, one can monitor cellular metabolism, track various metabolic pathways and modify cellular activity. In order to successfully load exogenous molecules, one has to select an optimal loading method and identify the optimal conditions for the specific model organism. The ideal, optimized loading method will produce rapid, uniform, controllable loading for a wide range of molecules with minimal reagent consumption, analyte degradation, and cell stress. In this experiment, three promising loading methods were optimized and compared in Dictyostelium discoideum. Methods included pinocytosis, electroporation and peptide myristoylation. Pinocytosis is based on cells first endocytosing surrounding media, and then applying an osmotic pressure to lyse formed pinosomes, hence releasing their contents into the cytosol. Electroporation involves a controlled disruption of the cell membrane followed by the entry of molecules through the artificially created pores. Peptide myristoylation is a method, which involves an addition of certain hydrophobic myristol group to the amino acid chain to facilitate the diffusion of the resulting peptide through the membrane. Of the three loading methods, myristoylation produced the greatest loading efficiency. Electroporation produced the most uniform cytoplasmic loading, while overall cell brightness was distributed more normally around the mean when cells were loaded by pinocytosis. The cells had shown similar viability after being loaded via different methods, while peptide reporter had a longer half-life in vivo after loading with electroporation than after loading with myristoylation due to lysosomal compartmentalization. Future work of this project will include analyzing the phosphorylation activity of protein kinase B on peptide reporter molecules loaded by each method to determine whether loading method affects reporter output.

#### 17.

#### SYNTHESIS, CHARACTERIZATION AND DETERMINATION OF THE EXHIBITION OF BETA-SHEET CONFORMATION IN AN ORGANOMETALLIC COMPLEX Thanh Nguyen '19

Faculty Sponsor: Timothy P. Curran

This research involves synthesis and characterization of a dialkynylferrocene with amino acid esters appended to the two alkynes, the synthesis and characterization of the complex of this dialkyne with tungsten (1), and the conformational analysis of 1. It has been proposed that intramolecular hydrogen bonds will form between the two peptide chains in 1, and that this hydrogen bonding will resemble that seen in beta-sheets. The use of an organometallic moiety to generate model beta-sheet systems would allow for experiments (like redox reactions) on these

systems that are only possible with metal ions. The general strategy of this research is to synthesize the dialkynylferrocene, complex it to tungsten, and then purify and examine the target molecule. Synthetic methods in both organic and inorganic chemistry will be used. To determine the conformation of the tungsten complex methods such as NMR spectroscopy, X-ray crystallography and DFT calculations will be employed. This poster will present the work accomplished so far on this project, which is funded by a grant from the National Science Foundation.

18.

## SYNTHESIS AND CHARACTERIZATION OF PEPTIDE DERIVATIVES OF A CONSTRAINED BIMETALLIC RING SYSTEM

Michael Phillip '19

Faculty Sponsor: Timothy P. Curran

The overall goal of this project, which was funded by NSF grant 1464761, is to create a model beta sheet by attaching peptides onto a bimetallic ring system. Using the model bis-alkyne tungsten complex synthesized by Allison Lawrence '10 as a framework, Niru Pokharel '15 and Lauren Davidson '16 briefly investigated if the rigidity of the model complex was affected by the groups attached to the alkyne group. It was found that the attachment of different phenyl groups to the alkyne group did not affect the rigidity of the model complex. The aim of this experiment was to characterize the Boc-valine and valine t-butyl ester derivatives of the bis-alkyne complex by coupling the ferrocene dialkynyldiamide with the two valine derivatives. To synthesize the tungsten complex, the dialkynylpeptide derivatives of valine were reacted with  $W(dmtc)_2(CO)_3$ . The resulting compounds from both reactions were purified using flash chromatography. After purification, both the dialkynylpeptide derivative of valine and the tungsten complex were characterized using <sup>1</sup>H NMR, ES-MS and HPLC. The synthesis and characterization of the dialkynylpeptide derivatives will be described in this poster.

19.

#### PREPARATION OF FERROCENE-COUPLED ALKYNE COMPOUNDS THAT ADOPT A PROPOSED BETA SHEET CONFORMATION WHEN COORDINATED TO TUNGSTEN

Joseph P. Sanderson-Brown '18 Faculty Sponsor: Timothy P. Curran

Dialkynyl peptides bonded to ferrocene have been shown to cyclise into bimetallacyclic complexes when coordinated to tungsten. The molecule 1,1'-ferrocenedialkynyldiamide has further been shown to adopt an anti-parallel  $\beta$ -sheet conformation when cyclised using tungsten. The intramolecular bonding between the amide groups of the peptide chains determine the conformation and orientation adopted. Alanine and phenylalanine peptide derivatives of 1,1'-ferrocenedialkynyldiamide have been previously synthesised and analysed but were not successful in forming the desired rigid  $\beta$ -sheet. Derivatives of 1,1'-ferrocenedialkynyldiamide were investigated; specifically, an alkyl (CH<sub>3</sub>) derivative sourced from acetic acid, and a boc derivative sourced from di-tertbutyl dicarbonate. Acetic acid was reacted with 1-amino-4-chloro-

2-butyne under basic conditions (Et<sub>3</sub>N) to form CH<sub>3</sub>CONHCH<sub>2</sub>CCCH<sub>2</sub>Cl. The chlorine of CH<sub>3</sub>CONHCH<sub>2</sub>CCCH<sub>2</sub>Cl was substituted via an overnight reaction with NH<sub>3</sub> in MeOH at 70°C to yield CH<sub>3</sub>CONHCH<sub>2</sub>CCCH<sub>2</sub>NH<sub>3</sub>Cl. The substitution reaction was followed by the attempted coupling of CH<sub>3</sub>CONHCH<sub>2</sub>CCCH<sub>2</sub>NH<sub>3</sub>Cl to a prepared ferrocene diacid chloride using dimethylformamide (DMF) and diisopropylethylamine (DIEA). The coupling was unsuccessful. The alkyl group was replaced with a boc group. An identical reaction pathway was attempted but similar problems were encountered with the coupling reaction. 1-amino-4-chloro-2-butyne was reacted with the ferrocene diacid chloride under basic conditions (DIEA) to form Fer(CONHCH<sub>2</sub>CCCH<sub>2</sub>Cl)<sub>2</sub>. The chlorine was substituted via an overnight reaction with NH<sub>3</sub> in MeOH at 70°C to yield Fer((CONHCH<sub>2</sub>CCCH<sub>2</sub>NH<sub>3</sub>Cl)<sub>2</sub>). The resulting hydrochloride salt compound was reacted with di-tertbutyl decarbonate under basic conditions to give Fer(CONHCH<sub>2</sub>CCCH<sub>2</sub>NHCOOC(CH<sub>3</sub>)<sub>3</sub>)<sub>2</sub>. The coupled compound was isolated and will be coordinated to tungsten in the near future to determine if the desired intramolecular hydrogen bonds form and if the rigid  $\beta$ -sheet is adopted. The structures of the reaction products were confirmed using electrospray mass spectrometry and proton NMR spectroscopy.

### **COMPUTER SCIENCE**

20.

#### AAProxy: ANONYMOUS AUTHENTICATION PROXY

Brian Cieplicki '19, Zorawar Moolenaar '20 Faculty Sponsor: Ewa Syta

Services on the internet constantly attempt to profile their users to provide them with a more personalized service. From advertisements to content-delivery, in some way every service tries to improve and target its audience with relevant content in hopes of either selling them products or the service itself. In trying to create this individualized experience for the user, services require background information to know the client's preferences.

Unknowingly or in lieu of saving time, most users will grant services the ability to "know who they are" and grant these services access to their "basic profile" and other information through single-sign on. Further, by reusing user names and other similar information across different services, users create a large digital footprint that corporations, state-funded actors, and other malicious users could potentially use to track users' habits and conduct highly specialized attacks, such as social-engineering attacks, against them. In a scenario where a service becomes compromised, this also leaves users vulnerable and exposed for anyone to potentially obtain their sensitive information.

Anonymous Authentication Proxy (AAProxy) aims to serve as an intermediary system for services to afford their users the protection of masking their real identities by using pseudo-identities, and authenticating users at the service's discretion.

## **ENGINEERING**

#### 21. POWER DELAY PROFILE REALIZATION WITH SIMULATED VEHICLE MOBILITY DATA Kun Chen '18

Faculty Sponsor: Lin Cheng

Inter-vehicle communication in urban settings is dynamic and extremely mobile. Relatively low antenna heights on communicating vehicles and roadside infrastructure further compounds the challenge of signal propagation. To help realize the challenges we would face in real-time situations, we utilize Simulation of Urban Mobility (SUMO) data to generate the power delay profile of receiver vehicles during vehicle to vehicle (V2V) and vehicle to infrastructure (V2I) communications. Simulation of multi-channel communication using SUMO data helps to provide insights into how signal propagates during real-time traffic situation.

#### 22.

#### AUTOMATIC DETECTION OF HEART SOUNDS AND CLICKS WITH MURMURS

Kun Chen '18, Fabiana Guajardo '20 Faculty Sponsors: Taikang Ning, Lin Cheng

Our research consists of creating an algorithm that automatically detects heart sounds and clicks. The algorithm can accurately detect the first heart sound  $(S_1)$ , the second heart sound  $(S_2)$ , and any systolic or diastolic click that occurs. Phonocardiogram data are divided into short data segments in several milliseconds where defined features carrying essential information regarding loudness and marked time locations are extracted. These features are examined through an expert system to identify  $S_1$ ,  $S_2$ , systoles, diastoles, and clicks. The performance of the algorithm was validated with a variety of clinical heart sounds episodes featuring murmurs and clicks.

#### 23.

## APPLICATION OF NEURAL NETWORKING IN RESPIRATION MONITORING AND APNEA DETECTION

Akrit Mudvari '18, Ahmed Eldmerdash '20 Faculty Sponsor: Taikang Ning

Apnea is a common sleep disorder with detrimental effects on health, performance and safety. This underlying research uses modern computing technologies to assist the diagnosis of sleep apnea. The method is based on several signal processing algorithms that can extract useful respiration features such as signal energy, respiration rate, and respiration waveform slope. These features are examined, and respiration signals are categorized into normal breathing, apnea, and breathing episodes combined with body motion artifacts that are caused by body

movement. Single layer neural networks are employed to differentiate between instances of normal breathing and breathing with motion artifacts.

#### 24.

#### ANALYSIS OF CHEMICAL KINETICS AND COMBUSTION COMPUTATIONS OF DIMETHYL METHYLPHOSPHONATE (DMMP)

Alex Sinson '20, Donovan Palmer '20, Farhan Rozaidi '20 Faculty Sponsors: John D. Mertens, Olivier Mathieu, Eric Petersen

Shock tubes are essentially two pipes—one with high pressure, and one with low pressure separated by a barrier, known as a diaphragm, that is ruptured to generate supersonic shock waves to heat gases to controlled high temperatures and pressures. With well-designed experiments, optical diagnostic measurements of chemical species in the hot gases can be used to measure the reaction rate coefficient-known as variable k-of a specific gas-phase chemical reaction. Experimental measurements shared from shock tube experiments performed in the lab of Prof. Eric Petersen at Texas A&M University were used to study the combustion of dimethyl methylphosphonate (DMMP), which has the chemical structure C<sub>3</sub>H<sub>9</sub>O<sub>3</sub>P. The optical diagnostic measured the time histories of an electronically excited state of hydroxide (OH\*), which occurs when a combustion reaction takes place. DMMP is being studied as a potential sarin surrogate, to potentially improve sarin gas destruction. An extensive chemical kinetics model was developed for DMMP by merging previously published reaction mechanisms, one containing a database of the DMMP reactions (Glaude, Melius, Pitz, & Westbrook, 2002), and two hydrocarbon mechanisms (Matheiu et al., 2016). Using these large compiled mechanisms and the Cantera (Goodwin, Moffat, & Speth, 2017) open source differential equation solver, the important chemical reactions were identified, and sensitivity analysis and production rates were used to understand and develop a flowchart of the combustion process. Several rate coefficients were adjusted within acceptable values so that the calculated OH\* profiles aligned with the measured OH\* profiles, and a preliminary measurement of the rate coefficient of the bimolecular DMMP dissociation reaction was achieved for the first time. This work is ongoing, and the new reaction mechanism is being used to identify conditions for additional DMMP experiments that will be performed at Texas A&M in the next month.

### **ENVIRONMENTAL SCIENCE**

#### 25.

#### THE EFFECTS OF CLEAR-CUTTING ON SOIL ALUMINUM AND CALCIUM LEVELS IN THE WHITE MOUNTAIN NATIONAL FOREST, NEW HAMPSHIRE AND MAINE

Jack Agosta '17, Kevin Oleskewicz '19

Faculty Sponsors: Jonathan Gourley, Andy Coulter, U.S. Forestry Service

Clear-cutting is and has been a highly efficient and profitable method of harvesting timber for well over a century in the White Mountain region of New Hampshire. However, it can have serious negative effects on the surrounding forest ecosystem. One of these effects is the significant alteration of soil nutrient concentrations due to increased soil nutrient leaching and runoff. This study focuses on measuring changes, if any, in soil aluminum and calcium levels. Aluminum is a key nutrient for plant growth and calcium is important for root, leaf and flower stability and health. Working with the USDA Forest Service, soil samples were taken on transect lines at multiple different clear-cut study sites in the White Mountain National Forest in New Hampshire and Maine. Samples were taken from the O and B- soil horizons at each site following a series of random transects. The site are as follows: Hogsback, Millstone, and Douglas Brook. The samples were processed using acid digestion before being run through the Inductively Coupled Plasma-Optic Emission Spectrometer (ICP-OES) where aluminum and calcium concentrations were measured.

The same sites/transects were sampled and analyzed both prior-to and post clear-cutting (at both 1 and 2-year intervals) to measure and compare baseline nutrient concentrations with post clear-cut nutrient concentrations under the hypothesis that measurable effects, if any, may not manifest until several months after logging operations have concluded.

#### 26.

## CHARACTERIZING HOW SUNLIGHT IMPACTS THE EFFICACY OF CURRENT OIL SPILL RESPONSE TOOLS

Cassia Armstrong '18

Non-Trinity Faculty Sponsors Collin Ward, Christopher Reddy, Woods Hole Oceanographic Institution; Department of Marine Chemistry and Geochemistry

Photochemical weathering is often considered too slow to impact the efficacy of chemical dispersants applied to floating oil in the aftermath of spills. However, using the Deepwater Horizon spill as a case study, we showed that photochemical oxidation was a dominant weathering process that oxidized over half of the floating oil on time-scales of days. These rapid changes to oil chemistry by sunlight suggest that photo-oxidation could conceivably impact the "window-of- opportunity" for chemical dispersant application during the emergency phase of a spill response. Here we show that on timescales of hours to days, sunlight substantially altered the chemical and physical properties of oil, which in turn decreased the efficacy of chemical dispersants used in response to the Deepwater Horizon spill. We further demonstrate that the impacts of photochemical weathering on the dispersant efficacy of Macondo Well oil greatly outweigh the impacts of evaporation, a weathering process regularly factored into oil spill response plans. Lastly, accounting for the decrease in dispersant efficacy due to photochemical oxidation, we revised downwards estimates for the amount of oil dispersed by aerial applications in the aftermath of the Deepwater Horizon spill. Collectively, our findings strongly suggest that photochemical weathering substantially impacts the "window-of- opportunity" to apply chemical dispersants in response to future oil spills in sunlit waters.

27.

ACCUMULATION OF HEAVY METALS IN GREATER HARTFORD AREA PONDS Shane McLaughlin '19, Brendan Lynch '20, Anna Maria Imwalle '20 Faculty Sponsor: Amber L. Pitt

Ponds in urban environments serve many purposes, including storm water catchment, recreation, and aesthetics. They are also crucial centers of biodiversity. We sought to investigate the health of ponds in the Greater Hartford Area and their suitability as wildlife habitats. We identified potential study ponds via Google Maps satellite imagery and through in-person assessment. We selected nine study ponds. We collected soil samples from three distinct sites at each pond and analyzed them for eight different metals using Inductively Coupled Plasma-Optical Emission Spectrometry and a Direct Mercury Analyzer. We compared the results to effect levels from MacDonald et al. (2000). Cadmium, copper, zinc, mercury, and lead were found above the threshold effect concentration (TEC) in multiple study ponds. Only one pond, Pope Park pond, had all three sample sites below the TEC for all metals. Only lead was found above the probable effect concentration (PEC) where adverse effects to wildlife are likely. Lead levels exceeded the PEC at four ponds in at least one sampling site, and only two ponds had all three sampling sites below the TEC for lead. While only preliminary, our results suggest potential consequences for wildlife. The metal levels found stood directly in contrast to what we expected from a visual assessment of the ponds-generally, the less aesthetically pleasing ponds had lower levels of metals than the more picturesque, naturalized ponds with vegetation and other characteristics that may attract wildlife. Wildlife may thus be attracted to ponds that do not provide suitable habitat. This could signify a threat to the health and future of these wildlife populations. Future work will include analyzing land use data and other factors that may influence heavy metal concentrations in ponds, and investigating bioaccumulation of these metals in wildlife.

#### 28.

#### **THE EFFECT OF MERCURY POLLUTION ON TERRESTRIAL VASCULAR PLANTS** Joseph Ruggiero '19

Faculty Sponsor: Jonathan Gourley

The presence of heavy metals in soil have a significant impact on quality of soil and plant life. Previous studies have shown that the accumulation of mercury in various parts of terrestrial vascular plants will result in reduced growth and impaired metabolism. The goal of this study will be to observe and trace the accumulation of mercury within different sections of vascular plants and soil samples. Plants will be grown for approximately 6-8 weeks until maturity. Mature plants will be subjected to different concentrations of mercury over multiple trials for a varying amount of days and weeks before cultivating and dividing into three parts: the root, stem and the leaves. Previous analysis has indicated the highest concentration of Hg is measured in the roots, and the distribution of Hg throughout the plant is dependent on the species observed. Therefore, it is hypothesized that the bean plants cultivated will show the highest Hg concentrations in the roots due to their fine roots and rapid metabolism. The results of this study will be applied to other research projects, which will investigate the uptake of Hg by higher order terrestrial species over elevation change within the White National Forest.

### **MATHEMATICS**

#### 29.

#### CRAZY BIFURCATIONS: BIFURCATION AND STABILITY ANALYSIS IN A NONLINEAR MASS-SPRING SYSTEM

Alison Adamski '19 Faculty Sponsor: Ryan Pellico

In this research, analysis of the behavior of non-linear simple harmonic oscillators was conducted. With the help of MATLAB as a programming tool, the differential equations used in this research model spring-band-mass systems. The particular focus on the parameters, which represent the different physical constants that affect the system's response, helped make novel conclusions about the stable and unstable solutions for the O.D.E.s (Ordinary Differential Equations). Utilizing Newton's Method to find solutions and varying the physical parameters, bifurcation diagrams were studied in 2D and 3D to visualize the space of periodic solutions. New solutions and complicated behavior were discovered at specific values of the nonlinear parameter. We investigated stability by studying the eigenvalues of the linearized system, and from that, more insight into the effects of parameter changes and multiple solutions for these specific O.D.E. models were gained.

#### 30.

## MODELING RESOURCE FLOWS WITH STATE-DEPENDENT RANDOM WALKS ON NETWORKS

Sabina Adhikari '19 Faculty Sponsor: Per Sebastian Skardal

In this work, we introduce a state dependent random walk model to study the flow of resources through networks. Random walk models describe the trajectory of one or more random walkers through a network in subsequent time steps via certain transition probabilities that are typically assumed to remain constant. Here we allow these transition probabilities to change at each time step, specifically defining them in terms of the fraction of walkers present at network neighbors. We introduce a biasing parameter that allows us to study two cases: when walkers are preferentially funneled towards neighbors who currently have a (i) smaller or (ii) larger fraction of random walkers, i.e., towards poorer or wealthier neighbors, respectively. Quantitatively, we find that varying this parameter can either exacerbate or mitigate the inequality present when transition probabilities remain fixed. Moreover, more extreme choices of this parameter result in the typical fixed-point solution losing stability and giving rise to period-two solutions where the steady-state behavior oscillates between two states. These behaviors depend sensitively on networks structures; the fixed-point solution tends to remain stable for longer in dense networks in contrast to spare network, delaying the transition to steady-state periodic behavior.

#### 31. SYNCHRONIZATION IN NONLINEAR HARMONIC OSCILLATORS

Yesenia Garcia Balbuena '19, Daniel Melesse '20, Fumihiro Tamada '20 Faculty Sponsor: Ryan Pellico

Our research is focused on coupled harmonic oscillators with a phase shift parameter. Our coupled nonlinear model is based on combing two nonlinear oscillators, each with a spring and rubber band. Our model demonstrates when and how synchronization between the oscillators happens as we increase the coupling strength. It shows the angles between the large and small amplitude periodic solutions. Another model shows how varying the amplitude of the periodic forcing affects synchronization between the oscillators, in which quasi-periodic behavior was also observed. We used MATLAB to simulate and examine how the change in parameters affects these properties of the coupled system.

#### 32.

#### **SPECTRAL ANALYSIS ON GRAPHS RELATED TO THE BASILICA JULIA SET** Samantha Jarvis '19

UCONN Mathematics REU Program, National Science Foundation

We analyze the spectra of a sequence of graphs constructed from the Schreier graphs of the Basilica group. Our analysis differs from earlier work of Grigorchuk and Zuk in that it is based on a macroscopic decomposition of the graphs. This method gives precise information about the multiplicities of eigenvalues and consequently good information about the spectral measures of large graphs. It also permits a proof of the existence of gaps in the spectrum of limiting graphs.

#### 33. CHAOTIC DYNAMICS OF ASYMMETRIC COUPLED OSCILLATOR COMMUNITIES

Ryan Tassan '18, Matthew Bicknese '20 Faculty Sponsor: Per Sebastian Skardal

Coupled oscillator models and the synchronization dynamics they exhibit have a wide range of applications, ranging from cardiac pacemaker cells to the flow of electricity in power grids. In many real-world applications coupled oscillators are connected through complex networks with community structure, where two or more groups exist such that coupling is strong within each group, but relatively weaker between different groups. Here we study the synchronization dynamics of communities of coupled Kuramoto oscillators via appropriately defined order parameters as various system parameters are varied. We pay special attention to the case of three oscillator communities with varied asymmetry in the relative sizes of the communities. In particular, we show that this kind of asymmetry is enough to generate chaos in the macroscopic

system dynamics. Using numerical methods, we construct a bifurcation diagram that illustrates the routes towards chaos, e.g., through period-doubling cascades, and quantify the sensitivity to initial conditions using the spectrum of Lyapunov exponents. The Lyapunov spectrum then allows us to quantify the fractal dimension of the strange attractor in the chaotic regime.

### **NEUROSCIENCE**

#### 34.

## LONG-TERM BEHAVIORAL EFFECTS OF PRENATAL EXPOSURE TO THE KETOGENIC DIET

Amro M. Arqoub '18

Faculty Sponsors: Luis A. Martinez, David N. Ruskin, Susan A. Masino

For many women with epilepsy, the decision of which treatment options to pursue during pregnancy has critical implications both for their own health as well as their developing fetus. Fetal exposure to specific anti-epilepsy drugs (AEDs) confers a great risk for congenital malformations. The ketogenic diet (KD), a diet composed of mainly fats and low but varying levels of proteins and carbohydrates, is a non-AED therapy shown to be highly effective in treating epilepsy. The present study aims to investigate the lasting effects of prenatal exposure to the KD on social and anxiety-like behaviors in mice. In this study, adult female and male CD1 mice will be randomly assigned to either a KD or a control diet (CD)- composed of typical rodent chow diet- and then mated with an individual of the opposite sex from the same diet treatment. The young adult offspring of these mice will then be assessed for sociability, selfrepetitive behaviors, and depressive/anxiety-like behaviors. Since former research has shown that postnatal exposure to the KD improves sociability in mice, we expect that prenatal exposure will have similar effects. Moreover, we expect to replicate previous findings showing decreased depressive/anxiety-like behaviors following prenatal exposure to the KD. Improving our understanding of the lasting effects that the KD has on offspring is critical for helping prospective mothers with epilepsy choose the safest therapeutic option; furthermore, it may help uncover novel therapeutic possibilities for the KD, including serving as a prenatal preventative therapy for children at risk for developmental disorders such as autism.

35.

## ANALYZING THE EFFECTS OF COCAINE AND A KETOGENIC DIET ON BRAIN NEUROTRANSMITTER CONTENT IN RATS

Ahmad Chughtai '20 Faculty Sponsor: William H. Church

Chromatography was used to measure the neurotransmitter contents of rat brains following cocaine administration. Rat brains were dissected using razor blades and tissue samples from the nucleus accumbens, cortex, striatum, and midbrain were obtained. These samples were sonicated and filtered before being analyzed using an HPLC apparatus. Calibration standards containing known amounts of each desired neurotransmitter were created to allow for the identification of the neurotransmitters present in each sample. These standards were also used to create a calibration curve on the computer, which was then used to determine the amount of each neurotransmitter present in each sample using the Chromeleon 7.2 program.

#### 36.

#### THE KETOGENIC DIET SHIFTS CEREBROSPINAL METABOLOME AND HAS DIFFERENTIAL EFFECTS IN RESPONSIVE VS. NON-RESPONSIVE PEDIATRIC EPILEPSY PATIENTS

Natalie R. Freedgood '18 Faculty Sponsors: David N. Ruskin, Susan A. Masino

The high-fat low-carbohydrate ketogenic diet (KD) has shown significant anticonvulsant effects in treating refractory epilepsy in children. The difficulty with understanding how the KD works in treating epilepsy has to do with the challenges in conducting controlled studies in this population. In this study we investigated the effects of the KD on refractory childhood epilepsy by analyzing their cerebrospinal fluid metabolome using metabolomic analyses. Cerebral spinal fluid (CSF) was collected from 25 children with drug refractory epilepsy from the Karolinska Hospital in Stockholm, Sweden. CSF samples were sent to Metabolon for analysis of metabolites. 271 metabolites were identified. Patients with 100% (n=5) or 0% (n=5) seizure reductions were then further assessed. Statistical analyses included a principle component analysis, ANOVA, and t-tests. Principal component analysis showed a strong effect of KD treatment on metabolites, in that the baseline and during treatment groups are well separated across component 2's correlations. Comparing responders and non-responders for the hallmark metabolic effects of KD, there was a greater elevation of ketone bodies and a larger drop in glucose in responders. These data show that KD treatment strikingly modifies the central metabolome, and that responders may have a stronger metabolic response to the KD.

#### 37.

## THE MEDIAL PREFRONTAL CORTEX- BASOLATERAL AMYGDALA CONNECTION

Tom Gitchell '18, Natalie Bruno '20 Faculty Sponsor: J. Harry Blaise

The two regions of interest in our study are the medial prefrontal cortex (mPFC) and the basolateral amygdala (BLA). We stimulated the mPFC which is responsible for complex behavior, decision making and logical reasoning. We recorded in the BLA which is responsible for autonomic responses, it is particularly known to act in emotional circumstances. A stereotaxic surgery was performed to acquire a signal with characteristics specific to the mPFC-BLA pathway. This surgery is new in our lab and therefore much of our time this summer has been concentrated on finding consistent methodology to acquire a successful and consistent signal in each attempt. The signal we are looking for has a unique feature of a negative peak at a latency of 25ms. To carry out these surgeries electrodes are created including the ground/reference electrode, stimulating/bipolar electrode and recording/monopolar electrode. Once the electrodes are created the experimenters may start their surgery. The rat is put under anesthesia using initially isothesia liquid to relax the rat and then is injected with a mixture of

ketamine anesthetics at the intraperitoneal site and is then prepared for surgery. An incision is made and the skull of the rat is cleaned to the point where Lambda and Bregma can be identified. Lambda and Bregma are reference points as to where to implant the electrodes. Once the electrodes are in place, they are capped with dental cement and the rat is left for five to seven days to recover. Histology was performed to verify the location of electrode implant. With the connection between the mPFC and BLA established we can begin LTP recordings to find control data. We then hope to assess how various types of stress impact this synapse. In conclusion, we established a consistent and successful methodology for a surgery that examines the synapse between the mPFC and BLA.

#### 38.

#### TREATMENT MODULES FOR PROSPECTIVE MEMORY TRAINING POST TBI

Anna Hackett '20, Anna Lee '20 Faculty Sponsor: Sarah Raskin

Traumatic brain injuries (TBI) can lead to numerous difficulties in daily live. One major side effect of TBIs are loss of prospective memory function, or in other words, "remembering to remember." The goal of this study was to make a series of treatment modules that patients and practitioners could use as training guides to improve prospective memory in people with brain injury. The memory of each patient is first tested using the Memory for Intentions Test (MIST) a standardized test of prospective memory. Based on the results, the participant would then begin whichever module was determined to be best suited for his or her condition working from their baseline determined from the MIST. Seven modules were created: Enactment, Visualization, Increasing Cognitive Load, Increasing Cue-Intention Relatedness, Decreasing Cue Focality, Implementation of Intentions, and Time Perception training.

#### 39.

## EFFECTS OF THE KETOGENIC DIET ON BEHAVIORAL RESPONSES TO COCAINE IN MALE AND FEMALE RATS

Meghan Lees '18

Faculty Sponsors: Luis A. Martinez, Susan A. Masino, David N. Ruskin

Abstract: The ketogenic diet (KD) is a high fat, low carbohydrate and adequate protein formulation that has traditionally been used as a treatment for epilepsy; however, there is growing evidence that this diet has broader therapeutic potential due to its diverse, positive effects on nervous system function. Recent drug addiction studies suggest that activation of the brain adenosine system decreases behavioral responses to many drugs of abuse, including cocaine. Given that one consequence of the KD is an increase in brain adenosine, we sought to address whether the KD has potential as a novel therapy for drug addiction. In this study, Sprague-Dawley were placed male and female rats on а strict 6.6:1 (fat:[carbohydrates+protein], by weight) KD or control diet at 5 weeks of age and then maintained on those diets for 3 weeks prior to behavioral testing. During testing, rats received daily i.p. injections of cocaine (15 mg/kg/ml) or saline vehicle for one week, were abstinent for a subsequent week, and then all animals received a final challenge injection of 15 mg/kg/ml cocaine. Our preliminary results indicate that both males and females on the

KD showed a less robust increase in ambulatory activity and stereotyped responses as would normally be expected following repeated cocaine injections. These results suggest that the KD may indeed hold potential as a therapy for drug addiction. Future studies will focus on how the KD affects the reward pathway using conditioned place preference to cocaine, as well as exploring the neural mechanisms underlying the behavioral effects of this diet. **40.** 

#### ANALYZING THE EFFECTS OF PROBIOTICS ON SUPEROXIDE PRODUCTION BY HUMAN NEUROBLASTOMA CELLS

Morgan McKeown '20 Faculty Sponsor: William H. Church

Lifestyle (exercise) and nutrition (supplements, low calorie diets) have been implicated in maintaining appropriate cognitive function as humans age. As a result, understanding the mechanism(s) by which neurons are protected from toxin-induced neurodegeneration can provide useful information regarding these health benefits. This research project characterized an assay for an important measure of cellular health: mitochondrial superoxide radical (mSOR) production. SH-SY5Y neuroblastoma cells were subjected to hydrogen peroxide treatments and mROS levels were quantified using a commercially available kit. Following a 30 min. treatment with  $5\mu$ M H<sub>2</sub>O<sub>2</sub>, mSOR generation was attenuated with increasing concentrations of a probiotic plant dietary supplement (72hr pre-treatment; Immunity©, BodyandEden ) These results provide interesting preliminary data regarding the utility of using mitochondrial generated superoxide radical to explore neuroprotective mechanisms in a human neuroblastoma cell line.

#### 41.

#### CONCENTRATION-DEPENDENT ASCORBIC ACID DECREASES CELL VIABILITY IN UNDIFFERENTIATED SH-SY5Y CELL LINE

Georgia M. Mergner '18 Faculty Sponsor: William H. Church

Previous research in this lab has shown that ascorbic acid treatment is neuroprotective in differentiated SH-SY5Y cells. However, the use of ascorbic acid treatment in undifferentiated SH-SY5Y cells resulted in loss of cell viability as it acts as a pro-oxidant rather than antioxidant. This study sought to reproduce previous results using a new cell viability protocol based on literature discussed over the course of the summer. SH-SY5Y cells were grown in 96 well plates to 30-40% confluence over the course of 3 days. Undifferentiated cells were then treated for 1 hour with varying concentrations of ascorbic acid (0, 0.2, 0.5, 1 and 2 mM). Cell viability was determined using a commercially available fluorescent assay (LIVE/DEAD Viability Cytotoxicity Assay Kit; Molecular Probes). Fluorescent microscopy was used to assess viability assays. Results indicate that percentage of cell death using ascorbic acid was concentration dependent. Higher concentrations and differentiated cells need to be analyzed in order to confirm significance of results.

## **KETOGENIC DIET IMPACT ON SOCIAL & REPETITIVE BEHAVIORS IN AN AUTISM MODEL OF MICE**

Mira Nakhle '20, Roxana Alvarez '20 Faculty Sponsors: Susan A. Masino, David N. Ruskin

In animal studies, it is important to have a quantifiable method of analyzing behavioral changes. Behavioral evaluation is vital when using animals to study human illnesses with both behavioral and biological components. Video scoring is a crucial way to evaluate behavioral abnormalities by allowing for isolation of desired variables. The scoring process consists of recording a rodent in a certain environment and later analyzing the behavior throughout a specific time frame. This form of analysis allows for thorough observation of subtle differences in behavior that might be missed when watching behaviors in real time. This form of testing is ideal for behavioral analysis as it is able to be completed without human interaction or proximity which could potentially influence test results. In this study, the data collection technique has allowed for the analysis of social and repetitive behaviors in an autism model, BTBR mice. Of particular interest is if the ketogenic diet (KD) can reverse or minimize the decreased social and increased repetitive behaviors typically associated with the condition. This is done by comparing the amount of time that a subject mouse spends with a stranger mouse versus the amount of time it spends on its own. Additional forms of measurement include comparing time spent with frontal contact on a corral as well as time spent grooming under various circumstances. It was expected that the ketogenic would improve the severity of the autistic behaviors, however none of the three methods of measurement used showed any significant differences between control diet and KD results. Potential explanations for this outcome could be a weakening of the phenotypic traits of the BTBR mice due to a large generational distance from original breeding animals, or irregularities in the conditions of the environment in which the mice live.

43.

42.

#### THE EFFECTS OF KETOGENIC DIET ON APOMORPHINE-INDUCED STEREOTYPED MOTOR BEHAVIOR IN MICE

Lillian Russo-Savage '18, Khaoula Ben Haj Frej '18 Faculty Sponsors: David N. Ruskin, Susan A. Masino

The ketogenic diet (KD) has been implicated as a method of reducing repetitive behaviors expressed by different models of autism in mice. Dopamine receptor agonists have also been used to induce repetitive behaviors in mice. In addition, it has been demonstrated that KD increases adenosine levels in the brain, which is known to have an antagonistic relationship with dopamine. The present study explores the possibility KD could decrease repetitive behaviors induced by the dopamine receptor agonist apomorphine in different mouse strains. Four different mouse strains (C57 wild-type, C57 adenosine receptor 1 knockout, BTBR, and EL) known to

express different levels of stereotyped autism motor behaviors were placed on either a control diet or KD for 3 weeks before behavior testing. After habituation to the test apparatus and 10 minutes of baseline filming, the mice were injected with apomorphine (3.0 mg/kg), and filmed for another hour. Roughly 120 mice have been filmed and scored so far. Scoring will continue and additional animals injected throughout the semester.

#### 44.

#### USING AUDIOVISUAL TRAINING FOR PATIENTS WITH HEARING LOSS: TRANSITIONING FROM CI SIMULATION ON COLLEGE STUDENTS TO CLINICAL PARTICIPANTS

Rachel Scheub '20, Anneliese Pedro '20 Faculty Sponsor: Elizabeth Casserly

Patients that are profoundly deaf can use electronic aids to partially restore hearing, but in order to adapt to these aids, training programs are needed to teach patients how to hear with their devices. The overall research goal is to develop a more interesting and effective training program to fit that need. An audiovisual program using clips of TV interviews was first tested on college students by simulating a cochlear implant, and results from the first experiment demonstrated that this interview-based program was effective enough to proceed to a clinical experiment. To ethically apply the two hour long auditory training program to clinical patients above the age of 54, the program has to be genuinely interesting to the patients, and consisting of follow-up episodes to last the period of training. Secondly, a few clinical patients are needed to participate in the current auditory training program that was tested on college students to gauge how well clinical patients respond to those specific video clips used. Therefore, to prepare for a clinical trial, we created and administered a questionnaire that asked participants above the age of 55 open-ended and 'Yes or No' questions about their preferences for specific television programs and channels. The results showed that the target age group enjoys watching comical television programs such as Ellen, and news programs. This data can be used to meet the research goals- ensure that the extended training program (currently in development) is interesting, and will constitute multiple sessions over a period of time. We also began testing the existing program's application to clinical participants with hearing loss. Future research is needed to expand on the clinical pilots and transform it into a valid clinical study.

### **PHYSICS**

#### 45.

**COINCIDE – DIGITAL DATA ACQUISITION OF MULTI-PARTICLE EVENTS** Stephen J. DeMonico'17, Aashwin Basnet'19, Alex Bellas'20 Faculty Sponsor: David Branning

In nuclear, particle, and optical physics experiments, pairs of particles are often detected simultaneously or "in coincidence." For instance, this technique is particularly important in the study of photons that exhibit quantum entanglement. There are many existing solutions to this problem, but their usability and functionality correlate directly with cost, leaving most devices outside the means of small educational labs, such as one might find here at Trinity. However, the recent explosion of inexpensive and powerful DIY electronics enables the development of an all-digital coincidence counter with a low price-point and a full-featured stand-alone interface. The

proposed device will offer a touch-screen interface for quick and convenient alterations in the lab as well as a LABVIEW-based program designed for more advanced data analysis.

#### 46.

## ROOTED BINARY TREE MODEL FOR MECHANICAL PROPERTIES ANALYSIS OF ACYCLIC VASCULAR NETWORKS

Christopher Giottonini '19

Non-Trinity Faculty Sponsor: Peter Yunker, Jonathan Michel, Georgia Tech

Research done at Georgia Tech by Shane Jacobeen in collaboration with Dr. Will Ratcliff and others in the Yunker Lab has led to a more thorough understanding of organism evolution to multicellularity through a biophysical lens. A genetic mutation of the *ACE2* gene in baker's yeast *Saccharomyces cerevisiae* impedes separation of cells during mitosis and leads to the development of branching networks in snowflake yeast clusters. These clusters exhibit mechanical properties uncharacteristic of normal bulk materials which motivate our efforts to reveal the fundamental physics underlying networks similar to those found in snowflake yeast. We have developed a versatile program to generate binary rooted tree networks to serve as a strong groundwork for continued research into the mechanical properties of acyclic vascular networks. Furthermore, we present the *Spatially Disjoint Binary Tree Condition*, a novel approach to bound our network construction program and guarantee non-overlapping networks. Research into demystifying the mechanical properties analysis utilizing the Finite Element Method.

## **PSYCHOLOGY**

#### 47.

#### THE INFLUENCE OF EXOGENOUS CUES ON VISUAL ATTENTION

Carty Campbell '18 Faculty Sponsor: Michael A. Grubb

Research was conducted to examine the cost and benefits of exogenous visual attention, a type of attention considered to be automatic or reflexive, driven by an external stimulus. An example of exogenous attention would be if you saw lighting in the sky, and you noticed it without looking at it. In this example, the lighting would be considered an exogenous cue. The central focus of the study was to examine the effects of two baseline cues, a distributed cue vs. a central cue, used in many attention studies. An example of a distributed cue is when 2 flashes go off, left and right on a screen before a stimulus appeared. It would be considered a central cue if the flash appeared in the center of the screen before a stimulus appeared. Both examples are considered to neutral cues because they do not favor either the left or right side or indicate where the stimulus will appear. In order to determine which cue serves as a more naturalistic baseline/neutral cue, the accuracy and reaction time were compared for all participants in a two-alternative forced-choice visual task (n=22). It was hypothesized, in line with much previous research, that there would be significant difference in accuracy and reaction time, when comparing invalid vs valid cues, and this would verify that our procedure was capable of producing a significant cuing

effect. The results for invalid vs valid accuracy and reaction time had a p-value less than .05, indicating that participants answered more accurately and faster in the presence of valid cues vs invalid cues. For the experiment, all participants (n=22) were ask to sit at a computer screen, and to record their answers using a keypad. In the beginning of the experiment, participants were asked to stare at a fixation point on the screen and this stage of the experiment was considered to be the fixation period. The fixation period lasted for 1000 milliseconds, and was followed by another period, where the participant would be expose to either a valid, distributed, neutral, or invalid cue for 33 milliseconds. Participants would then experience a 67 milliseconds interstimulus interval, followed by interval (17 milliseconds), where participants would be exposed to two gradients, left and right of the fixation point. The gradients would be either rotated counter clockwise or clockwise. Shortly after, a response cue appeared either left or right, indicating which gradient orientation the participant had to record. The last interval was a feedback loop notifying the participants whether they recorded the orientation correctly. There were a total of 10 block with 96 trials within each. A 2 tailed paired t-test was used to test the differences between the two averages of accuracy, distributed vs central. P-value was less than .05, indicating that there was a significant difference between the two cues. Participants were able to respond more accurately in the presence of a distributed cue but not necessarily faster. Previous visual attention research has used both kinds of cues as baseline conditions to estimates of costs and benefits of visual attention, without considering that they may differ. Future projects will work toward assessing which cue is a better reflection of a neutral attentional state.

#### 48.

#### DOES HOMETOWN OF STUDENTS MATTER AT INTER-DISTRICT MAGNET SCHOOLS? DIFFERENCES IN STUDENTS' METACOGNITION, MOTIVATION, AND ACADEMIC PERFORMANCE

Daisuke Katsumata IDP; Phuong (Kelly) Nguyen '19

Faculty Sponsors: Dina Anselmi, David Reuman, Debra Avery, Andrea Heller, Timothy Roarty, and Chris Ewing

Inter-district magnet schools face challenges that are unique to serving a student population from diverse backgrounds that are not typical in more traditional school districts. Our study looked at kev differences in socio-economic status (SES), metacognition, motivation, and grades among select 6<sup>th</sup> through 8<sup>th</sup> grade students at one inter-district magnet school that serves the greater Hartford region in Connecticut. Metacognition and motivation were assessed using a variety of student self-report measures, as well as teacher ratings. SES indicators were measured in aggregate using the District Reference Group (DRG) classification system. Our results indicated a consistent trend in which students from more affluent districts had higher metacognition and motivation across a variety of measures, as well as better grades. Identifying such effects of hometowns is especially important at an inter-district magnet school because its mission revolves around addressing racial and socio-economic isolation of the districts it serves. District and statelevel assessment initiatives focus on students' academic performance, but not directly on students' metacognition and motivation. Our study provides teachers and school administrators with new information about students that may be helpful in designing and implementing effective pedagogies for a demographically heterogeneous student population. It is important to note that our findings compared students according to the districts they came from without accounting for individual differences within those districts. There are also notable limitations with respect to potential biases related to self-report measures, and our sample only included classes from select teachers from a single school.

#### 49. MIRROR MIRROR ON THE WALL: WHAT WE KNOW ABOUT MIRROR BEHAVIOR

Brii Kuzmickaite '18 Faculty Sponsor: Elizabeth Casserly

Growing up in the West means growing up in an environment that has mirrors everywhere. Despite this, scientific evidence regarding how they affect behavior is sparse. This project aimed to find out what is known about mirrors' influence on behavior and also to observe the differences directly, in various circumstances.

Only a few animals (chimps, orangutans, etc.) have been observed to recognize themselves, using the mirror-mark test. In human children, mirror recognition seems to start around the age of 18 months but can vary. Those two areas – self-recognition in human development and across species – are the most researched, whilst other areas are inconclusive. We summarize findings regarding task effort, mirror use in disorders such as body dysmorphia, and reports of negative self-image post mirror-viewing.

Direct observations, especially in areas where mirrors are expected (e.g. commercial areas) were also conducted. In such areas, people seemed to avoid looking at the mirror unless they had to. When a mirror was unexpected, it seemed almost everyone looked at the mirror.

Finally, in a pilot study designed to document mirror-related behavior in the laboratory, participants were asked to memorize a monologue near a mirror while their behavior was recorded. We found that people avoided looking at the mirror directly, although most faced it without looking, and that accuracy varied mostly as a function of time taken to memorize.

It seems that we use our schema of an environment in order to subconsciously decide whether to avoid looking at a mirror or to directly look at it, and it does seem to play some part in changing behavior due to this. For future research, situations in which a mirror is ignored versus specifically looked at could be studied, and how this changes behavior.

#### 50.

## INTER-TRIAL EFFECTS MODULATE RESPONSE TIME VARIABILITY WHEN MONETARY REWARD IS ON THE LINE

Benjamin M. Liske '20, Kalsang W. Sherpa '20 Faculty Sponsor: Michael A. Grubb

It is commonly observed that physically salient stimuli capture attention. However, even nonsalient stimuli capture attention if associated with monetary reward. In order to study the latter phenomenon, known as value-driven attentional capture, one particular protocol has been used in our lab. Participants first complete a training phase composed of several iterations of a visual task, where the goal is to report the orientation of the line located inside the red or green circle. The experimental group receives a trial-by-trial reward amount, based on their performance of the task; meanwhile, the control group receives a flat rate reward. Then, those participants complete a test phase, where the target line is located inside of the unique shape. Using this method, it has previously been shown that the stability of the strategic balance of speed and accuracy decreases when the subject undergoes a performance-contingent reward learning phase. In our study, we asked if the temporal presence of value-related distractors might affect performance despite their spatial absence. In order to answer this question, we removed red or green shapes from all trials in the test phase. We found that the variations in response time were similar between our control and experimental groups, unlike those in the aforementioned experiment. However, we duplicated the previous result that response time variation and error rate are positively correlated. These results suggest that the effects of performance-contingent reward learning are lost when distractors are entirely removed from the scenario, even temporally. In addition, it can be confirmed that response strategy stability and performance are linked. These findings further develop a gateway to study attention and the effects of exogenous distraction.