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TENTH ANNUAL SUMMER RESEARCH SYMPOSIUM TRINITY COLLEGE

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BIOLOGY

1.

DEVELOPMENT OF GENE CONSTRUCTS TO TEST THE FUNCTION OF THE NOTCH INHIBITORY REGION WITHIN THE SERRATE LIGAND

James Curlin '15

Faculty Sponsor: Robert J. Fleming

The Notch signaling system is an important cell-signaling pathway responsible for key developmental changes that take place in a wide variety of organisms. In *Drosophila melanogaster*, the Notch pathway is controlled by two different ligands, known as Delta and Serrate. Serrate is composed of an extracellular domain that consists of 14 EGF like repeats (ELRs), each of which has a unique amino acid sequence. Serrate activates Notch when located on adjacent cells in a process known as *trans*-activation, but inhibits Notch when located within the same cellular membrane, known as *cis*-inhibition. Inhibition of the Notch pathway appears to be controlled largely by ELRs 4, 5 and 6. Removal of any or all of these three repeats results in a significant loss of inhibition, with no effect on Notch activation. Furthermore, when these three repeats are relocated to a C-terminal position after repeat 11 of Serrate, the resulting form activates but fails to effectively *cis*-inhibit Notch. This suggests that ELRs 4-6 do not function as an independent unit from the N-terminus of the ligand that is necessary for Notch activation. The dependency of ELR's 4-6 on the N-terminal regions may require specific positioning of this region with respect to the N-terminus for function. We are creating a construct that moves ELRs 4-6 one ELR toward the 3' end of Serrate from its normal position to determine how critical the positioning of this region is for *cis* inhibitory functions.

2.

VIEWING WILDLIFE IMAGES THROUGH CITIZEN SCIENCE: IMPACT ON THE ANXIETY LEVELS OF MILITARY VETERANS WITH VARYING LEVELS OF PTSD

Nicole Evancho '15

Faculty Sponsors: Scott Smedley, Kathyne Marinchak, Trinity College Counseling Center

Citizen science is becoming a popular way to involve the public in scientific research that is too cumbersome for a single lab to undertake. In our study of the effects of residential composting on scavenger ecology, citizen scientists identify animals in numerous wildlife camera images through crowd sourcing. So far participants have included students (secondary and college) and high school science teachers, but we are eager to involve military veterans since they possess advanced observational skills and also have the potential to benefit educationally and psychologically from involvement. However, as a precaution before engaging veterans, we are determining whether or not those with post-traumatic stress disorder (PTSD) experience adverse reactions when viewing infrared illuminated images (similar to military night vision optics) or images of animals that could be considered menacing. In an ongoing pilot study, veterans with varying levels of PTSD (as measured by the PCL-M) are assessed for state anxiety levels (as measured by the STAI-S) immediately before and after viewing a set of twenty wildlife images from our study. Our pilot project results to date show no adverse response to viewing the wildlife images; in fact, the veterans overall illustrate a marginally significant ($P=0.065$)

decrease in anxiety after viewing. Participants with higher levels of PTSD, show the greatest anxiety reduction ($P=0.047$). In a follow-up study, a psychophysiological measure of anxiety and a non-veteran control group will be added to our current design.

3.

DETERMINING THE ROLE OF CELL DIVISION IN THE EMBRYONIC DEVELOPMENT OF *TRIBOLIUM CASTANEUM*

James Graydon '15

Faculty Sponsor: Terri Williams

The process of segmentation is vital to the developmental strategies of three major phyla, Arthropoda, Annelida, and Chordata, and contrary to traditional thinking, may share many commonalities between the species that are segmented. Despite the prevalence of segmentation, the cellular behavior that gives rise to highly ordered segments is not well understood. In order to understand what cell behaviors drive segmentation, segment addition, and elongation, embryos of *Tribolium castaneum* were injected with hydroxyurea, a known inhibitor of cellular mitosis. Embryos were injected during either one of two phases: an early phase characterized by high rates of mitosis and low rates of segment addition, and a late phase characterized by high rates of segment addition. The two distinct phases were chosen in order to determine the importance of cellular division during periods of high rates or low rates of mitosis. Hydroxyurea was found to inhibit cellular mitosis, and despite the vastly reduced rates of mitosis, elongation and segment addition both occurred, suggesting that cellular division is not critical in driving elongation and segment addition. Further research is needed to verify that the observed results were not due to off-target effects of hydroxyurea.

4.

INVESTIGATING NOTCH SIGNALING AND SEQUENTIAL SEGMENTATION IN THE FAIRY SHRIMP, *THAMNOCEPHALUS PLATYURUS*

Sara Khalil '15

Faculty Sponsor: Terri Williams

Segmentation is a key feature of arthropod diversity and evolution. The process by which segments form has been well studied in the model system *Drosophila melanogaster*. In *D.melanogaster*, segments develop simultaneously, by a progressive subdivision of the embryo (Peel. 2004). By contrast, most arthropods add segments sequentially from a proliferative posterior region called the growth zone. The underlying mechanisms that establish and maintain the growth zone and regulate elongation and segmentation are not well understood in most arthropods. It is also unclear whether common genetic and regulatory mechanisms exist among all segmented animals. In vertebrates, Notch synchronizes oscillations of gene expression and plays a role in posterior segmentation. It is part of a "clock" mechanism that controls segmentation and establishes somite borders.

Previous experiments in *T.platyurus* used pharmacological reagents to show that Notch signaling plays a role in segmentation. However, these experiments did not document the expression of

Notch genes through *in situ* hybridization or systematically investigate function using RNAi. In order to find relationships between gene regulation and segmentation in these animals, we will be tracing expression of Notch pathways genes using *in situ* hybridization and investigating their functions through RNAi knockdown experiments. Here we report on cloning potential homologs of two important Notch ligands, Delta and Serrate, from *T.platyurus* and initial experiments to visualize expression by *in situ* hybridization.

5.

CHEMICAL COMPOSITION OF CORN SNAKE EGGSHELLS AND ULTRASTRUCTURE OF CORN SNAKE AND KING SNAKE YOLK

Glory Kim '17, Cionna Rosenthal '17, Kathryn Powers '17

Faculty Sponsor: Daniel G. Blackburn

Corn snakes are oviparous and lecithotrophic species; they lay eggs and use a yolk sac to provide nutrients to their developing offspring. Corn snakes serve as valuable models for studies of the development of squamate embryos. In our preliminary studies, we have utilized an analytical technique of the Scanning Electron Microscope (SEM), energy-dispersive X-ray spectroscopy (EDS), to measure the relative percentages of calcium and other elements present in corn snake eggshells harvested during various stages of embryonic development. Our goal in gathering these datum was to track how and when calcium is mobilized from the outer surface of the eggshell to be taken up by the embryo. In addition to gathering calcium level datum, we also investigated yolk through photos taken with the SEM. By mid to late development, the yolk sac is entirely cellularized and vascularized in both corn snakes and king snakes, indicating a significant, unique evolutionary characteristic. These eggshell and yolk studies contribute to an understanding of the evolutionary history of reptiles as they help reveal the specializations in viviparous species and ancestral patterns from which viviparity has evolved.

6.

SELECTION OF LUMEFANTRINE RESISTANT *PLASMODIUM FALCIPARUM* PARASITES OF VARIOUS GENETIC BACKGROUNDS

Lulama L. Lunika '16, Nahla B. Gadalla, Thomas E. Wellems

Faculty Sponsors: Chris Swart, Nahla B. Gadalla, Ph.D.

Lumefantrine is a synthetic antimalarial drug used in combination with artemether to treat malaria. The genetic determinant of *Plasmodium falciparum* lumefantrine resistance has not yet been identified. Using three parasite lines from various genetic backgrounds: 803 (South East Asia), GB4 (Africa) and 76H10 (a progeny of 803 and GB4); our study has selected parasite lines that are resistant to lumefantrine *in-vitro*. Our aim is to identify the genetic loci associated with resistance employing advance comparative genomics. After a six-month period of selection, no changes have occurred in the SNP's of candidate genes, *pfmdr1* and *pfcr1*, associated with antimalarial susceptibility.

7.

EFFECT OF PREDATION ON CELL PROLIFERATION IN THE FOREBRAIN OF ELECTRIC FISH, *BRACHYHYPOPOMUS*

Michael Ragazzi '16

Faculty Sponsor: Kent Dunlap

Adult neurogenesis, the formation of new brain cells, occurs in specific regions of the adult brain. Electric fish, in particular, generate new neurons at very high rates, and the rate of proliferation of new cells is influenced by a variety of factors, including social and psychological stimulation. We studied the effect of predation on neurogenesis from natural populations of *Brachyhypopomus*, sampling fish from six different populations throughout Panama that vary in predation levels. We identified cell proliferation in the adult fish forebrain through immunohistochemical localization of Proliferating Cell Nuclear Antigen (PCNA). Fish with injured tails exhibited less cell proliferation than fish with intact tails. Moreover, we found a negative linear correlation between the percentage of fish injured and the amount of cell proliferation in the brain, suggesting that higher levels of predation decrease cell formation in the brain. The high predation sites contrasted with the low predation sites by showing lower levels of neurogenesis. Finally, no correlation was found between neurogenesis and body length or the sex of the animal. Our current results suggest that predation inhibits the proliferation of brain cells in the forebrain of electric fish. This could occur because the fish in high predation sites experience stress in their environment, which restricts rapid cell division in the brain.

8.

A *XHO* I CONTAINING FORM OF SERRATE HAS FUNCTION IN NOTCH SIGNALING COMPARABLE TO WILD TYPE

Kathy Rodogiannis '17

Faculty Sponsor: Robert J. Fleming

Notch signaling is an evolutionarily conserved developmental pathway that regulates several cellular processes in a wide range of organisms. In *Drosophila melanogaster*, the Notch ligands, Delta and Serrate (Ser) mediate cell-to-cell communication through (1) trans-activation when the Notch receptor and ligand are present on adjacent cells and (2) cis-inhibition when the Notch receptor and ligand are located on the same cell. In this study, an artificial *Xho* I site was built into the coding DNA of the intracellular region of Ser to facilitate the building of altered Ser constructs in the future. Expression of the *Xho* I containing form of Ser via the patched and Ser promoters was used to determine the functionality of the construct. This *Xho* Ser construct was found to trans-activate and cis-inhibit the Notch pathway similarly to wild-type Ser. This construct will be used in the future to replace the transmembrane and membrane adjacent regions of Ser in order to investigate the importance of an extracellular metalloprotease cleavage site in Ser for trans-activation and cis-inhibition properties of this Notch ligand.

9.

IS THE RED ALGA *LOPHOCLADIA TRICHOCLADOS* (CERAMIALES, RHODOMELACEAE) BOTH CRYPTIC AND MORPHOLOGICALLY PLASTIC IN THE WESTERN ATLANTIC?

Dylan S. Spagnuolo '17

Faculty Sponsor: Craig W. Schneider

In the western Atlantic Ocean, the red algal genus *Lophocladia* is currently known as a single species, the generitype *L. trichoclados* with a type locality in the West Indies. Using molecular-assisted alpha taxonomy (MAAT), this study evaluates the possible presence of two cryptic species now designated as *L. trichoclados* in the Florida Keys, which have been distinguished using conserved mitochondrial gene sequences (COI-5P) as the barcode marker for species diversity. Specimens collected in Bermuda and Key West, while appearing to be morphologically distinct, have been found to be genetically the same species (morphological plasticity). Morphological measurements have shown the Bermuda collections to differ from those in Key West primarily in the dimensions of the plants' periaxial cells while the collections from Key West have not revealed any distinct differences in their morphological characteristics from one another (crypsis).

10.

REFERENCES AMONG BEETLE FAMILIES INHABITING COMPOST PILES WITH VARIED KITCHEN SCRAP CONTENTS

Bridget Tevnan '15

Faculty Sponsor: Scott Smedley, Dr. William Krinsky, Division of Entomology, Yale Peabody Museum

Composting is a common, environmentally sustainable method of organic waste disposal; however no experimental data exist to examine the claim that adding animal-based products to compost piles increases wildlife visitation. Seasonal replicates in which wildlife monitoring cameras record mammalian and avian visitation to three compost piles with varying kitchen scrap content (vegetable products only – VEG, vegetable and animal product mix – MIX, and control with no scraps – CON) have taken place in Andover, Connecticut since February 2008. Because compost piles also offer resources and habitat to invertebrates, an invertebrate survey was conducted during the summer of 2009. Every ten days, samples were collected from the straw and leaf mulch substrates of the three piles. Tullgren funnels were used to extract and preserve the invertebrates. The beetles were then identified to at least the family level. Thirteen beetle families were encountered, four of which had sufficient sample sizes ($n \geq 15$ /substrate type) for statistical analysis. The Hydrophilidae, Nitidulidae, Ptiliidae, and Staphylinidae were analyzed using chi-square (goodness-of-fit) tests to determine their frequency among pile substrates and treatments. In both the mulch and straw substrates, Hydrophilidae and Staphylinidae showed a significant preference for the MIX pile. In the mulch substrates, Nitidulidae showed a significant preference for the VEG and MIX piles, while Ptiliidae showed no pile type preference. Amongst the straw substrates, Nitidulidae showed a significant preference for the VEG pile, while Ptiliidae preferred the CON and MIX piles. Beetles may

preferentially inhabit substrates and pile types due to scavenging habits and predation-prey relations. Pile location and microclimate may also influence their occurrence.

CHEMISTRY

11.

ELIMINATING THE CARRYOVER EFFECT OF DIHALOMETHANES FROM SPME FIBERS

Ashira Anderson '16, Christina McGuire '13, Edward Harrington '11
Faculty Sponsor: Maria J. Krisch

This research project deals with an improved method to quantify dihalomethanes using Gas Chromatography-Mass Spectroscopy (GC-MS). Solid phase microextraction (SPME) was used to prepare samples that were run through the GC-MS. At lower concentrations, residual chemicals stuck to the SPME fiber, thus displaying a carryover effect. In order to improve the removal of residual chlorodiodomethane (CH_2I_2), a conditioning run between samples was introduced by adding a second injector port. The work here details experiments that were run to see how temperature affected the measurements. The results of the varying temperature runs gave very similar slopes, indicating that the better results were the effect of the cleaning run.

12.

ALKYNYL β -SHEET PEPTIDOMIMETICS RETAIN THEIR ANTI-PARALLEL SHEET CONFORMATION WHEN COORDINATED TO TUNGSTEN

Adam N. Boynton '12, Shawna M. Berk '13, Elena-Marie C. Pedro '17
Faculty Sponsor: Timothy P. Curran

β -sheet proteins and their aggregation in cells are areas of study due to apparent associations with the incidence of such neurodegenerative diseases as Alzheimer's disease. Exploring factors that stabilize or destabilize these proteins may provide information on these diseases. The research objective was to investigate whether peptide derivatives of 2-amino-2'-carboxydiphenylacetylene maintained their β -sheet conformation when the alkyne bond in the diphenylacetylene was coordinated to the transition metal tungsten. Research conducted yielded results that showed that the peptide portions of the synthesized mono-alkyne complexes maintained their β -sheet conformation. The structure and purity of these compounds were then verified by re-synthesis, followed by flash chromatography and high performance liquid chromatography to confirm the purity and mass spectrometry and ^1H NMR spectroscopy to prove its characterization. Details regarding this work will be presented.

Because protein aggregation in diseases such as Alzheimer's disease is manifested in the body's cells, examining what makes these proteins stable and unstable in an aqueous solution would allow investigation of the behavior of β -sheet proteins in an environment similar to the one in which it causes health concerns. Therefore, research is currently being investigated to displace a CO ligand from the monoalkyne complexes previously studied, and to replace it with a ligand (for example an alkynylcarbohydrate, propargylglucose) that would make the anti-parallel β -

sheet soluble in aqueous solutions. Details about the synthesis, purification and coordination of propargylglucose to tungsten will be presented.

13.

CONSTRUCTING A SYSTEM FOR MICROCHIP ELECTROPHORESIS LASER-INDUCED FLUORESCENCE (LIF)

Eleanor Clerc '17

Faculty Sponsor: Michelle L. Kovarik

Microchip electrophoresis laser-induced fluorescence (LIF) is an effective method for the analysis of single cells. The microchip allows for precise fluid control, as well as a confined space in which to analyze small volume samples such as lysed single cells. These microchips are fabricated in the lab using a soft lithography technique. Once the sample has been loaded into the microchip, electrophoresis is an efficient method to separate the contents of the cell. Electrophoresis uses voltages applied to the chip to separate the contents of the cell based on their charge and size. Since the sample volume is limited, a sensitive detection method, such as LIF is required to analyze the sample. A program in LabView was created in order to control the voltages used for electrophoresis, as well as record the output of the detection system to a text file for analysis. The laser used in LIF was aligned and a power source was built for the photomultiplier tube (PMT), which was attached to the microscope for detection. After the system was fully constructed, a limit of detection was determined using fluorescein injections and optimal laser power and microscope objective were found. The LOD was 10^{-19} moles, the optimal laser power was 2mW and the optimal microscope objective was 60x magnification. Images taken during these injections were analyzed in ImageJ and data from the PMT was analyzed in Cutter 7. After the limit of detection was determined, peptide separations were conducted. The next step in this project is to optimize a buffer for the separation of reporter peptide fragments. In the future, this newly-built system will be used to conduct enzymatic assays of lysed single cells containing a fluorescent reporter peptide.

14.

SYNTHESIS OF A PHOSPHONAMIDITE FOR FUTURE SOLID-PHASE OLIGONUCLEOTIDE SYNTHESIS

Florence Dou '16

Faculty Sponsor: Richard Prigodich

Organic and inorganic cations bind to the anionic phosphate groups of nucleotides, oligonucleotides and nucleic acids. In fact, an increase in metal cation concentration can induce the right-handed B-DNA helix to change conformation into a left-handed Z-DNA helix. This cation-DNA interaction can be studied by monitoring the ^{31}P NMR chemical shifts. However more detailed information can be obtained if at a single site on the oligonucleotide, the 3'-O of the ribose ring is changed to a methylene group. This will produce a distinct, unique signal in the ^{31}P NMR spectrum that can be used to monitor cation binding at that site in the oligonucleotide. In order to make this change, a phosphonamidite will be synthesized by a seven-step reaction sequence. The phosphonamidite can then be used in standard oligonucleotide solid-phase

synthesis. In the first step, the 5'-hydroxyl group of thymidine was protected with tert-butyldiphenylsilyl chloride. In the second step, the 3'-hydroxyl group was substituted by an iodine atom. The third step will be a Grignard reaction to displace the iodine atom with a vinyl group.

15.

ONE STEP DEUTERIUM EXCHANGE ON TERNARY METAL HYDRIDES

Ryan Geelan '15

Faculty Sponsor: Ralph Moyer

One-step deuterium exchange chemistry was attempted on the ternary metal hydrides Sr_2IrH_5 , Sr_2RuH_6 , Eu_2IrH_5 , and Eu_2RuH_6 through heating the compounds to a predetermined temperature under approximately 1 atm of deuterium gas. Comparison of Infrared spectra of the sample before and after the treatment revealed a shift in the wavelength consistent with the d to h reduced mass of 1.40, indicating that a successful exchange had occurred. Additionally, powder x-ray diffraction patterns revealed that the crystal structures remained intact throughout the exchange process. The one step reverse exchange process was also demonstrated to work under the same conditions for Sr_2RuH_6 , Eu_2IrH_5 , and Eu_2RuH_6 in a hydrogen gas environment. The iridium containing samples underwent the exchange under milder conditions than ruthenium containing samples. We hypothesized this difference may be related to the structural differences between the two types of compounds.

16.

THE NEW ROUTE FOR THE SYNTHESIS OF TRIARYLMETHYL CATIONS

Mazin Khalil '15

Faculty Sponsor: Cheyenne Brindle

As a result of the difficulties with the traditional carboxylic acid to acyl chloride to triarylmethyl cation route, it was decided that an alternative method was required. We decided to change the starting material and in turn began using compounds such as 1-bromo,2,6-difluorobenzene, which would change the electronic aspect of the experiment since fluorine is so electronegative and mimicked the size of a hydrogen ion such as in the original 3-fluoro-2-bromotoluene. We decided to incorporate new electrophiles to try and tweak the reactivity. Our results in the end were fairly inconclusive.

17.

IMPLICATIONS OF INCREASED REPEATS IN THE HEXANUCLEOTIDE EXPANSION OF CHROMOSOME 9 ON AMYOTROPHIC LATERAL SCLEROSIS (ALS) PATHOPHYSIOLOGY

Heather S. Loring '15

Faculty Sponsor: Terence R. Flotte, M.D.

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disorder caused by the degradation of motor neurons. A subset of familial ALS has been linked to a mutation in the C9ORF72 gene. An intron within the gene contains a hexanucleotide repeat "GGGGCC" that occurs 23 times in healthy individuals. In individuals afflicted with ALS, the hexanucleotide repeat occurs hundreds to thousands of times. The cause of ALS associated motor neuron degradation is thought to be increased toxicity within nerve cells due to the buildup of longer RNA transcripts, which sequester proteins. A plasmid was packaged with 200 repeats and injected via the intracerebral ventricles of wild-type mice. These mice were then injected with a "GGGGCC" probe and analyzed by fluorescent *in situ* hybridization (FISH) to detect the presence of RNA transcripts containing the "CCCCGG" repeat. RNA transcript build up was evident in the cortex and spinal cords of mice injected with the plasmid containing the expanded repeat but not in normal mice. Based on this data, the cause of the progressive motor neuron deterioration, evident in the expanded repeat injected mice, is the toxicity associated with the buildup of RNA transcripts and their sequestration of proteins.

18.

SYNTHESIS OF TURBOMYCIN ANALOGUES FOR THE DEVELOPMENT OF NEW ANTIBIOTICS: VARIATION OF THE PHENYL COMPONENT

Brooke Moore '15, Ifeanyi Okoh '15, Abigail Whalen '15, Christine Reavis '15,

Phong Quach '17

Faculty Sponsors: Cheyenne Brindle, Lisa-Anne Foster

Increasing bacterial resistance to commercial antibiotics is one of the leading health concerns in modern medicine. The natural product turbomycin has antibiotic activity that act on an array of bacterial strains. Through research experiment, several analogues of turbomycin were prepared and tested against a variety of bacteria, such as *Bacillus cereus*, *Streptococci pyogenes*, *Staphylococci epidermis*, *Enterococcus faecalis*, and *Escherichia coli*, to investigate what structural characters of the molecule impart antibiotic activity resulted in a more potent antibiotic. Compounds were prepared using a one-step procedure for the synthesis of the unoxidized turbomycin analogues. This allowed for the synthesis of four new analogues this summer. One of the previously made precursor analogues was oxidized to remove the hydrogen atom on the central carbon. Turbomycin B has a cationic central carbon, therefore the precursor was oxidized in order to determine if it has the same biological activity as turbomycin B. It was previously found that solutions of pure DMSO and drug kill many strains of bacteria that we wished to use in our testing. Therefore, minimizing the amount of DMSO used to dissolve the drug should alleviate this problem. We made decreasing percentages of DMSO in water by using serial dilutions. Initial biological assays indicate that the precursor to turbomycin is also an effective antibiotic. Future efforts will investigate whether this activity is due to the different substituents of the phenyl group or direct action of the precursor itself.

19.

LOADING PEPTIDES INTO *DICTYOSTELIUM DISCOIDEUM* USING MYRISTOYLATION AND PINOCYTOSIS

Berjana Nazarko '15

Faculty Sponsor: Michelle L Kovarik

The main goal of the research was to transport an exogenous peptide across the membrane of *Dictyostelium discoideum* for future assays of the activity of protein kinase B (PKB). PKB is a signaling enzyme involved in many pathways and is related to survival, cell proliferation and prevention of apoptosis. Since the phospholipid bilayer of the cells is permeable only to small hydrophobic molecules, larger reporters are not readily transported into cells. Therefore a method for loading larger molecules had to be optimized. Two methods were used for this purpose: myristoylation and pinocytosis. Myristoylation is a chemical modification of the peptide by covalently binding it to a lipophilic myristoyl group that helps the peptide insert itself into the membrane and flip into the cytoplasm. The cells were incubated with the peptide for 20 minutes at different concentrations: 0, 1, 5, 15, 20 μM . They were also incubated at a concentration of 10 μM for 1, 10 and 40 min. All these experiments gave 100% viability of the cells based on Trypan blue exclusion assay. Loading of the peptide was assessed by intensity of fluorescence from a fluorescein tag; the brightness of the cells is proportional to the amount loaded. Although loading was successful, the peptide appeared to be localizing in endosomes.

To determine whether endocytosis was causing this localization, cells were treated with caffeine, which the literature suggested inhibits endocytosis. The average intensity of cells treated with caffeine was lower than the intensity of untreated cells. Therefore endocytosis was responsible for some peptide loading. However inhibition of endocytosis by caffeine treatment did not eliminate the localization of the peptide in endosomal compartments. Pinocytosis is an alternative loading method that takes advantage of endocytosis, but uses osmotic pressure from hypotonic media to cause the pinosomes to burst and release the molecules being loaded. In experiments with pinocytosis, the cells were not very bright and seemed to be stressed but still viable. Future work includes steps to reduce localization during myristoylated peptide loading and to increase loading by pinocytosis, followed by the use of labeled substrate reporters to assay the activity of the PKB.

20.

EXPRESSION OF FIBROSIS-RELATED mRNAs AND miRNAs IN PEDIATRIC IDIOPATHIC DILATED CARDIOMYOPATHY (IDC)

Hieu "Hugh" Nguyen '17

Faculty Sponsors: Michelle L. Kovarik, Shelley Miyamoto¹, Brian Stauffer², Kika Sucharov²

¹Department of Pediatrics and Children's Hospital Colorado, Aurora, Colorado

²Division of Cardiology, University of Colorado Denver School of Medicine, Aurora, Colorado
Children's Hospital Colorado

Fibrosis is nearly universal in adults with idiopathic dilated cardiomyopathy (IDC) and contributes to the poor function of the heart. Fibrosis is much less commonly seen in children with IDC, which could help explain why some of the medicines effective in adults are not as

effective in children. However, no prior studies have investigated the myocardial expression of genes regulating fibrosis in children with IDC. The hypothesis of this study was that pediatric hearts with IDC will have higher fibrosis-related gene expression and lower expression of microRNAs (miRs) that are known to target fibrosis-related genes than normal pediatric hearts. Left ventricular (LV) tissue from IDC pediatric hearts (age<18; n=41) and non-failing controls (age<18; n=24) were subjected to Quantitative Real-Time Reverse Transcription PCR (RT-qPCR) in order to measure gene expression of: AT1R, ST2L, Galectin-3, Corin, gelatinases (MMP-2, -9), MMP inhibitors (TIMP-1 to -4), and the miR-29 family. Additionally, RT-qPCR was also done on adult LV samples (non-failing: n =10; IDC: n =10) to test for expression of the genes. While all of the 14 genes included in this study have been shown to be differentially regulated in adults with IDC relative to healthy controls, only MMP-2, TIMP-1, TIMP-4, and miR-29s were noted to be significantly different between pediatric IDC and NF. Our findings suggest that these genes may play some role in pediatric IDC, and may help explain the lower prevalence of fibrosis in children compared to adults with IDC.

21.

POPPING BOTTLES

Haley Peterson '15

Faculty Sponsor: Maria J. Krisch

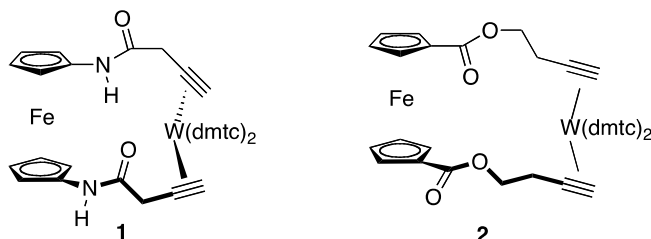
Carbon dioxide gives champagne its “bubbly” or effervescent effect. The effervescence of champagne gives the drinker visual, aromatic, and mouth sensations. For these reasons, it is ideal to try and optimize the amount of CO₂ present in champagne, and to maintain high CO₂ levels for as long as possible. The goal of this study was to examine whether a cover would make a significant difference in the CO₂ retention compared to an open glass. Over the course of various trials it was found that when the champagne was left covered for extended periods of time (25 minutes) significant amounts of additional CO₂ were retained versus when the champagne was left uncovered. When the cover was taken on and off repeatedly over the course of the 40-minute period (with the cover on for three minutes and off for a minute at a time), the amount of CO₂ was almost identical to when the champagne was left uncovered. The next step would be to determine exactly where the point is where it makes a difference to cover the champagne.

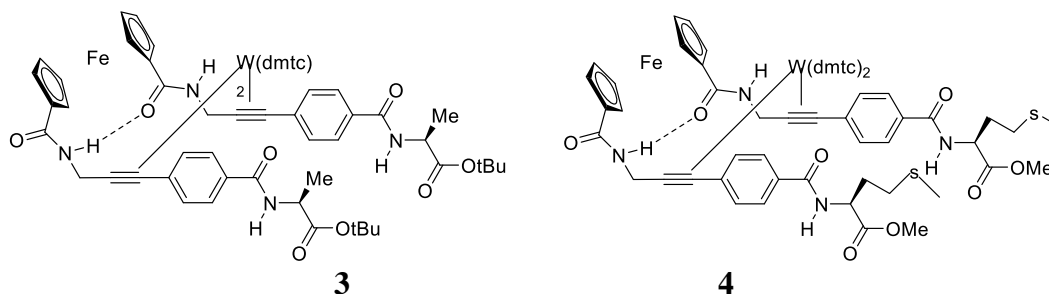
22.

EXPLORING WHETHER A NOVEL MACROCYCLE CONTAINING IRON AND TUNGSTEN CAN BE USED TO NUCLEATE A β -SHEET

Niranjana Pokharel '15

Faculty Sponsor: Timothy P. Curran





The β -sheet is one of the two secondary structures observed in proteins. β -sheets are stabilized by hydrogen-bond interactions between lateral chains of amino acids. Their shape and physical properties have been implicated in several diseases, including Alzheimer's disease. Our lab has been interested in finding organometallic models for β -sheets. It might be possible to use a molecule with rigid conformation to hold two peptide chains in close proximity so that a β -sheet structure is obtained. In prior work in our laboratory, dialkynylpeptides were complexed to tungsten, forming novel metallacyclicpeptides featuring a cyclic tungsten bis-alkyne complex. Most of these complexes synthesized were found to be flexible about the tungsten-alkyne bond. This posed the question of whether all cyclic tungsten bis-alkyne complexes would be flexible. In 2010, Lawrence synthesized complex **1**, a bimetallic ring system with a ferrocene unit at one end, and a tungsten bis-alkyne complex at the other end. In order to study the rigid behavior of complex **1**, complex **2** was synthesized. The flexibility of complex **2**, along with the results from Lawrence, Ji and Murtaugh, shows that the rigidity of the cyclic tungsten bisalkyne molecule depends on the ring size; complexes having the ring size found in **1** will be rigid. In order to explore whether peptides attached to **1** will adopt a β -sheet conformation, two metallacyclicpeptide complexes were synthesized and their rigidity was explored: complex **3**, which contains the amino acid alanine, and complex **4**, which contains the amino acid methionine. The ring system in **3** and **4** is identical to the ring system in **1**. Addition of the benzene ring and the amino acid did not change the rigid ring structure. The intramolecular hydrogen bonding in complex **3** and **4** is being studied using a DMSO titration experiment. The results from these studies will be presented along with the ideas for other possible derivatives of **2**.

23.

COMPARING MEASUREMENTS OF THE FORMIC ACID, WATER, AND SODIUM CHLORIDE LIQUID-VAPOR INTERFACE

Jeff Pruyne '15

Faculty Sponsors: Maria J. Krisch, Matthew A. Brown (Institute for Chemical and Bioengineering, ETH Zürich, CH-8093 Zurich, Switzerland), Ming-Tao Lee, Markus Ammann (Paul Scherrer Institute, CH-5232 Villigen PSI, Switzerland)

The liquid-vapor interface has experienced renewed interest from physical chemists in last two decades as techniques to study it have become more accessible and reliable. We compare two different methods of examining the liquid-vapor interface for a ternary (water, formic acid, and sodium chloride) system. An important property of the interface is the surface excess of a compound relative to its concentration in the bulk solution. Wilhelmy plate surface tension measurements from which the surface excess can be derived were compared with X-ray

photoelectron spectroscopy (XPS) measurements of the composition of a surface layer of the solution. We found sodium chloride acts as a weak salting out agent for the formic acid with a very small concentration dependence of surface excess on salt concentration. Our data are consistent with the surface tension measurements accessing a more surface selective region than XPS spectra. A simple model of the XPS experiment allowed us to replicate the XPS results under the assumption that the top nanometer of solution contributed most of the experimental signal.

24.

SYNTHESIS OF TURBOMYCIN ANALOGUES FOR THE DEVELOPMENT OF NEW ANTIBIOTICS: VARIATION OF THE INDOLE COMPONENT

Phong Kim Quach '17

Faculty Sponsors: Cheyenne Brindle, Lisa-Anne Foster

The world has entered an antibiotic crisis due to increased bacterial resistance. This necessitates the development of novel antibiotics. Turbomycin B is a naturally occurring compound that has been isolated from microorganisms found in soil and has shown antibiotic effects. Our research focuses on altering the structure of turbomycin B, specifically the indole portion to determine the consequences of such changes. Thus far, we have synthesized N-tosyl, N-acetyl derivatives precursors. Additionally, we are optimizing the synthesis protocol for synthesizing the 5-bromoindole and pyrrole derivatives. Moreover, we have made serial dilutions of the N-methyl derivative precursor in DMSO and water to test on *B. subtilis*, *E. coli*, *P. fluorescens*, *P. vulgaris*, *S. griseus*, *S. salivarius*, *S. viridans*. In the future, we plan to oxidize the precursor analogues, alter the indole portion with imidazole group and other substituents, and make dilutions of the analogues to test them on biological systems.

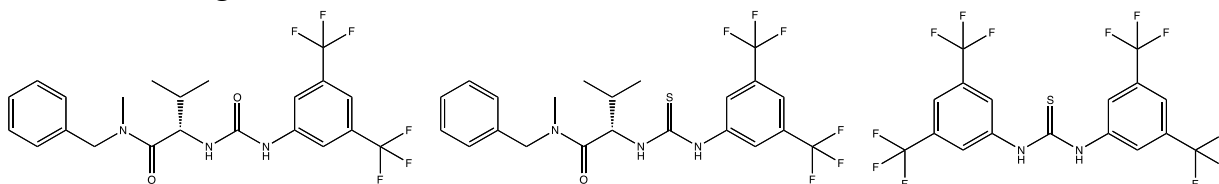
25.

THE IMPROVEMENT OF HYDROALKOXYLATION REACTIONS VIA UREA CATALYSTS

Jordan Reid '17, James Cescon '16

Faculty Sponsor: Cheyenne Brindle

Figure 1: Klausen's Urea, Klausen's Thiourea, and Schriener's Thiourea



Over the course of the ten-week period spent in Doctor Brindle's lab, the primary focus of our research was to develop a hydrogen-bond catalyzed hydromethoxylation reaction and see if the reaction could be improved in areas such as reaction speed and product formation via various catalysts.

The research was basically split into two sections over the ten-week span. The first section was dedicated strictly to hydroalkoxylation reactions, chemical reactions that combine alcohols with alkenes to produce ethers. The alkenes used were α -methylstyrene, 1-phenyl-1-propanol, and norbornene. MeOH and 2,6-dimethyl phenol as the alcohols while the solvents and acids varied from experiment to experiment. To conduct these hydromethoxylation reactions, the alkene, alcohol, solvent, and finally acid amounts were all calculated using mole equivalents, added to a vial around 2 drams, and stirred at a specific heat. Aliquots of around 200uL were taken at five different time intervals and analyzed for purity using Thin Layer Chromatography plates. Afterwards the aliquots were moved into vial specialized for Gas Chromatography Mass Spectroscopy, allowing us to follow the reaction at the five time points and analyze starting material, product, or product fragments resulting from either dehydration, normal synthesis, or dimerization. The second section of research was dedicated mainly to the formation of four hydrogen-bonding catalysts, Schriener's Urea, Schriener's Thiourea, Klausen's Urea, and Klausen's Thiourea. The catalyst were produced by first synthesizing 3,5-bis(triflouromethyl)phenyl isocyanate and isothiocyanate from 3,5 Bis(triflouromethyl)aniline and triphosgene and an amine HCl from the production of N-methyl-N-benzyl-NBOC-valine and the subsequent removal of its BOC group.

All four catalysts, not including Schriener's Urea, were successfully synthesized, and are subsequently being tested on our best hydromethoxylation reactions at this time, the first of which being one mole equivalent of α -methylstyrene with two mole equivalents of MeOH, a 0.01 equivalent of TsOH, and 0.1M Toluene.

26.

CHARACTERIZATION OF THE STABILITY OF SUPPORTED BILAYER MEMBRANES IN POLYDIMETHYLSILOXANE MICROFLUIDIC DEVICES

Livia Shehaj '15

Faculty Sponsor: Michelle L. Kovarik

Supported bilayer membranes (SBMs) have been used to coat capillaries and microfluidic devices. These coatings are similar to cell membranes and prevent adsorption of proteins, DNA, and other biomolecules to channel walls. Combined with the small dimensions of microfluidic devices, such coatings are an optimal tool for biological and biomedical research. To characterize the stability of SBMs, we prepared hybrid PDMS-glass devices containing straight, 3 cm channels. The channels were filled with small unilamellar vesicles, which self-assemble into supported bilayers. Conductivity measurements of the electroosmotic flow were used to characterize the presence and stability of the SBMs. Our primary focus was the stability of coatings using natural vs. synthetic lipids. No substantial difference was observed, suggesting that the variations in length and saturation of natural lipid tails do not affect the stability of the coatings. Electroosmosis measurements of the two lipid coatings were relatively stable (RSD 8-18%) for 2-4 hours when a constant voltage was applied across the channel. At longer time points, electroosmotic mobility increased to that of a bare chip suggesting the coating was destroyed. The stability of these membranes was also studied over a two week period to determine how long a device can be stored before use. Our results suggested that a chip can be stored for about a week and still yield consistent results. After a week, the coatings deteriorated,

as evidenced by increasing electroosmotic mobility. On-going work is exploring effects of cholesterol on SLBs to further enhance the stability and storability of these coatings.

27.

**APPLICATION OF ANALYTICAL TECHNIQUES IN ART CONSERVATION:
DART-TOF-MS & SEM-EDS**

Sarah Talcott '17

Faculty Sponsor: Henry DePhillips

A number of analytical techniques have been developed for the determination of resins, binders and pigments, both organic and inorganic, in artifacts, in particular, easel paintings. Typically, those methods require that the sample be modified (solubilized, derivatized) and given that samples taken from easel paintings are very small, treatment usually means loss of the original sample. Hence, any technique that permits direct analysis of sample components with no prior treatment is preferable. One such technology is Direct Analysis in Real Time, Time of Flight, Mass Spectrometry (DART-TOF-MS), especially for organic materials. The conditions required to obtain a Mass spectrum using DART-MS considerably reduce the amount of fragmentation produced. Inorganic components are determined by their elemental composition. Using Scanning Electron Microscopy with an Energy Dispersive Spectroscopy accessory (SEM-EDS) we have determined the elemental composition of a number of pigments to complement the data accumulated from prepared and commercial paint samples.

This poster shows the first steps in the development of a library of information on artificially heat aged resins, binders and pigments analyzed by DART-MS and SEM-EDS. In addition, results are shown for artists' paints prepared and aged in our lab as well as commercial artists' paints aged and analyzed in the same way. Finally, these methods permitted us to determine the resin, binder and pigment from a painting sample submitted by a private owner for materials analysis.

COMPUTER SCIENCE

28.

**A GPU BASED APPROACH TO THE 0-1 KNAPSACK PROBLEM USING THE
DISCRETE SHUFFLED FROG LEAPING ALGORITHM**

Pranav Bhandari '17, Rahul Chandrashekhar '17

Faculty Sponsor: Peter A. Yoon

The Knapsack Problem is a popular combinatorial optimization problem which is of the type NP-hard. It assumes a case where there is a knapsack which can hold a maximum weight W and there is a set of items N provided from which each item n_i has a certain weight w_i and a value v_i . The task is to pack the knapsack with the maximum possible value while staying under the weight limit of W . The 0-1 Knapsack Problem is a unique case of the classic Knapsack Problem in which each item from the set either goes in or out of the knapsack in entirety. Fractional portions of items are not allowed. The conventional method to tackle is problem is the brute

force method in which all the subsets of the set N which equals to 2^n are compared from which the one with the highest value under the weight limit W is considered the optimal solution. But as n increases, the number of subsets also increases exponentially making this conventional approach computationally impractical. We use the Discrete Shuffled Frog Leaping Algorithm to solve this problem which employs the use of dividing the problem into multiple sub-problems hence making it more suitable for parallel computation. The GPU based approach is theoretically a much more efficient alternative for a large scale implementation of the Knapsack Problem. This approach employs the use of multiple GPU threads which simultaneously work on the different sub-problems, hence making the computation much faster and efficient. Each iteration performs an improvement to the multiple subsets followed by a shuffle operation. As it is a meta-heuristic algorithm, it terminates when a desired amount of improvement has been undergone. The subsets combine to generate the final solution set.

29.

HYPERGRAPH PARTITIONING ON GPUS AND IMAGE CLASSIFICATION

Hyunsu Cho '15, Sam Johnson '17

Faculty Sponsor: Peter A. Yoon

A standard method of classifying images is pairwise comparison. We compare images one-to-one and record their similarities. Graphs provide a nice abstraction: Nodes represent images and edges similarities among them. There is an extensive body of literature describing how to partition the nodes into two homogeneous groups. Unfortunately, graphs prove inadequate for real-world datasets. We could gain better insight by comparing multiple images at once than comparing only two at a time. Hypergraphs are a promising alternative: each edge connects more than two nodes. Several researchers such as Yu (2012) obtained better classification results using hypergraphs. Our contribution is to accelerate hypergraph partitioning by using a commodity hardware known as Graphics Processing Units (GPUs). We present some preliminary results.

30.

MULTI-GPU BISECTION ALGORITHM FOR TRIDIAGONAL SYMMETRIC EIGENVALUE PROBLEM

Barok Imana '16, Nicky Thai '15

Faculty Sponsor: Peter A. Yoon

The goal of the research was implementing bisection algorithm on a multiple-GPU system for the computation of eigenvalues of symmetrical tri-diagonal matrices. The eigenvalue computation is one of the important problems in numerical linear algebra. It has applications in Mathematics, Physics, Engineering, Epidemiology and many other fields. The focus of the research was finding the eigenvalues of matrices in the symmetric tri-diagonal class. The symmetric tri-diagonal class represents matrices with entries only on the main diagonal and on the diagonals above and below the main diagonal. In this research, it has been attempted to compute the eigenvalues of such matrices in an efficient way by first creating a serial program. As the research progressed, programs that would perform the computation on a multi-core CPU

and on a single GPU has been successfully created. And currently, a multi-GPU code is on a working progress.

31.

GROUP STEINER PROBLEM ON THE GPU

Basileal Imana '17, Venkata Suhas Maringanti '17

Faculty Sponsor: Peter A. Yoon

Previous literature on VLSI routing and wiring estimation typically assumes a one-to-one correspondence between terminals and ports. In practice, however, each terminal consists of a large collection of electrically equivalent ports, a fact that is not accounted for in layout steps such as wiring estimation. In this research, we address the general problem of such minimum-cost routing tree construction in the presence of multi-port terminals, which gives rise to the Group Steiner problem (GSP). The minimal tree problem states that given a weighted undirected graph G , a minimum spanning tree is a sub-tree that contains all the vertices in G and minimizes the sum of the weight of the edges. Our research focuses on an extension of the problem where we are allowed to introduce special Steiner nodes with zero weight as junctions in order to minimize the cost of the tree. Common approach to the Group Steiner problem, known as the strong connectivity version, allows all the nodes of a group to be implicitly connected with each other which allows the solution to the problem to be a forest. However, the version of the problem that we are working on involves weak connectivity whose solution must strictly be a tree. The problem of interconnecting a net with multi-port terminals is a direct generalization of the NP-hard Steiner problem, and is therefore itself NP-hard which means that, theoretically, it cannot be solved in polynomial time. To that end, our research is focused on developing an efficient algorithm of the GSP on the GPU (Graphical Processing Unit) using CUDA programming and comparing the results with the serial implementation of the GSP on the CPU.

32.

HIGH-SPEED HYPERGRAPH MATCHING ALGORITHM

Kevin Liu '17, Reid Delaney '16

Faculty Sponsors: Peter A. Yoon, Lin Cheng

A hypergraph is a mathematic model that generalizes a graph. Its hyperedges can contain more vertices than the two in a normal graph, and this feature allows hypergraphs to model complex relations such as the inter-personal relationships of a social network website, or the reactions between compounds in a complex chemical reaction. Our research focuses on implementing a fast multi-GPU algorithm based on existing algorithms for solving hypergraph matching problem. The hypergraph matching problem occurs frequently in computer science and engineering, especially in areas such as image comparison and object recognition. So a faster hypergraph matching algorithm would greatly benefit these areas. Past algorithms of hypergraph matching are mostly designed for single CPU and are, therefore, slow. Now the multi-core GPUs are cheaply available and they are better at doing large number of floating operations than CPUs, we naturally want to design an algorithm that solves the hypergraph matching problem on the GPU. Starting from a probabilistic approach designed by Zass and Shashua, we now have a

single core parallel implementation that works for hypergraphs with degree two. We've begun to develop a parallel implementation of the algorithm, using specialized tools to utilize the multi-GPU system. Our next attempt will be to move onto a tensor-based approach and to extend the algorithm for hyper graphs of higher degrees.

33.

EXPANDER GRAPHS AND THEIR EIGENVALUES

Yicheng Shao '16, Peter Reheis '16

Faculty Sponsor: Professor Takunari Miyazaki

Expander graphs are a family of sparse and highly connected graphs. Their quality is often measured by the second largest eigenvalues of their adjacency matrices. A classical method to compute eigenvalues is the power method, which uses iteration towards convergence. We studied necessary conditions for the power method to maintain certain levels of accuracy on graphs of different sizes. We also compared the power method with Matlab's eigenvalue function. We conclude that for large graphs the power method performs better.

It has been theoretically proved that many random graphs are expander graphs. In this project, we conducted experiments with randomly generated 3-regular bipartite graphs. We found that, up to manageable sizes, very large percentages of such graphs are indeed expander according to the measure defined by second largest eigenvalues.

ENGINEERING

34.

TRANSCRANIAL DOPPLER ASSESSMENT OF CEREBRAL BLOOD FLOW VELOCITY DURING VISUOSPATIAL TASKS

Erin Barney '15

Faculty Sponsor: Dr. Gregory Bashford (University of Nebraska - Lincoln)

Transcranial Doppler (TCD) uses ultrasound to measure the blood flow velocity of cerebral arteries. In healthy subjects, if arterial diameter does not change, cerebral blood flow velocity is linearly related to cerebral blood flow and perfusion pressure. Changes in cerebral perfusion pressure allow the brain to regulate the amount of oxygen it receives. Typically, as the brain becomes more active, cerebral blood flow velocity increases. In this study, cerebral hemodynamics were studied during cognitive tasks requiring visuospatial processing. Participants' middle cerebral arteries were monitored with TCD while viewing images that required either visual searching for a letter or memorization of a scene for later recall. The majority of the participants showed an increase in blood flow velocity between the resting phase and the task phase, indicating an increase in brain activity. When comparing blood flow velocities from the left and right middle cerebral arteries, the left tended to have higher velocity than the right. This indicates that the left hemisphere was dominant in performance of these tasks. By studying typical physiological responses to cognitive tasks, it is possible to learn more

about the functional organization of the human brain, and it can provide a baseline for comparison with injured brains.

35.

STUDY OF NITROMETHANE COMBUSTION USING A SHOCK TUBE

Binod Giri '15

Faculty Sponsors: John D. Mertens, Eric L. Petersen

Combustion characteristics of Nitromethane (CH_3NO_2) were studied with the help of a shock tube at the Turbo Lab at Texas A&M University. An experimental matrix was designed to study the combustion at various pressures, temperatures, and percent dilution. The mixtures of Nitromethane and Oxygen diluted in Argon were introduced into the driven section of the shock tube, and the ignition delay times were measured. The ignition delay times obtained experimentally were compared to those modeled in Chemkin.

36.

DESIGN AND IMPLEMENTATION OF SEVERAL ENGINEERING PROJECTS

Bobby Tella '17, Catherine Poirier '17, Phillip Winser '17, Subekshya Bidari '17,
Sydney Doolittle '17, Tristan Peirce '17

Faculty Sponsor: John D. Mertens

The Trinity College Engineering Department conducts research in a broad spectrum within its field. Areas of interest include prosthetics, alternative fuel sources, materials, robotics, software, and neural engineering. Commercially available rapid prototyping machines have allowed prosthetics research to move towards the rapid creation of cheap, personalized prosthetic devices. A prototype of a prosthetic hand with moving finger and thumb phalanges was created for individuals suffering from digital amputations. In working with the robotics team, research was conducted in the design and discovery of engineering materials appropriate to create a durable, weatherproof cover for "Q," the Trinity College Robotics Team's autonomous robot. In the Biomedical Engineering lab, electrodes were prepared and a stereotaxic apparatus was used to insert them into rats' brains to study electrophysiological patterns. In researching alternative fuel sources, a 1985 Mercedes was modified to run on waste vegetable oil instead of diesel fuel, helping to minimize fuel costs and reducing dependence on fossil fuels.

ENVIRONMENTAL SCIENCE

37.

THE EFFECT OF CLEAR CUTTING ON SUB-ALPINE FOREST SOIL NUTRIENT CONCENTRATIONS OF ALUMINIUM AND CALCIUM WITHIN THE WHITE MOUNTAIN NATIONAL FOREST, NEW HAMPSHIRE

Jack Agosta '17, Justin Beslity '15

Faculty Sponsors: Jonathan Gourley, Robert A. Colter (Soil scientist from USDA Forest Service, White Mountain National Forest)

Clear-cutting is the most popular and economically profitable method of logging and has been in use for centuries to provide lumber. However, there are several negative side effects which may lead to an increase in soil erosion and nutrient loss. Aluminum and calcium have been found to be critical nutrients for forest ecosystems. Aluminum is an important nutrient for plant growth, especially for trees, and a deficiency in calcium could result in root degradation, leaf necrosis and inability to properly produce flowers and fruit. Collaborating with the USDA Forest Service, three small timber sales were selected for a long term study, which would be sampled for measurements of aluminum and calcium in the O and B layers of soil. Samples were collected prior to the cutting of the timber sales to measure the baseline soil nutrient content of the three plots in the summer of 2013, and samples have been collected one year after the initial sampling in order to analyze the changes in soil nutrient concentrations. The Inductively Coupled Plasma-Optical Emission Spectrometer (ICP-OES), is used to measure the concentrations of aluminum and calcium in the soil and GIS to provide an interpolated map of each timber sale. In order to confirm the accuracy of results and to compare future nutrient concentrations, samples with high deviations above ten percent or concentrations observed to be orders of magnitude higher or lower than expected, were reprocessed. The reanalyzed samples were then selected based off of precision and accuracy.

38.

QUANTIFYING VEGETATION COVER CHANGES IN RESPONSE TO INVASIVE PLANT REMOVAL TREATMENTS

Jacob Ammon '17, Gregory Reardon '15

Faculty Sponsor: Cameron Douglass

Vegetation cover is a measurement of the percentage of the ground covered by plants. This quantity can be assessed as overall cover of vegetation, or on an individual species basis. Vegetation cover is important in the analysis of ecosystem health because it describes vegetation density, and can be used to quantitatively measure changes in plant community composition over time. In order to estimate vegetation cover, visual estimates are normally made of the proportion of a given area. In an effort to avoid the observer bias that frequently influences visual estimations, we experimented with quantifying vegetation ground cover in digital photographs. A camera support apparatus was constructed to ensure photos were taken at the same angle. Digital images of vegetation sampling plots at Knox Preserve (Mystic, CT) were taken beginning in the fall of 2013 and continuing through the summer of 2014. The images were processed in Photoshop to correct white balance and then imported individually into ImageJ, a free NIH-

developed program. Selecting different hue and brightness threshold levels allowed for the selection of particular color ranges in the imagery, in this case, that of plant leaves. Images were then processed and the area consisting of green leaves measured.

Mean vegetation cover from the visual estimates and digital measurements were very similar. However, variability in data from digital measurements was consistently much lower than that of visual estimates. Between fall 2013 sampling and summer 2014 sampling, numerous improvements were made to the digital image collection and analysis process. These improvements were found to have further reduced variability among ground cover measurements made via digital imagery. In particular, the use of a polarizing lens filter and color balance card in each image significantly reduced variations between photos taken at different times throughout the day and in different conditions. Additionally, refining the image processing protocol minimized processing errors that had previously occurred. Our results indicate that this process can be reliably used to quantify vegetation cover going forward.

39.

UNDERSTANDING THE CHALLENGES OF MANAGING INVASIVE PLANT ASSEMBLAGES, A CASE STUDY AT THE KNOX PRESERVE

Tracy R. Keza '17

Faculty Sponsor: Cameron Douglass

Invasive plants are a significant environmental concern not just because they out compete native plants but also because they can reduce the functional capacity of ecological systems. Generally invasive species are considered problematic because they create dominant mono-cultural communities that negatively impact ecosystems. However, in particular habitats – such as shrub lands and forests in the northeastern U.S. – instead of invasive mono-cultures, co-dominant assemblages of multiple problematic species are becoming increasingly common. It's especially hard to manage assemblages of invasive plants because due to each species unique morphological or physiological attributes they typically require different control strategies. In this project we intended to investigate whether using integrated and adaptive management strategies it was possible to effectively manage multiple co-occurring invasive species while minimizing negative ecosystem impacts.

We established the study at Knox Preserve (Mystic, CT) in an area where multiple co-occurring invasive vines and shrubs were to be controlled. In treatment areas plots were installed in September 2013 so that baseline data on the plant communities could be collected, additional plots (including an untreated control) were set up in spring 2014. Preliminary results suggest that herbicide treatments followed by mowing provided effective control of targeted invasives but reduced desirable plant species richness and cover. Mowing alone initially did not effectively control invasive assemblages but did result in a significant increase in plant community diversity and cover. These data were used by the land owner (Avalonia Land Conservancy) to optimize follow up treatments made in the summer and fall of 2014. We will continue to collect data and establish plots to better determine the long term effects of Avalonia's invasive management, and hope to learn generalizable lessons from this project that can be applied to similar systems.

40.

PERSISTENCE OF FIRE INDUCED MAGNETIC SIGNALS IN SOIL

Kyaw San Min '17

Faculty Sponsor: Christoph Geiss

Soils contain iron minerals that make them slightly magnetic, and the resulting magnetic properties can reflect the environmental conditions during pedogenesis. An earlier study of prairie soils discovered that the fires have such effects on soil magnetic properties by increasing the abundance of very fine iron-oxide particles in the topsoil (Roman *et al.*, 2013). However, it is not known why this effect fails to persist for long. This study looks into whether this could be caused by the magnetic particles being washed away by rain and are accumulating downhill. Therefore, samples of soil are collected along a hillside and are measured for low-field magnetic susceptibility, frequency-dependent magnetic susceptibility and Anhysteretic Remnant Magnetization (ARM) as well as Coercivity Distributions. Our results so far are mixed. While the low-field magnetic susceptibility (χ) measurements and Coercivity distribution analysis show no pattern of being washed down hill, frequency dependent susceptibility (χ_{fd}) and Susceptibility of Anhysteretic Remanent Magnetization (χ_{ARM}) show a pattern of increasing magnetic properties as we go downhill. This only indicates that further study is needed.

41.

SOIL CHEMICAL AND PHYSICAL PROPERTIES ALONG ENVIRONMENTAL AND ECOLOGICAL GRADIENTS: A CASE STUDY AT KNOX PRESERVE, STONINGTON, CT

Jenna Wilborne '15, Jordyn Fisk '17

Faculty Sponsor: Cameron Douglass

Invasive plants are known to cause widespread ecological disturbances that can lead to reductions in biodiversity and change soil properties. For example, invasive plant species can alter soil carbon-nitrogen ratios because of their abundant biomass production and tissue chemistries. This project sought to characterize chemical and physical properties of soils collected across a habitat gradient - from an open grassland to a dense shrubland - at the Knox Preserve, near Mystic, Connecticut. Between fall 2013 and spring 2014, 867 soil samples were collected every 2.5 meters along transects following the prevailing habitat (and elevation) gradients. Samples were analyzed for soil moisture and carbon content using the loss-on-ignition method, and a soil conductivity meter was used for measuring soil salinity.

Soil moisture levels did not differ significantly (mean = 24.82%) between habitat types (forested vs grassland), but there were higher moisture levels at the northern edge of the grassland near a small pond. Organic carbon levels from forested soils were 47% higher ($P < .0001$) than grassland soils. Carbon levels in forested soils were more positively correlated ($R^2 = 0.35$, $P < .0001$) with soil moisture than carbon levels in grassland soils ($R^2 = 0.08$, $P < .0001$). Soil salinity levels were dramatically higher in samples collected surrounding the pond (mean = 35.41 mS/cm). Excluding these very high sample levels, salinity levels were significantly ($P < .0001$) higher in samples collected from the grassland habitat (mean = 490 μ S/cm) compared to the shrubland habitat (mean = 29 μ S/cm). Overall, there was a positive correlation ($R^2 = 0.34$, $P <$

.0001) between soil moisture and salinity levels in the grassland site, but a much weaker correlation ($R^2 = 0.09$, $P < .0001$) between these parameters in forested soils. Changes in elevation within the Knox Preserve likely explain the variation in salinity levels. Further studies will be carried out in the coming year to determine additional possible driving factors for habitat differences in organic carbon levels.

NEUROSCIENCE

42.

EFFECTS OF KETOGENIC DIETS ON AUTISTIC SYMPTOMS OF FEMALE EL MICE

Subrina Bisnauth '15

Faculty Sponsors: David Ruskin, Susan Masino

The ketogenic diet (KD) is a high fat, restricted carbohydrate, sufficient protein metabolic therapy that has been shown to significantly reduce seizures in persons with epilepsy which is often comorbid with autism. A restrictive KD reverses symptoms in the BTBR mouse model of autism but is a factor in its clinical applicability. In a study with the EL mouse model of epilepsy and autism, sex-dependent effects were found and effects were seen in females only. The strict and a milder KD was tested on female EL mice to compare their effects on blood chemistry and body weight. The moderate KD trended towards having lowered blood glucose and both diets resulted in significantly increased blood ketones. Only the strict diet resulted in weight loss. The difference in the ratio of fats, carbs and proteins as well as in the other nutrient components in the two diets is important to the weight and blood effects seen in the animals. The current study will investigate if increased ketones and lowered blood glucose are necessary for behavioral improvement. Further studies should be carried out on male EL mice to determine whether there are also sex-dependent effects of the moderate KD, as well as studies to determine which components of the diet are most important for improving symptoms and how the balancing of nutrients could impact the therapeutic effects.

43.

NEONATAL DESENSITIZATION AND IMMUNE-TOLERANCE IN RATS

Tasmerisk Haught '15

Non-Trinity Faculty Sponsors: Virginia Mattis, Ph.D. , Genevieve Gowing, Ph.D. , Clive Svendsen, Ph.D.

Neural xenografts are stem cell transplants from one species to another. Without appropriate immunosuppression, neural xenografts in adult rodent brains are rejected. However, by taking advantage of the underdeveloped immune system of neonates, a neonatal desensitization and immune tolerance to xenogeneic donor cells during this period could allow for transplant survival. Using one human fetal neural progenitor cell line and two induced pluripotent stem cell (iPSC) derived neural progenitor cell lines expressing luciferase, intraperitoneal injections were performed the day rat subjects were born to induce desensitization. Bilateral stem cell striatal injections were performed two months later to assess for xenograft acceptance following

desensitization. Luciferase imaging of desensitized rats done 73 days post-adult striatal transplant show preliminary evidence against xenograft survival. To determine if cells were surviving in the desensitized animals, the animals were sacrificed, stained and co-localization of human stem cell labeling, SC101, and the nuclear marker DAPI were imaged and surviving cells were counted. If cell survival is shown to take place, desensitization of neural stem cell transplants would allow for more cost efficient and effective animal models. However, if counts are consistent with the preliminary luciferase data already obtained, alternative methods such as the use of immunosuppressant drugs for survival of neural stem cell xenografts will remain necessary.

44.

A COMPARISON OF THE MEMORY FOR INTENTIONS SCREENING TEST AND THE CAMBRIDGE PROSPECTIVE MEMORY TEST

Constance Ky '17

Faculty Sponsor: Sarah Raskin

The Memory for Intentions Screening Test (MIST) is a paper and pencil test, designed to be a clinical measure of prospective memory through the use of time versus event and action versus verbal cues. The Cambridge Prospective Memory Test (CAMPROMPT) is another test that quantitatively measures prospective memory, but unlike the MIST, subjects are given the opportunity to record their instructions in order to aid them during the test. Although the CAMPROMPT creates a more realistic setting for the subject, it reduces the extent to which the participant must rely on prospective memory, whereas those who take the MIST must rely solely on prospective memory. Therefore, the goal of this research was to compare the MIST to the CAMPROMPT and determine if participants would receive the same scores on both tests. The results of the test reveal that participants scored differently on the two tests, with only a correlation between the event-based prospective memory questions, indicating that the different methods of testing had a significant effect.

45.

QUANTIFICATION OF PURINE CONTENT CHANGES IN MOUSE BRAIN FOLLOWING CHRONIC KETOGENIC DIET

Jacob Rubin '15

Faculty Sponsor: William H. Church

The ketogenic diet (KD) is well known for its efficacy in reducing frequency and severity of seizures in children with drug-resistant epilepsy. However, very little is known about the mechanisms by which the KD elicits these effects. Previous work in this lab examined KD-induced changes of catecholamine neurochemistry in several regions of the mouse brain. Dopaminergic metabolite ratios were increased in both the motor and somatosensory cortices suggesting an activation of the meso-cortical dopaminergic system. Adenosine has also been purported to be involved in the ketogenic diet's mechanism of action, so changes in the purinergic neurochemical system was investigated in the current work. Tissue homogenates of the motor cortex, somatosensory cortex, nucleus accumbens, anterior caudate putamen, posterior

caudate putamen and midbrain regions were analyzed using high pressure liquid chromatography to quantitate uric acid, guanine, hypoxanthine, xanthine, inosine, guanosine, and adenosine. At this point data from the analysis of 72 out of 96 brain samples have been processed. The remaining brain samples will be completed during the fall semester at which point data will be processed to assess how the KD influences purine levels in the aforementioned mouse brain regions. Information gathered from the current study may shed light on the mechanisms involved in the KD's anti-epileptic effects and thus afford the scientific community with the insights that will aid in the development of pharmaceutical alternatives to the KD.

46.

CELL DIFFERENTIATION STRATEGIES FOR NEONATAL INTRAVENTRICULAR HEMORRHAGE

William Schreiber-Stainthorp '15

Non-Trinity Faculty Sponsors: Joshua J. Breunig, PhD; Aslam Akhtar, MS

Intraventricular hemorrhage (IVH) is a condition which affects premature and low birth-weight infants. Over 12,000 cases are seen every year in the United States alone, with consequences that can often result in severe, lifelong disabilities or death. Cognitive impairments correlate well with white matter destruction in the brain. White matter is made up of oligodendroglial cells, which are generated during the myelination process that occurs from birth to young adulthood. Previous studies suggest that oligodendrocytes are preferentially lost in IVH and, thus, may constitute a target for cell therapy.

To better study IVH and evaluate potential therapies, we have designed a new animal model of the disease. Recent studies have shown that increased secretion of vascular endothelial growth factor (VEGF) is seen in patients and can cause IVH symptoms in the mouse brain. To this end, we created a plasmid to overexpress VEGF. This plasmid will be electroporated into the neural stem cells lining the lateral ventricles of newborn mice, and we hypothesize that that the increased VEGF secretion will lead to vascular leakage and IVH.

Using our model of VEGF induced IVH, we will test the strategy of directing differentiation of neural stem cells into oligodendrocytes as a cell therapy. Previous studies have shown that overexpression of several transcription factors – Sox10, Olig2, and Nkx6.2 or Zfp536 – can reprogram fibroblasts into functional myelinating oligodendrocytes. We created inducible plasmids containing these four transcription factors. With our inducible system, administration of doxycycline results in gene expression. Additionally, neural stem cells can be cultured from IVH patients as part of standard of care treatment and represent an autologous source which avoids the need for immunosuppression. Thus, these plasmids will be nucleofected into a culture of these neural stem cells and then transplanted into the IVH brain. Expression of the four transcription factors will then be induced by administration of doxycycline, differentiating the stem cells to oligodendrocytes which will potentially re-myelinate the neurons and promote neuronal function. Outcomes will be assessed based on survival, the differentiation of the stem cells to oligodendrocytes, and myelination in the IVH brain.

47.

REVERSIBILITY OF DIETARY EFFECTS ON SOCIABILITY IN BTBR MICE

Sierra Slade '15

Faculty Sponsors: David Ruskin, Susan Masino

Autism is a neurological disorder characterized, in part, by a deficit in sociability. The ketogenic diet (KD), a high-fat and very low-carbohydrate diet, has been shown to improve sociability in the BTBR mouse model of autism. KD is also used to reduce epileptic seizures in humans and rodent models of epilepsy, a neurological disorder comorbid with autism. Interestingly, reductions in seizures have been shown to be sustained in epileptic patients and rodent models after the diet is no longer administered. This study aimed to determine whether the increase in sociability seen in BTBR mice on KD is maintained or reversed when they are returned to a standard diet. In this study 5 week old BTBR mice were fed a 3:1 (fat : carbohydrates + protein) ketogenic diet for 3 weeks. After 3 weeks of dietary treatment their sociability was tested using the 3-chamber test of sociability in rodents. Immediately following, the reversal (experimental) group was switched to a standard diet while the control group remained on KD. One week later, both groups were re-tested. Results of the re-test showed that after being switched to the standard diet the mice had significantly lower sociability than displayed in their initial testing and significantly lower sociability than the control group which had remained on KD. The control group maintained a trend of increased sociability insignificantly different from the initial test. The study concludes that the dietary effects on sociability were reversed when diet therapy ended. Methodological concerns include the brief duration of the study and the abrupt removal of the reversal group from KD. The results imply different mechanisms by which the diet ameliorates epileptic seizures and the behavioral symptoms of autism. Future studies will examine the reversibility of the dietary effects after extended (longer than 3 week) use of KD.

48.

ANALYSIS OF THE ACTIVITY OF AN ASTROCYTE SECRETED FACTOR ON SH-SY5Y NEUROBLASTOMA CELLS

Nathaniel Thiemann '17, Sheila Njau '17, Thomas Naragon '17

Faculty Sponsor: William H. Church

Astrocytes are glial cells that are found in the central nervous system and perform various functions that aid with the growth and survival of neurons. Neuroblastoma is a type of solid extracranial tumor that usually develops in young children. When astrocyte conditioned media is applied to SH-SY5Y neuroblastoma cells, there is a significant increase in cell death from that observed in the control (DMEM F-12). This suggests that astrocytes release a factor that induces SH-SY5Y cell death. Addition of TIMP-1, a tissue inhibitor of metalloproteinases, prevents cell death in cells treated with wild type astrocyte media. Heating the wild type media eliminated the increase in cell death, suggesting an enzyme like factor. To begin to narrow down the size of this cell death inducing factor, fractions of the media were obtained through ultracentrifugation of wild type astrocyte media and TIMP-1 KO astrocyte media at 50kDa and 100kDa. The results suggest that this factor weighs between 50kDa and 100kDa and somehow interacts with TIMP-1 to mediate cell death. Also, there may be a neurotrophic factor that weighs above 100kDa that

protects the SH-SY5Y cells against the CDIF. Learning the identity of this factor could have implications on research dealing with cancer as well as neurodegenerative diseases.

POLITICAL SCIENCE AND PUBLIC POLICY

49.

EXTERNAL FORCES, INTERNAL RESPONSES: LOCAL GOVERNMENT POLICIES TOWARD IMMIGRANTS OVER TIME

Magdalena Filippone '15, Bettina Cecilia Gonzalez '16

Faculty Sponsor: Abigail Fisher Williamson

In studying local government responses to immigration, we find that external forces play a role in the tenor of local reactions. Federal policies and national political context together with local characteristics shifts local policy responses. Additionally, external scrutiny, or reactions by the media and outside organizations, put pressure on municipalities that can cause them to rein in their restrictive ordinances. We examined 54 cities and towns that passed immigrant-related restrictive ordinances in 2006-2007 and hypothesize that there is a relationship between external scrutiny and the reining in of restrictions on immigrants. Our findings suggest that heightened attention to external anti-discrimination policies and the economic costs of bigotry gives immigrants the definition of minorities in need of protection, which leads to the taming of local restrictive ordinances. Meanwhile, when national rhetoric classifies an immigrant as an illegal alien, restrictive responses are not considered to be discriminatory.

PSYCHOLOGY

50.

DESIGNING A RELIABLE CODING SYSTEM FOR METACOGNITION

Melody Fulton '15, Jen Schackner '15, Julia Sager '15

Faculty Sponsors: Dina Anselmi, David Reuman

There are only a few quantitative measures of metacognition suitable for children and adolescents. Moreover, there are no explicit qualitative measures of metacognition aligned with Ambrose et al.'s 5-step model of metacognition. A qualitative measure of metacognition was developed last year and given to 8th grade social studies students as part of a metacognitive intervention study. In order to validate this new measure as a reliable assessment of students' metacognition, we practiced the coding of student responses based on a coding system until we became proficient coders. A correlation of $r=.8$ is considered an acceptable standard for inter-rater reliability for research purposes. After making adjustments to the coding system and performing several practice checks, we reached an inter-rater agreement of $r=.87$. Based on these results, future coders should now be able to apply the MC-5 coding system to accurately assess the qualitative metacognitive skills of 8th grade social studies students.

51.

SPEECH PRODUCTION CHANGES AND INTELLIGIBILITY WITH A REAL-TIME COCHLEAR IMPLANT SIMULATOR

Lily Talesnick '15

Faculty Sponsor: Elizabeth Casserly

Subjects hearing their speech through a real-time cochlear implant (CI) simulator alter their production in multiple ways, e.g. reducing speaking rate and constricting F1/F2 vowel space. The motivations behind these alterations, however, are currently unknown. Two possibilities are that the changes in speech are due to the influence of a direct feedback loop in which the subject is adjusting speech production to minimize acoustic “error,” or that the changes could reflect the indirect influence of a high cognitive load (stemming from the challenge of hearing through the real-time CI simulator). We explored these two possibilities by conducting a playback experiment in which 35 naïve listeners assessed the intelligibility of speech produced under conditions of normal versus vocoded feedback. Intelligibility of vocoded isolated word stimuli in each condition was tested in both a two-alternative forced choice task (“Which recording is easier to understand?”) and an open-set word recognition task. Listeners found normal-feedback speech significantly more intelligible in both tasks (p 's $< .0125$), suggesting that speakers were not adjusting for direct error correction, but rather because of the influence of an intervening factor, e.g. high cognitive load. Confusion matrix analyses further illuminate the perceptual consequences of the effects of CI-simulated speech feedback.