

Undergraduate Research Symposium

Icahn School of Medicine at Mount Sinai

**Saturday, September 16th 2017, 9am-4pm
Guggenheim Pavillion, 1468 Madison Ave, New York, NY 10029**

Abstract Book



**Icahn
School of
Medicine at
Mount
Sinai**

*Graduate School of
Biomedical Sciences*

Event Schedule

9:00 am (ongoing) – Annenberg Atrium

Symposium Check-In & Breakfast

10:00 am – 10:15 am – Hatch Auditorium

Opening Remarks & Welcome Address
(Deans of Graduate School & Organizing Committee)

10:15 am – 11:30 am – Annenberg Atrium

Morning Poster Session*
(Odd Numbers)

11:30 am – 11:45 am – Hatch Auditorium

PhD Programs Overview

11:45 am – 1:00 pm – Hatch Auditorium

Plenary Talks Selected from Abstracts

1:00 pm – 1:30 pm – Outside Hatch Auditorium

Lunch
(Provided by the Graduate School of Biomedical Sciences)

1:30 pm – 2:45 pm – Annenberg Atrium

Afternoon Poster Session*
(Even Numbers)

2:45 pm – 3:00 pm – Hatch Auditorium

Master Programs Overview

3:00 pm – 3:45 pm – Hatch Auditorium

Data Blitz
(Current students of the Graduate School of Biomedical Sciences)

3:45 pm – 4:00 pm – Hatch Auditorium

Award Presentations and Closing Remarks

Invited Talks

1. Niuska Alvarez
2. Samvida Venkatesh
3. Alexa Friedman
4. Ann Lin
5. Chi Nguyen
6. Suppawat Kongthong
7. Thomas Nguyen

Posters

Presenter (Last)	(First)	Poster #
Alvarado Mateo	Diego	1
Alvarez	Niuska	2
Alvarez	Jonas	3
Babalola	Kabiru	4
Babar	Mustufa	5
Bartholomew	Myles	6
Basri	Noah	7
Bedard	Gabriel	8
Behun	Marissa	9
Cha	Jennifer	10
Cimino	Francesco	11
Coronado	Daniela	12
Duan	Nicole	13
Friedman	Alexa	14
Gabbireddy	Sairisheel	15
Gobin	Dupah	16
Goradia	Eshani	17
Gurkov	Iurii	18
Halawath	Jeran	19
Hoying	Madelyn	20
Iglesias-Montoro	Patricia	21
Jeronimo	Carlos	22
Jones	Amber	23
Khalizova	Nailya	24
Kihui	Maryanne	25
King	Sarah Ann	26
Kongthong	Supawat	27
Lee	Serena	28
Lee	Zachariah	29
Limon	Angelina	30
Lin	Ann	31
Malik	Hagerah	32
Martin	Hannah	33

Presenter (Last)	(First)	Poster #
Mikofsky	Rachel	34
Mirkovic	Jovan	35
Morris	Aaron	36
Murillo	Fernanda	37
Na Javeed	Suha	38
Nation	Rachel	39
Nguyen	Chi	40
Nguyen	Thomas	41
Noreña	Luke	42
Oguntuyo	Kasorelope	43
Pace	Jesse	44
Parham	Imari	45
Patel	Saloni	46
Pizano	Katherine	47
Pugh	Derian	48
Rucinski	Violetta	49
Samuel	Savannah	50
Samuel	Roy	51
Scarlata	Miranda	52
Shah	Lopa	53
Smith	Ian	54
Steinsnyder	Zoe	55
Stimpson	Don	56
Stoerback	Amanda	57
Tyer	Margaret	58
Vargas	Danielle	59
Venkatesh	Samvida	60
Vo	Phuong	61
Wang	Yuqiao	62
Woodbury	Jesse	63
Xiong	Laura	64
Yang	Leon	65
Yen	Frederick	66

THE ROLE OF GLUA1 AND GLUA3 TRAFFICKING IN THE BASOLATERAL AMYGDALA DURING PAVLOVIAN REWARD CONDITIONING

Diego Alvarado Mateo
York College

Mentor: Peter Serrano

Pavlovian Conditioning (PC) is a behavioral model of learning that occurs through associations. In our present study, we sought to characterize the molecular mechanisms in the brain that underlie PC. Specifically, we focused on a paradigm of PC that pairs a conditioned stimulus (CS), a light, and an unconditioned stimulus (US), a food reward. Accordingly, rats underwent 8 days of PC, followed by 5 days of extinction training or control exposure. Rats were subsequently tested for 4 trials, sacrificed, and their brains collected. Previous studies with rodents suggest that certain structures in the brain (basolateral amygdala (BLA), Nucleus Accumbens (NucAcc), Dorsal Striatum (DS)) play pivotal roles in PC. From the tissue collected, we analyzed protein concentrations in synaptic fractions of these brain regions from the respective groups. The proteins that were analyzed include: AMPA receptor subunits GluA1, GluA 2 and GluA 3, and Protein Kinase C ζ . These markers can reveal changes in synaptic plasticity that may occur as a result of PC and extinction. Behavioral analyses from the test day showed that the PC group made more conditioned magazine approaches in response to the CS than either of the other two groups. Western blotting has revealed that in the BLA, PC and PC+Extinction groups, both displayed higher levels of GluA2 and PKC ζ expression. In the same region, GluA1 expression increased in the extinction group only. Additionally, the NucAcc shows increased expression of GluA1 in both PC+Extinction and No PC groups. Finally, the DS shows significantly higher GluA3 expression in the PC group compared to other groups. These data suggest that the trafficking of specific AMPA receptor subunits could underlie distinct aspects of learning. We provide a novel perspective on the mechanisms underlying PC.

Evaluating Human Cell Survival and DNA Damage After Exposure to Various Amount of Chlorine Dioxide and Exploring its use as a potential cancer chemotherapy agent; especially for cancer cells which harbor defected DNA repair genes

Niuska Alvarez
University of New Haven

Mentor: Ali Senejani

Chlorine Dioxide is currently been used for the treatment of drinking water and food preservation. The present study examines the cell viability and DNA damage in response to Chlorine Dioxide administration.

Multiple ClO₂ dilutions were established ranging from 0.1% to 100% to which human and mouse embryonic cells were submitted for 30 minutes on ice. 24 hours after the treatment, the metabolic activity of the cultured samples was determined via MTT, a colorimetric assay. The desired concentrations were selected for the treatment of HEK293 cells to be used for the COMET assay employed to determine single and double strand breaks caused by ClO₂ dosing.

The MTT assay analysis of the HEK293 cell cultures showed a 50% survival at a dose of 0.02% when compared to the untreated control. Additionally, the cell types MEF₁ (+/+) and MEF₁ (-/-), the latest missing an essential DNA repair enzyme, both also treated with the same conditions showed to maintain a survival relative to the control at doses lower than 0.05%. However, in comparison to the wild type, the pol β deficient MEF₁ cells expressed a considerable reduced viability.

Furthermore, according to results obtained from the COMET assay, a significant difference could be observed on the analysis of the tail moment in response to an increase in dose from 0.1% to 1%, having the 1% treated sample the most damaged DNA.

Chlorine Dioxide appears to decline the survival of the cells studied and it caused possible irreparable damages at doses higher to 0.02%. Nonetheless, DNA repair deficient cells seem to be more susceptible to the treatment, a particularity that could use for medical advantage.

APPLYING METABOLOMICS TECHNIQUES TO IDENTIFY BIOMARKERS IN ALZHEIMER'S DISEASE

Jonas Alvarez
University of Pennsylvania

Mentor: Arjun Sengupta

The field of Metabolomics and the techniques that it encompasses offer a great insight into the chemical mechanisms that manage metabolism. Many diseases arise from irregularities in metabolism. Conversely, several diseases affect specific physiological pathways which are ultimately reflected in global metabolic status of organism. Recent literature suggested that classical “non metabolic disorders”, such as Alzheimer's disease (AD) can be better understood using metabolic profiling of biofluids. In this study, we sought to gain better insight into the mechanism of progression of AD and identify potential biomarkers that are linked to the symptoms of AD. We examined the Cerebral Spinal Fluid (CSF) samples of patients with AD or mild cognitive impairment and age matched healthy controls using Nuclear Magnetic Resonance (NMR) spectroscopy. Multivariate statistical analytical strategies were employed to analyze the data. Preliminary results suggest distinct metabolic characteristics that varies with cognition state and clinical biochemistry features of AD patients. Further processing of the data is ongoing that will help in gaining insightful knowledge of clinical and translational relevance.

SERO PREVALENCE OF BRUCELLOSIS IN BOVINE IN ILE IFE ABBATTOIRS OSUN STATE NIGERIA

Kabiru Babalola

Obafemi Awolowo University, Ile Ife, Osun State, Nigeria.

Mentor: Nkem Torimiro

Background

A cross-sectional study was conducted to determine the sero prevalence of bovine brucellosis among cattle slaughtered at several abattoirs of Ile Ife, Osun State, South West, Nigeria. In Nigeria, bovine brucellosis is a major animal health problem affecting the growth of the cattle industry. It remains a significant disease in animals and humans worldwide and an important cause of reproductive failure such as abortion in cows and sterility in bulls (Mekonnen, et al., 2010; Gul et al, 2013). Bovine brucellosis is a disease with a significant economic and public health importance due to losses incurred as a result of infertility in animals and extensive chronic morbidity in humans (Gwida et al, 2016, Al Dahouk et al, 2013).

Research question

This study is aimed to determine the sero prevalence of Brucellosis in cattle slaughtered for consumption in the study areas in the south west Nigeria, especially in Osun state, Ile Ife.

Approach and experiments

A total of 24 sera samples were collected over a period of seven days from 24 slaughtered cattle in five different local abattoirs in Ile Ife and were screened for brucellosis using Rose Bengal Plate Test(RBPT).

The Rose Bengal antigen will react with the brucella antibody in the sera if positive to give any degree of agglutination or clumping.

Conclusion

It was observed in this study that; Brucellosis prevalence is very low in the study centers.

The study therefore suggests surveillances of cattle slaughtered for Brucellosis to avert a future outbreak of disease.

STRUCTURE AND ACTIVITY RELATIONSHIP STUDY OF NOVEL LPXC INHIBITORS THROUGH RESIDENCE TIME

Mustufa Babar
Stony Brook University

Mentor: Peter Tonge

As resistance to currently available antibacterial agents continues to grow, particularly resistance resulting from Gram-negative bacteria, huge threats are inundated onto the human population. LpxC, an enzyme that catalyzes the first committed step in lipid A biosynthesis pathway, has recently been under scrutiny by the scientific community for developing novel antibacterial agents. Although various LpxC inhibitors have been reported in journals, none have reached the market due to safety concerns. We outline synthetic steps toward a structure and biological activity study of LpxC inhibitor derivatives of PF5081090, introduced by Pfizer company, which has shown superior Gram-negative antibacterial efficacy when compared to other previously reported inhibitors. We will determine *in vitro* activity of the inhibitor through a kinetic parameter, residence time, as it models the dynamic *in vivo* environment. We believe through optimization of this kinetic parameter of inhibitors, we would be able to not only improve their time-dependent activity but also their safety profile.

Acknowledgments: This project couldn't be conducted without the support of Peter Tonge, the PI, as well as all the members of Tonge Lab

MUTAGENESIS OF CLINICAL *PSEUDOMONAS AERUGINOSA* ISOLATES TO ASSESS PHAGOCYTTIC SUSCEPTIBILITY

Myles Bartholomew
Dartmouth College / Xavier University of Louisiana

Mentor: Brent Berwin

Pseudomonas aeruginosa is a gram-negative opportunistic pathogen. It is notorious for causing chronic lung infections in immunocompromised individuals and especially amongst patients diagnosed with cystic fibrosis (CF). A major factor in the pathogenicity of *P. aeruginosa* is its flagellar motility. Lack of flagellar motility has been shown to be a determining factor in the bacteria's ability to evade phagocytosis by host immune cells. For CF patients, the prodromal stage transitions to a chronic infection characterized by the bacterial downregulation and loss of flagellar expression. In order to understand the importance of this trend, it is crucial that research is conducted on clinical isolates of identical *P. aeruginosa* that differ only in motility. We hypothesize that, with transposon mutagenesis, we will engineer paired clinically derived motile and non-motile *P. aeruginosa* isolates, thus identifying the contribution of motility to phagocytic susceptibility. To address this hypothesis, we are currently engineering non-motile *P. aeruginosa* through transposition with *E. coli* SM10. With these phenotypically non-motile mutants, we will identify the mechanisms and differences in phagocytic susceptibility in *P. aeruginosa* clinical isolates as compared to their close mutant relatives. Our long-term plan is to leverage these mechanistic findings and studies to identify novel treatments for CF patients with chronic bacterial infections.

Notes:

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SilenshR: Bacteria-Mediated Oncogene Silencing as Living Cancer Therapeutic

Noah Basri
Columbia University

Mentor: Harris Wang

RNA interference (RNAi) therapies modulate endogenous gene expression in target cells through introduction of exogenous short interfering RNAs (siRNA) or their precursors, short hairpin RNAs (shRNA). Challenges for efficient and cell-specific RNAi therapies abound, such as rapid renal clearance, degradation by serum nucleases, traversing the lipid bilayer and escape from the intracellular endosome. Bacteria have shown to innately colonize the hypoxic and immune-privileged cores of tumors and as such have been explored as potent delivery systems for RNAi-based therapeutics for cancer. We aim to engineer a bacteria-mediated RNAi gene therapy, utilizing recombinant *E. coli* with the capacity to invade mammalian cells and deliver an shRNA payload targeting the aberrantly expressed receptor tyrosine kinase EGFR (Epidermal Growth Factor Receptor) and transcription factor c-Myc. *E. coli* uptake by mammalian cells and subsequent endosomal breakdown is mediated by a quorum-inducible invasin HlyA operon. We are now characterizing the circuit by assessing RNAi efficiency in vitro using immortalized HeLa and prostate cancer cell lines. Target oncogene knockdown is assessed through flow cytometry and qRT-PCR, and bacterial invasion is evaluated via gentamicin protection assays.

Is it Really Iminium Catalysis? Mechanistic Investigation of the Hantzsch Ester Reduction using Kinetic Isotope Effects (KIEs)

Gabriel Bedard
State University at Binghamton, Binghamton New York

Mentor: Mathew Vetticatt

ABSTRACT: The selective reduction of carbon-carbon bonds in an α,β -unsaturated aldehydes using the Hantzsch ester was reported in 2004 by List and coworkers. Hantzsch esters have unique reductive properties similar to their biological counterpart, NADH, when presented with a good electrophile. Addition of secondary amine salts to enals were thought to create *in situ* iminium ion, thus lowering the LUMO activation energy barrier and creating a better electrophile without the use of expensive metal catalysts, however the exact rate determining step (RDS) of the proposed mechanism is unknown. Herein we report the Kinetic Isotope Effect (KIE) for the title reaction in order to elucidate mechanistic details. We report a 1.011 and 1.036 isotope effects on the carbonyl and beta carbon, respectively, which may refute previously proposed pathways and support a concerted mechanism involving a loss of water and addition of the hydride to a carbinolamine intermediate. Computational analysis, however, supports a co-rate determining model in which formation of the iminium ion and attack of the nucleophile are equivalent in energy (approximately 0.5 kcal/mol difference). Our insight may suggest a paradigm shift in understanding secondary amine catalysis with α,β -unsaturated aldehydes.

FABRICATION OF A TIMED-PRESSURE REGULATOR (TPR) TO ENABLE THE STUDY OF BLADDER PAIN

Marissa Behun
Duquesne University

Mentor: Benedict Kolber

Interstitial cystitis/bladder pain syndrome (IC/BPS) is a common, poorly treated chronic pain disorder. Many IC/BPS symptoms suggest a central nervous system component. The role for the brain in processing and modulating bladder pain is still largely unknown. *In-vivo* recordings of the left and right central amygdala using carbon-fiber electrodes were made during noxious bladder distention in *Mus musculus* (mouse). Once a neuron was detected during the recording process, urinary bladder distensions (UBD) were used to observe how the neuron responded to a noxious stimulus. Currently, UBD is done using a manual distention device. To better standardize UBD data, a timed-pressure regulator (TPR) has been designed and built. The TPR consists of a microcontroller, pinch valve, and pressure sensor that will be able to regulate the various air pressures during UBD, while also monitoring the duration of UBD. TPR instrumentation is currently undergoing testing before being implemented in the recording process.

A better understanding of how the brain modulates bladder pain is an important finding because with this data, better treatments can be developed to not only help treat chronic bladder pain patients, but a variety of chronic pain patients. The lateralized effect of this data shows that hemispheric brain lateralization, well known in the context of language and memory, may play important roles in the development and treatment of disease states.

Fatty acid β -oxidation as a novel target for cisplatin-resistant ovarian cancer

Jennifer Cha

Rensselaer Polytechnic Institute/Albany Medical College

Mentor: Ellen Cheon

Ovarian cancer is the most lethal gynecological cancer and is predicted to affect one in 70 females in the United States. Cisplatin is the most common treatment for ovarian cancer but fast development of cisplatin resistance severely limits treatment success. The goal of this study is to uncover the mechanism by which cisplatin resistance is acquired and identify novel therapeutic targets to overcome chemoresistance. We identified collagen type XI alpha 1 (COL11A1) as a novel biomarker strongly associated with poor survival, chemoresistance, and recurrence in high grade serous ovarian cancer, the most common type of ovarian cancer. COL11A1 inhibits cisplatin-induced apoptosis in ovarian cancer cells but not by cisplatin efflux, inactivation, or DNA repair. Our proteomics data instead suggests that mitochondrial fatty acid β -oxidation (FAO) is the most upregulated pathway by COL11A1 via the overexpression of several key FAO enzymes. The role of FAO in ovarian cancer chemoresistance is largely unknown. Our results showed that COL11A1 enhances key FAO enzymes – CPT1A, HADHA, ACSL1, and ACAA2. Significantly, we confirmed that ovarian cancer cells increase both fatty acid uptake and oxygen consumption rate (OCR) in response to palmitate in the presence of COL11A1. Inhibition of FAO using shRNA against CPT1A attenuated the function of COL11A1 in cisplatin resistance. Taken together, our results suggest novel mechanisms by which COL11A1 confers cisplatin resistance and uncovers FAO as a promising therapeutic target for cisplatin-resistant ovarian cancer.

Investigating a Novel Cytotoxic EGFRvIII-Targeted Fusion Protein for Use in Cancer

Therapy

Francesco Cimino
Fordham University

Mentor: Patricio Meneses

Cancer claims millions of lives annually; in both 2014 and '15, the CDC reported that over 500,000 Americans died from cancer. Despite numerous advancements in the field of cancer biology, many problems remain. Adverse side effects of cancer treatments, including hair loss, fatigue, nausea and loss of appetite, arise from healthy tissue damage. The challenge to creating an effective cancer therapy that is preservative of healthy tissue, lies in the ability to destroy only cancerous cells. This research aims to develop a toxic, target-specific fusion protein, utilizing two molecules previously investigated in cancer therapies. Employing a peptide (PEPvIII) specific to the mutant Epidermal Growth Factor Receptor variant III (EGFRvIII), we intend to deliver the fusion protein exclusively to cancerous cells so that the other half of our molecule, a toxin named Streptolysin-O (SLO), may induce a pore, resulting in concentration gradient disruption and eventual cell death. The transformation of our protein into bacterial and mammalian cells will produce cells that can express our protein, as well as permit extraction of the isolated protein, upon secretion from mammalian cells. The cells and isolated protein will be administered to known EGFRvIII cell lines to assess their viability as therapeutic agents. EGFRvIII expressing cancers are especially resistant to apoptosis and currently account for 87% of Glioblastoma Multiforme, 67% of breast cancers, and 16% of lung cancers. Our intent is to provide new insights into possible alternate methods of cancer therapeutics, and if successful, to minimize tumor growth and metastasis in EGFRvIII expressing cancers.

TOWARDS AN UNDERSTANDING OF THE ROLES OF THE NF- κ B TRANSCRIPTION FACTOR RELB IN INNATE IMMUNE RESPONSES

Daniela Coronado
National Institutes of Health

Mentor: Ulrich Siebenlist

Allergic asthma is a chronic inflammatory disease that consists of both innate and adaptive immune responses. Previous studies conducted in the laboratory linked the non-canonical NF- κ B transcription factor RelB to lung inflammation in response to House Dust Mites (HDM) challenges, a model for allergic asthma. Lung inflammation in patients with severe asthma has also been linked to type I interferons. Therefore, there is a potential relationship between RelB and type I interferons in the inflammatory responses of patients with allergic asthma. By investigating this connection, we will be able to deepen our understanding of how the body regulates lung inflammation in patients with allergic asthma.

In order to begin our study of this relationship, we generated bone marrow-derived macrophages *ex-vivo*, and compared how the loss of RelB in these macrophages impacted the production of innate immune response cytokines under polyI:C stimulus. We focused on the cytokines IFN β , IFN α , and CXCL10 because IFN β and IFN α are type I interferon cytokines known for their presence in viral infections, while CXCL10 is an interferon-induced chemokine, which has also been associated with infectious diseases. We found that loss of RelB in bone marrow derived macrophages impacted the polyI:C-induced levels of both RNA and protein of these inflammatory mediators.

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

Nicole Duan
Trinity College

Mentor: 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.

Alexa Friedman
University of Connecticut

Mentor: Nichole Broderick

The gut microbiome of many model organisms has been shown to play a large role in host immune response. Preliminary studies in our lab found that axenic flies may have a reduced susceptibility against acute (high dosage) bacterial infection. To further investigate the impact of the microbiome on infection, we studied both an acute (high dosage OD600 200) and chronic (low dosage OD600 2) infection of *Pseudomonas entomophila* (*Pe*) on conventionally reared and germ-free (axenic, Ax) Canton (Cs) *Drosophila* flies. Results suggest susceptibility to infection may change based on the dosage of the infection. Furthermore, results suggest that chronic infections cause a further decline in fertility compared to acute and control conditions.

NOTCH3 proteolysis between EGF2/EGF3 in CADASIL: Probing the effect of proline at the cleavage boundary

Sairisheel Gabbireddy
University of Michigan

Mentor: Michael Wang

Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL) is a form of cerebral small vessel disease that can lead to large changes in brain vasculature. One of these changes is the degeneration of smooth muscle around blood vessels, which may lead to stroke. CADASIL is attributed to an autosomal dominant mutation in the gene NOTCH3, however, the exact mechanism of disease progression linked to the gene is unknown. One hypothesized outcome of mutated Notch3 is the impairment of normal disulfide bonding leading to protein deformity. We have recently discovered that the Notch3 protein is cleaved by an unknown mechanism at the first two boundaries between EGF repeats 1-2 and 2-3. A common dipeptide sequence, aspartate and proline (DP), is found at both of these boundaries. We hypothesize that the proline residue at the second position of this cleavage target sequence is required for efficient processing at the boundary sequence between EGF2 and EGF3. After mutating the proline residue in the second boundary sequence, we found that Notch3 proteins containing proline in the EGF2/EGF3 boundary were cleaved, on average, 4.5 times more than proteins that contained non-proline residues. Most residues yielded less cleavage than the wild type protein. Thus, we found that proline at the boundary sequence was required for efficient proteolysis of Notch3. A possible explanation of the proteolysis event is that proline's ring structure allows for a favorable interaction between mutated cysteine residues and the carboxyl terminal of aspartate, resulting in the cleavage of the boundary sequence. Future studies should investigate whether the mutation of specific cysteine residues changes processing of Notch3 at the DP2 boundary region. Additional research should analyze if Notch3 proteolysis results in different protein conformations, suggesting different mechanisms for Notch3 cleavage.

Analysis of How Recent Urban Constructions Limits Stingless Bee (*Tetragonisca angustula*) Gene Flow in Faria De Santana Brazil.

Dupah Gobin

Home university is CUNY York College however research was conducted at State University of Faria de Santana, Brazil

Mentor: Eddy Oliveria

In this study, I examine the influence of how recent urban constructions limits stingless bee (*Tetragonisca angustula*) gene flow in Faria De Santana, Brazil. *Tetragonisca angustula*, popularly known in Brazil as Jataí bee which is a meliponini bee that can be characterized by its rusticity and its important ecological role. *Tetragonisca angustula* is one of the most popular stingless bees in the Neotropical region. Over recent years, there have been potential declines in native pollinator communities that have raised serious concerns about pollinator conservation and dispersal of native pollinators across human-altered landscapes such as in urban landscapes in Faria De Santana, Brazil. Stingless bee species as of recently are becoming endangers because of the habitat fragmentation and anthropogenic activities. Evidence of local dispersal was gathered by investigating the relatedness, or the degree of shared genotypes, between individual stingless bees over small spatial scales. I examined the genetic structure of *Tetragonisca angustula*, in order to determine if urban transformation influences local genetic differentiation patterns and investigate whether bees exhibit fine-scale relatedness indicative of local dispersal. Specifically, I hypothesize that (i) recent barriers can generate local genetic differentiation (ii) stingless bees will exhibit high levels of relatedness at the 1–5 km spatial scale, indicative of local queen dispersal. I test these hypotheses using field surveys, land use maps and local and fine-scale population genetic analysis. Microsatellite markers were developed from an enriched genomic library of *Nannotrigona testaceicornis* and was characterized using *Tetragonisca angustula* samples. The genetic differentiation patterns measured at local scales may reveal of dispersal patterns and could possibly provide insight into local barriers to stingless bee dispersal, such as topographic features (urban constructions such as tunnels, bridges, and viaducts) and human-altered habitat. Results might have implications for understanding the impacts of urbanization trends on stingless bees. Also, the genetic data in this area could essentially be used to better understand the dynamic of *Tetragonisca angustula* and for the development of conservation strategies.

The Effects of Storage Conditions on Stability of an Antigen-Model Protein within the ImmunoMatrix Patch

Eshani Goradia
Stony Brook University

Mentor: Katarzyna Sawicka

According to the World Health Organization, vaccinations help prevent approximately 2 to 3 million deaths annually and could help protect up to 1.5 million more if vaccine coverage were improved. Traditionally, vaccines are administered as intramuscular injections and require refrigeration for storage. In order to improve access to vaccines researchers around the world aim to create devices that are patient-friendly and offer improved stability of immunizations, especially in areas lacking medical infrastructure such as refrigeration. The ImmunoMatrix needle-less vaccination patch, previously developed in our lab, non-invasively administers vaccines through the skin, thus removing the need for medical personnel, bio-hazardous waste, or expensive syringes and needles. The noninvasive vaccine patch is composed of electrospun nanofibrous mats that are characterized by large surface area to volume and mass ratios for improved skin to patch interaction. In an effort to understand and compare the storage stability of our patch to that of a traditionally composed protein/vaccine solution, in this study we examined the biological activity of horseradish peroxidase (HRP), a 44,000 Dalton enzyme, for 15 weeks at various temperature and humidity storage conditions. We found that the biologic activity of HRP in liquid formulation decreased by as much as 50% within the first 24 hours when stored in refrigeration. The enzymatic activity of HRP degraded at a slower rate for the electrospun nanofiber mats under controlled humidity conditions. We were able to determine two compositions of the patch that prolonged the storage stability of the enzyme for 8 weeks. Although conventional methods of protein stabilization such as lyophilization are widely employed, they have the potential to alter protein structure and thus reduce their biological activity. None of the methods offer notable improvement over the current vaccine paradigm, thus the ImmunoMatrix patch offers significant advantage for antigen shelf life under certain storage conditions.

Ether-Linked Lipid Biosynthesis is Important for Growth, Chemotaxis, and Development in *Dictyostelium discoideum*.

Iurii Gurkov
Hunter College, City University of New York

Mentor: Derrick Brazill

Signal transduction is a fundamental process by which cells regulate their metabolism, development, and movement. Signals from outside of the cell are transmitted across the membrane triggering series of molecular events that lead to a cellular response. Inositol phospholipids, as membrane components, play important roles by acting as second messengers in a variety of signaling pathways. Therefore, regulation and synthesis of inositol phospholipids is central to normal cellular activity. One of the enzymes that has been shown to provide both substrates required for ether linked, inositol phospholipid synthesis is FARAT (Fatty Acy Reductase Acy Transferase). FARAT is an evolutionary conserved protein, that has two domains, FAR (fatty acyl reductase) and AT (acyl transferase), that are responsible for the synthesis of fatty alcohols and 1-acyl DHAP (1-acyl dihydroxyacetone phosphate) molecules respectively. In order to assess FARAT's biological importance, the social amoebae *Dictyostelium discoideum* was used as a model organism due to its shared biochemical pathways with mammalian systems. We find that GFP-FARAT partially localizes to sites of lipid synthesis, consistent with its proposed function. Wild type cells and cells lacking or overexpressing FARAT were characterized for developmental morphology, growth and chemotaxis to folate. Spore counting assays demonstrate no impact on the ability of cells to develop into spores, suggesting that FARAT is not important in differentiation. Growth curves for the deletion mutants show extended lag phase when cultures were started from an extended stationary phase, implying that FARAT plays role in cell growth, specifically recovery from stationary phase. Most interestingly chemotaxis assays in the deletion mutants demonstrate a significant impact on cells ability to efficiently move towards folate. These results suggest an important role for FARAT in intracellular signaling.

This work was supported by grants to D.B from the National Science Foundation(MCB-1244162) and Research Centers in Minority Institutions Program grants from the National Institute on Minority Health and Health Disparities(8 G12 MD007599) from the national Institutes of Health.

Immuno suppression/modulation of *Euphlyctis hexadactylus* (Ranidae: Raninae) by aquatic heavy metal pollution in an urban wetland in Sri Lanka

Jeran Halawath
University of East London

Mentor: David Guliano

Contamination of aquatic environments, particularly wetlands due to anthropogenic activities is increasingly evident globally. The urban wetlands are strongly influenced by long term discharge of untreated domestic and industrial wastewaters, storm water runoff, accidental spills and direct solid waste dumping due to rapid urbanization accompanied by weak urban planning (Mackintosh and Davis, ???). In Sri Lanka, urbanization has caused a steady degradation of urban wetlands during the past few decades (CEA, 1994), threatening the sustainability of the ecosystem services provided. Heavy metals which are disposed off through industrial and domestic wastes and effluents are amongst the most ubiquitous and vicious pollutants, posing severe threats to biota in the wetland ecosystems (Rai, 2009).

Owing to the intimate association with the wetland ecosystem through diverse lifestages and other vulnerable traits, amphibians appear to be particularly sensitive to xenobiotics, including heavy metals (Hopkins 2007). The exceptionally sensitive nature of amphibians has lead them to be treated as sentinel species which warn mankind of serious biodiversity issues that may culminate in the Earth's next mass extinction, if not dealt with appropriately. Recent decline and malformations reported in amphibians of many parts of the world were associated with aquatic pollution as it supports multiple stressors hypothesis where xenobiotic induced susceptibility to diseases and other agents evidently elucidate the mass decline reported in contaminated habitats (Hayes, 2010; Blaustein et al., 2011). Amphibian decline occurred due to disease outbreaks in contaminated habitats suggestive of xenobiotic driven immunosuppression, thus increasing their susceptibility to disease (Carey et al., 1999). Hence, there is an urgent need to study the extent to which xenobiotics indeed contribute to immunosuppression.

Stabilizing a reference laser for a modified Michelson interferometer

Madelyn Hoying
Duquesne University

Mentor: Theodore Corcovilos

Controlling the wavelength of a laser to high precision has a wide range of applications in spectroscopy and metrology. With a simple ratio, a stabilized laser can be used to determine the wavelength of an unknown laser to that same precision. The helium-neon reference laser for a modified Michelson interferometer was successfully stabilized using a digital proportional-integral-derivative (PID) controller implemented using an Arduino microcontroller. Our PID controller uses three variable parameters to provide a feedback mechanism that controls a heater inside the laser. By controlling the temperature of the laser, we can vary the length of the laser cavity and the wavelength of the emitted light. The polarization of the laser was measured as a proxy for the wavelength, and when the polarization changed, the PID controller changed the output of the heater to return the polarization and therefore the wavelength back to its original value.

Peptide Hydrogels for Neural Regeneration

Patricia Iglesias-Montoro
New Jersey Institute of Technology

Mentor: Vivek Kumar

Stroke is one of the most prevalent diseases worldwide and it affects millions of people annually. In 2010, stroke affected 33 million people globally and was the second-leading cause of death behind heart disease in 2013. Side effects of having a stroke include an abnormal gait, speech complications, as well as paralysis, all which greatly affect quality of life. Stroke and traumatic brain injury therapies on the market fall short when treating the disabilities resulting from the brain damage that drastically reduces independent daily living. There is currently no FDA approved revascularization for micro-vessels, a limitation this peptide hydrogel has the potential to overcome.

Our goal is to stimulate neural proliferation using multi-domain peptides (MDPs). Our ependymin peptide mimic conjugated to our peptide hydrogel can be injected to damaged areas of the brain which do to our MDP's biocompatible, slow degrading, injectable qualities make it a promising choice for developing material-based drugs for enhanced neural regeneration post-stroke and post-injury. This research aims to fuse the gap between stroke management and post-stroke/brain injury recovery in an attempt to restore bodily activities to victims with ischemic or impact induced brain damage.

We have synthesized our peptide, SLnc, done characterization tests such as mass spectrometry to identify impurities, infrared spectroscopy to determine anti-parallel beta sheet formation, rheology to test mechanical properties, and SEM to show the peptide's nanofibrous matrix structure. *In vitro* experiments to determine the neuroprotective potential of SLnc was tested by inducing neurotoxicity in rat cortical neurons using HIV-tat. *In vivo* experiments include subcutaneous implants in rats for biocompatibility and application of the peptide in an ischemic stroke model and traumatic brain injury model. We expect to see increased neuronal survival after brain injury due to our multi-domain peptide's potential as a potent drug delivery vehicle.

Acknowledgement: This work was funded in part by an Undergraduate Research and Innovation Phase II seed grant as well as by an NJIT Provost Fellowship.

Carlos Jeronimo
CUNY Lehman College

Mentor: Mary Baylies

Alveolar Rhabdomyosarcoma (ARMS) is a soft tissue sarcoma cancer that is made up of cells that express skeletal muscles genes. ARMS uniquely associates with chromosomal translocations t(2;13) (q35;q14) and t(1;13) (p36;q14), leading to the production of PAX3::FOXO1A or PAX7::FOXO1A fusion oncoproteins. The outcome for those ARMS patients with metastatic or recurrent disease remains dismal with standard treatments (<20% 5-year survival rate). Moreover, there have been no significant changes in treatments within the last thirty years and no precision treatments exist. The Baylies lab is using a *Drosophila* model of ARMS to identify novel therapeutic agents and treatment regimes. *Drosophila* is an appropriate *in vivo* model since expression of hPAX3::FOXO1 or hPAX7::FOXO in the fly mesoderm generates metastatic cells that invade nearby tissues in the larva, similar to what is seen in human patients. These transformed and invasive cells originate directly from dedifferentiating hPAX7::FOXO1 expressing muscles. Honokiol and IKK-16, two drugs that target the NfKB pathway, have been shown in the Baylies lab to prevent movement of the metastatic cells in flies and human tumor cells in culture. The purpose of these experiments is to develop a *Drosophila* high throughput screening methodology to measure the effects these experimental drugs. Previously, the lab had been dissecting larva tissues to measure metastatic behaviors: however, this is very time consuming and inefficient for the testing of these drugs. We designed a climbing assay to investigate whether pupation height, as a measure of larval muscle function, could be used as an effective screening assay. Honokiol and IKK-16 were tested at different concentrations. The heights that the treated larvae climbed were compared to those heights climbed by control, non treated PAX7::FOXO1 larvae. We found that pupation height increased in drug treated larvae when compared to non treated larvae. These data suggest that our assay works effectively and corroborates our previous data with these two drugs. Nevertheless, to confirm the assay's robustness, additional testing of other FDA approved drugs isolated with the lower throughput assay will be required. The ultimate goal is to develop therapeutic solutions for individuals with this disease by identifying novel drugs and translating them to the clinic.

Characterization of BMH-21 for Potential Use as Chemotherapy Treatment for Ovarian Cancer

Amber Jones
Hampton University

Mentor: Robert Cornelison

In women, ovarian cancer is the fifth leading cause of death in cancer related incidents, and it is the highest ranking cause of death of all the gynecological malignancies. Cancerous cells have a variety of mechanisms that promote their proliferation and survival. One of the major factors promoting cancer cell survival is their ability to develop chemoresistance against the standard chemotherapy treatments. The cytotoxic drug, BMH-21, is a small molecule DNA intercalator that inhibits RNA polymerase 1 (Pol 1) transcription by binding to ribosomal DNA. The drug is known for having an affinity for targeting rapidly dividing cells, like these chemoresistant cancer cells. We hypothesize that in the presence of BMH-21, ovarian cancer cells' ability to survive and proliferate would be significantly compromised. In order to try and characterize what effect BMH-21 has on cancerous cell lines, we tested the drug on the human derived ovarian cancer cell lines: OVSAHO, SKOV3-Parent, and SKOV3-TR. MTT assays were used to determine cell viability in response to varying doses of BMH-21. Immunofluorescence microscopy was then used to visualize the cellular effects induced by BMH-21. One of the main targets we were attempting to visualize was RRN3, which is a gene that promotes the efficient initiation of transcription by RNA Pol 1. In both the MTT assays and the immunofluorescence microscopy, BMH-21 had an adverse effect on the cells overall survival. With increasing doses, it significantly reduced the viability of all of the cell lines tested, and the microscopy showed fragmentation and degradation of the nucleoli, which is where RRN3 localizes. In order to further confirm this data, supplemental experiments are to be done testing the effects of BMH-21 on other ovarian cancer cell lines, as well as quantification of protein levels of RRN3 to ensure that an actual change in amount is occurring, rather than just a visual assumption. By better understanding BMH-21 and its role, it could potentially be used as a new novel chemotherapy option for treatment of ovarian cancer.

INCORPORATION OF T CELL RECEPTOR ALPHA LOCUS CONTROL REGION INTO LENTIVIRAL VECTORS FOR GENE THERAPY

Nailya Khalizova
CUNY Hunter College

Mentor: Benjamin Ortiz

Chimeric antigen receptor (CAR) T-cell gene therapy has emerged as a novel treatment for cancer and other diseases. However, this treatment is not without its limitations; the lack of control over the therapeutic gene bearing vector's integration site makes it susceptible to silencing from the surrounding chromatin environment. Additionally, the lack of a renewable source of T cells carrying the therapeutic gene can require multiple transfusions for the patient. Gene regulatory elements, such as Locus Control Regions (LCRs) may address some of these limitations by providing predictable spatiotemporally controlled therapeutic expression from a vector, regardless of the vector's site of integration. In order to translate our basic findings of LCR properties to gene therapy, we aim to incorporate LCR driven transgenes into lentiviral gene therapy vectors. We study the T-Cell Receptor alpha (TCR α) LCR, which as it exists endogenously is too large to fit into a lentiviral vector. Therefore, 'mini-LCRs' containing only characterized subregions of the TCR α LCR will be engineered into lentiviral vectors for therapeutic gene delivery. Here, we describe the construction of these viral vectors, and their use in the production of lentivirus-transduced mouse and human cell lines. Ultimately, we will utilize the viruses that are generated to transduce embryonic stem cells (ESC) and will analyze for mini-LCR-driven reporter gene expression during development of those ESC into T cells.

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Maryanne Kihui
Bryn Mawr College

Mentor: Varsha Gandhi

Ibrutinib is a highly potent target therapy that is used in the treatment of Chronic Lymphocytic Leukemia (CLL). It targets the B-Cell Receptor (BCR) signaling pathway by covalently binding to one of its key signaling molecules, Bruton's Tyrosine Kinase (BTK). The BCR pathway is essential for the growth and proliferation of B-lymphocytes. Usually, this pathway is implicated in CLL manifestation. Thus, the disruption of the BCR pathway, through the irreversible covalent inhibition of BTK with Ibrutinib, turns out to be very effective in preventing the growth of B- Lymphocytes and treating CLL. Even though Ibrutinib is very potent in CLL treatment, its mechanism of action is still not well understood. This, alongside other studies such as those focusing on resistance of CLL to Ibrutinib, highlight the need of an invitro model.

The purpose of this study is to establish an in-vitro model for CLL cells treated with Ibrutinib. To establish the suitability of the in-vitro model, we investigate whether the changes observed in the clinical trials of Ibrutinib are maintained in an in-vitro model using MEC-1 cells incubated with two doses of Ibrutinib, that is: 0.5 μ M and 1.0 μ M.

Data collected show a modest decrease of ibrutinib on cell growth and cell size. The data also show a significant decrease on the synthesis of macromolecules such as DNA, RNA and proteins. Further analysis on specific proteins' levels show a decline in pBTK, total BTK and Aminoacylase-1. Among the Bcl-2 family proteins, Bcl-2 and Bim proteins increased while Mcl-1 proteins decreased. An apoptosis assay confirms that Ibrutinib does not cause abnormal cell-death.

The time-frame in which most of the experiments are performed, (48hrs), is short in comparison to the 4week time-frame in the clinical analysis. Thus, the observations made could be limited to time. Nevertheless, most of the observed changes in Mec-1 cells are in concert with the observations made during therapy. Thus, it was concluded that Mec-1 cell line provides a relatively good model for studies related to CLL.

The Effect of Decreased Membrane Fluidity on the Efficacy of the Antimicrobial Peptide Maximin 3

Sarah Ann King
Purchase College

Mentor: Elizabeth.R Middleton

Antibiotic resistant microbes have become a serious problem for modern society due to the threat that super bugs and untreatable infection pose to individuals and agriculture alike. In recent years, a tentative solution to antibiotic resistant microbes has been identified in the form of antimicrobial peptides, or AMPs. Antibiotic molecules and modulators of the immune system, AMPs function via charge driven mechanisms which lack specificity and therefore not only effect microbes that have developed an immunity to some traditional antibiotics, but also may also remain effective for a longer period due to their smaller potential to act as a selective pressure for resistance. This report describes an assessment of the efficacy of the AMP Maximin 3 to induce leakage in man made vesicles of varying levels of lipid tail saturation. There is a strong correlation between degree of unsaturation in tails and leakage induced by Maximin 3; the peptide most effectively induces leakage in a target membrane that is moderately saturated. The current data also appears to suggest that interacting with saturated membranes may elicit changes in the mechanistic behavior of the peptide that are not observed in its reactions with less saturated membranes.

DISSECTING THE ROLE OF THE COMMENSAL MICROBIOTA IN INTESTINAL T CELL FATE

Suppawat Kongthong
Columbia University (Work was done at Rockefeller University)

Mentor: Angelina Bilate

The gut mucosa is constantly exposed to a large number of various antigens – both harmless and pathogenic. As a result, maintaining a balance between pro- and anti-inflammatory responses is of key importance. Two subpopulations of intestinal lymphocytes, peripheral regulatory T cells (pTregs) and intraepithelial CD4⁺ T cells (CD4-IELs), play a role in suppressing immune responses and maintaining intestinal homeostasis. Conversion and expansion of these two subpopulations can be affected by the intestinal microbiota through antigen recognition via the T cell receptor (TCR). To investigate how microbiota influence lymphocyte fate and the TCR repertoire, we performed a single-cell analysis of the TCR $\alpha\beta$ pairs on the Illumina MiSeq platform on intestinal lymphocytes from inducible fate-mapping reporter mice that enabled us to single-cell sort pTregs, Tregs that converted into CD4-IELs, and CD4-IELs. Our preliminary data show the TCR repertoire of pTregs and CD4-IELs in the intraepithelial compartment is restricted to a few expanded clones whereas pTregs in the mesenteric lymph nodes are as diverse (polyclonal) as expected. Our data suggest the intraepithelial pTregs and CD4-IELs are being activated by a restricted set of antigens. To screen for commensal bacteria that these TCRs recognize, we used an *in vitro* antigen presentation assay consisted of NFAT-GFP T cell hybridomas reconstituted with selected TCRs. When the hybridomas become activated, they express GFP under NFAT control. The next step is to find which bacterial antigens these expanded TCRs recognize. This can shed light on how immunity against commensals is achieved while keeping intestinal immune homeostasis.

Research was conducted during the summer of 2017 and was sponsored by The Rockefeller University as part of their Summer Undergraduate Research Fellowship (SURF) program.

CHRONIC ALCOHOL EXPOSURE AFFECTS GENERALIZATION AND EXTINCTION OF FEAR MEMORY

Serena Lee
Vassar College

Mentor: Hadley Bergstrom

Post Traumatic Stress Disorder (PTSD), one of the most prevalent psychiatric disorders in the world, is highly comorbid with alcohol use disorder. How alcohol might impact PTSD symptoms is currently unknown. We used auditory fear conditioning and a chronic ethanol (EtOH) exposure paradigm to investigate whether repeated exposure to alcohol modified fear memory generalization and extinction in C57BL/6 mice. After auditory fear conditioning, mice were given daily injections (intraperitoneal) of saline or ethanol (EtOH; 2.5 g/kg) over five days, which were then followed by a 3-day drug-free period. The mice then received 50 exposures to either the 5-kHz target stimulus tone they were conditioned with or a 3-kHz non-target stimulus they were previously naive to. EtOH significantly increased fear response (freezing) to the 3-kHz tone to the same magnitude of response as the 5 kHz control group, indicating that alcohol increased cued fear memory generalization. In addition, the alcohol 3 kHz group extinguished their generalized fear memory at a much faster rate than all other groups after the first 10 tones, demonstrating that alcohol also affected extinction rate. The increase in generalization due to alcohol may explain alcohol's role in exacerbating PTSD symptoms. However, these effects were not seen when the mice were tested 12 days after extinction. The reversibility of alcohol's effect on generalization over a larger period of time suggests that decreasing alcohol consumption may alleviate PTSD symptoms. To further examine these findings we will use designer receptors exclusively activated by designer drugs (DREADDs) to test a causal role for alcohol-induced neuroadaptations in specific brain regions in fear memory generalization and extinction.

Acknowledgements: This work was supported by the Undergraduate Research Summer Institute at Vassar College.

IDENTIFYING AN NO DETOXIFICATION REGULATOR USING S-NITROSYLATION IN *VIBRIO CHOLERAE*

Zachariah Lee
University of Pennsylvania

Mentor: Jun (Jay) Zhu

Cholera, a disease characterized by severe diarrhea and dehydration, is estimated to affect hundreds of thousands of people each year. In order to prevent the pathogenesis of *Vibrio cholerae*, the causative agent of cholera, the host immune system creates a system of stresses to prevent the pathogen from colonizing, such as oxidative stress and the release of nitric oxide (NO). Although the major genes responsible for *Vibrio cholerae*'s response to these stresses during colonization is known, gaps exist in the knowledge of their regulation that could provide new insight into bacterial adaptation to host environments.

Our research focuses on the identification of a regulator for the major NO detoxification protein possessed by *Vibrio cholerae*, HmpA, as well as characterizing the effects of this regulation on *Vibrio cholerae* pathogenesis. This study could advance our knowledge of bacterial stress responses employed by pathogens to overcome host stresses.

We demonstrate that AphB, a virulence gene activator, functions as a repressor of *hmpA*. This repression has shown to be important for survival when *Vibrio cholerae* are exposed to low or intermediate nitrate concentrations, while repression is not required at high nitrate concentrations. We also show that *hmpA* repression by AphB promotes survival during oxidative stress. Finally, using an adult mouse model, we determine that *hmpA* repression by AphB is necessary for stable colonization. Our data demonstrates that regulation of the NO detoxification pathway is critical for *Vibrio cholerae*'s response to nitrogen sources, oxidative stress, and host colonization.

This work was partially supported by University of Pennsylvania's Center for Undergraduate Research and Fellowships and the Ruth Marcus Kanter College Alumni Society Undergraduate Research Grant.

Title: Structure-Function Analysis of Antimicrobial Pore-Forming Protein Apolipoprotein L-1

Angelina Limon
CUNY Hunter College

Mentor: Jayne Raper

African Trypanosomes are protozoan parasites that cause sleeping sickness in primates and mammals. A subfraction of high density lipoproteins (HDL or “good cholesterol”), found in select primates, termed Trypanosome lytic factor (TLF) provides innate immunity from the species *Trypanosoma brucei brucei*. The parasites take up TLF by receptor-mediated endocytosis, allowing Apolipoprotein L-1 (APOL1), a protein component of TLF, to insert into the endosomal membrane and form a pore upon recycling to the plasma membrane. This pore disrupts osmotic balance within the parasite, resulting in water influx and eventual lysis. It has been shown that the pore formed by APOL1 is cation selective, depends on acidic pH for insertion into a lipid membrane, and subsequent neutralization of pH for opening of the pore. However, the protein’s structure, mechanism of insertion, and pore formation are unknown. Our hypothesis is that there are two pore-forming domains and one transmembrane domain in APOL1. This study outlines a preliminary investigation of APOL1 structure via computational methods. Using several servers to predict secondary structure and topology, such as TOPCONS, PHYRE2, and PSIPRED, we were able to create a consensus model of secondary structure and transmembrane domains. We also identified several template candidates for tertiary structure modeling, based on homology or fold recognition. In the future, we will test pore-forming domains identified computationally through in-vitro methods, such as residue substitution. We hope to use these models to elucidate the mechanism of parasitic membrane interaction of APOL1, such as conformational changes dependent on pH and potential oligomerization.

CRISPR/Cas9 mutagenesis invalidates a genetic target of clinical trials in cancer

Ann Lin
Stony Brook University

Mentor: Jason Sheltzer

The Maternal Embryonic Leucine Zipper Kinase (MELK) has been described as a genetic dependency in several cancer types, most notably in the highly-aggressive basal subtype of breast cancer. MELK inhibition through the use of either RNAi or small-molecule approaches has been reported to block the growth of cancer cells from these subtypes. Based on these results, the MELK inhibitor OTS167 is currently being tested as a novel chemotherapy agent in multiple clinical trials. Here, however, we report that mutagenizing MELK with CRISPR/Cas9 has no detectable effect on the fitness of basal breast cancer cell lines or cell lines from several other cancer types. Additionally, cells that lack wild-type MELK remain sensitive to OTS167, suggesting that this drug blocks proliferation through an off-target mechanism. In total, our results undermine the rationale for a series of current clinical trials based on MELK inhibition and provide an experimental approach for the use of CRISPR/Cas9 in preclinical target validation that can be broadly applied.

REGULATION OF THE CLASS II TRANSACTIVATOR BY 14-3-3 β

Hagerah Malik
Sarah Lawrence College

Mentor: Drew Cressman

The Class II Transactivator (CIITA) is an important transcription factor of Major Histocompatibility II (MHC II) genes. Defects in CIITA, as occur in Bare Lymphocyte Syndrome, lead to the loss of MHC II expression and subsequent inability to activate an immune response. Accordingly, CIITA has often been termed the master switch of the immune system. Its underactivation results in immunodeficiencies whereas its overactivation may lead to autoimmune disorders. Therefore, the activity of CIITA must be tightly regulated through multiple post translational modifications, including phosphorylation and ubiquitination, to ensure proper levels of activation.

We identified a sequence within CIITA from amino acids 283 to 289 that matches a consensus 14-3-3 binding site motif: Rx₃xpTxP and decided to explore the effects of the 14-3-3 β isoform on CIITA. We have shown through IPs that CIITA interacts with 14-3-3 β and that this interaction leads to the proteasome mediated degradation of CIITA. Mutants of the binding site are more stable than wild type CIITA, indicating that they are less prone to degradation. Additionally, phosphorylation at the threonine residue in this site is necessary for the interaction and effects of 14-3-3 β upon CIITA.

We have shown that 14-3-3 β leads to the degradation of CIITA by scaffolding it to a cellular factor and altering its stability. Much work remains to be done to determine what this cellular factor is and the changes in transactivation potential of CIITA as a result of 14-3-3 β interaction.

Purification and crystallization of the extracellular loop domains of TRPP and PKD ion channel proteins

Hannah Martin
St. John's University

Mentor: Yong Yu

Abstract: Members of the transient receptor potential (TRP) protein family of ion channels are essential for various cell signaling throughout the body. Proteins in the polycystin subfamily of TRP channels (TRPP) form homomeric tetramer by themselves and receptor-ion channel complexes with their polycystin kidney disease (PKD) protein partners in a three TRPP plus one PKD stoichiometry. TRPP2 forms a flow-sensing complex in primary cilia with its PKD partners that determines left-right asymmetry in the embryonic node and proper morphology of kidney tubules. Mutations in TRPP2 and PKD1 cause autosomal dominant polycystic kidney disease (ADPKD). TRPP3, with its binding partner PKD1L3, is a sour taste receptor candidate found in taste cells in the tongue. TRPP proteins assemble with PKD proteins through their C-terminal coil-coil domains and the extracellular loops. Since a significant portion of pathogenic mutations are located in these loops, they likely play crucial roles in subunit assembly and ion conductivity. However, the exact function of the loops and the mechanism by which the mutations lead to ADPKD are unknown. Structural analysis of these extracellular loops will greatly enhance our understanding of the molecular mechanism of the assembly of TRPP/PKD complexes and the function of these complexes in normal and disease conditions. In the current project, we are working on solving the structures of the complexes formed between the extracellular loops of TRPP2 and PKD1, as well as from TRPP3 and PKD1L3. Particularly, we cloned the fragments of the extracellular loops of the PKD1, PKD1L3, TRPP2, and TRPP3 proteins based on secondary structure prediction and functional importance, and co-expressed and purified the binding partners from *E. coli*. Our goal is to co-crystallize them and determine their structures. This study will shed light on the interaction and assembly of the extracellular loops of TRPP proteins, their relevance to the channel function, and the mechanism by which pathogenic mutations alter channel behavior.

TIME CORRELATED SINGLE PHOTON COUNTING IN MICE *IN VIVO* REVEALS DIFFERENTIAL ACTIVATION OF D1-EXPRESSING MEDIUM SPINY NEURONS IN THE DORSAL STRIATUM DURING A BEHAVIORAL TASK OF ANXIETY.

Rachel Mikofsky
Columbia University Medical Center

Mentor: David Sulzer

Anxiety disorders are the most common class of mental illness worldwide, costing more than 40 billion dollars in the US alone. Human magnetic resonance imaging studies implicate dysfunction in the striatum and increased striatal volume in the pathology of anxiety.

The dorsal striatum is largely comprised of spiny projection neurons (SPNs) that project through the basal ganglia circuitry in two major pathways, termed the direct and indirect paths. Direct pathway SPNs express dopamine D1 receptors while indirect pathway SPNs (iSPNs) express dopamine D2 receptors. The activity of the dSPNs and iSPNs during tasks of anxiety have never been examined *in vivo*. We hypothesized that these neurons may show a differential level of activation depending on the anxiety state of the mouse.

We used time correlated single photon counting (TCSPC) to record neuronal activity in the dSPNs and iSPNs during the elevated plus maze task of anxiety. The elevated plus maze consists of two open and two closed arms: time spent in the closed arms is thought to be associated with a lower anxiety state. D1-Cre (direct path) and A2A-Cre (indirect path) mice were injected with AAV9.Flex.GCaMP6f virus. Imaging fibers were implanted in the dorsal lateral striatum. Mice were recorded with a high speed video camera framelocked to the TCSPC sampling rate. This enabled correlation of anxiety related behaviors with activity of specific SPN pathways. The D2 expressing iSPNs showed a higher rate of firing in the closed arms of the maze (Two way ANOVA $P < .001$). D1 expressing dSPNs showed a trend towards increased firing in the closed arms, but this did not reach significance. ($P > .05$). Utilizing the high speed video analysis, we concluded that the increased calcium transients in the dSPNs correlated with behaviors associated with a lower anxiety state. We plan to perform future trials with benzodiazepines.

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IDENTIFYING THE NOVA MUTATIONS RESPONSIBLE FOR THE SPLICING FAILURE OF Z⁺ AGRIN

Jovan Mirkovic
St. John's University

Mentor: Mateo Ruggiu

The NOVA family of RNA-binding proteins consists of neuron-specific splicing regulators that are targeted in an autoimmune neurodegenerative motor disorder. They are sequence-specific splicing factors, with a modular structure consisting of three KH domains that mediate protein-RNA interactions.

One of the targets of NOVA is a neuron-specific isoform of the ubiquitously expressed gene AGRIN. This splice variant, termed Z⁺ AGRIN, is secreted by neurons and is found at the synaptic cleft at the neuromuscular junction (NMJ). By inducing the clustering of acetylcholine receptors (AChR) on the postsynaptic membrane, Z⁺ AGRIN promotes the formation, maintenance, and development of the NMJ. The Z exons are also of particular interest as they are potential mutation sites in congenital neuromuscular diseases.

The study of NOVA function in vertebrates is complicated by the presence of two NOVA genes termed NOVA1 and NOVA2. The tunicate *Ciona robusta* is a marine invertebrate that is the closest living relative to vertebrates. Interestingly, *Ciona* has only one Nova gene. We used the NOVA-AGRIN paradigm to study the evolution of protein-RNA interactions in the brain. We cloned and characterized Nova proteins from both mouse and *Ciona*. We also generated Agrin minigenes from both mouse and *Ciona* that recapitulate the action of Nova in a tissue culture cell system. Here, we identify the specific amino acid sequences that mediate Nova-dependent splicing of Z⁺ agrin in both mouse and tunicates. Our work provides evidence of co-evolution of a splicing factor –Nova – and the target it regulates –Agrin.

Effect of 20% Fructose consumption on NOX4 Expression in Rat Proximal Tubules

Aaron Morris
Morehouse College

Mentor: Jeffery Garvin

Hypertension is the leading cause of loss of health worldwide. In America, roughly 35% of people are hypertensive and half of them display sensitivity to salt. Angiotensin II (Ang II) is an important regulator of blood pressure, in part by controlling salt reabsorption in the kidney. Upon increases in salt intake Ang II levels drop to allow renal excretion of salt. Malfunction in Ang II signaling is associated with elevated oxidative stress, salt retention and hypertension. The effects of Ang II on the proximal tubules are mediated in part through the production of superoxide from NOX4 which leads to increased sodium transport. NOX4 is the main form of NADPH Oxidase found in the proximal tubules of the kidneys where Na reabsorption occurs in the renal system. An increase in proximal tubule transport contributes to an increase in blood pressure. Fructose increases renal sensitivity to Ang II. Therefore, the increased sensitivity to Ang II displayed in fructose fed animals may be increasing superoxide production by NOX4 and thereby Na transport. We hypothesize that a 20% fructose diet will lead to the stimulation of NOX4 leading to increased sodium transport in the proximal tubules. To test our hypothesis, proximal tubules suspensions were prepared from rats on either a normal diet or a 20% fructose diet. Protein samples were put through a gel electrophoresis, and then a blotted for NOX4. We found that NOX4 expression in tubules from rats in fructose was ~61% greater than in controls. Our findings indicate that a 20% fructose diet heightens NOX4 expression in proximal tubules. This likely elevates superoxide production leading to an increase in salt absorption and contributing to hypertension.

A bursting assay for Giant Unilamellar Vesicles containing gangliosides

Fernanda Murillo
Adelphi University

Mentor: Matthew Wright

Cell membranes are composed of phospholipid bilayers. Phospholipids are amphiphiles, which have a hydrophilic head group and two hydrophobic fatty acid chains. When they are in water, they self-assemble so that the head groups align with each other and interact with the water, shielding the tails from the water so that they only interact with each other, forming the phospholipid bilayer. Other amphiphilic molecules can also be incorporated into the bilayer, such as cholesterol and proteins. The molecules of a membrane are held together by the hydrophobic effect, so the lipids and proteins are free to flow and diffuse along the membrane. We know that some cell types are constantly exposed to flow, that these cells sense flow and that flow is essential for normal function in these cells. We want to study the effect that flow has on membrane proteins. Our lab creates phospholipid bilayers in the form of Giant Unilamellar Vesicles (GUV), which are spheres of membrane, and grows them from lipids through electroformation, in order to investigate the effect of flow on membrane proteins. If we dilute our vesicles in a saline solution, the vesicles begin to sink to the bottom of the solution and rest on the glass, since they are more dense than the solution. The vesicles rupture on the surface and form a flat sheet (splat), which we refer to as a supported lipid bilayer (SLB). We can apply a flow to the SLB to see its effect on membrane proteins. The particular system we studied is a lipid (ganglioside) and protein, Cholera Toxin subunit B (CTB). We use ganglioside GM1 as our lipid because it has a large head group that sticks out of the membrane and acts as a receptor for the CTB protein. We can use this system to apply flow and see the effect of flow on membrane proteins, but we need to have a better understanding of how the vesicles break. This work studied how to optimize the breaking conditions of the GUV to use the SLB to study the effect of flow, since GUV breaking kinetics depend on many factors, such as lipid change and composition, surface treatment and buffer/salt concentration. In order to characterize splatting conditions, I took a movie of vesicles splatting on a glass coverslip and made a Python program to detect the vesicles in each frame of the movie to observe the properties of the splatting. The program uses the HoughCircles function in python to detect circles (vesicles) in an image. Vesicles must be diluted the appropriate amount in order to get a clear image of well-separated vesicles. The vesicles are electroformed in sucrose so you can see the phase contrast, since phase contrast shows refractive index differences. Lastly I added a high salt buffer to dilute the vesicles in order for them to splat at a reasonable rate and then took images for the movie. As time passes, there should be a decreasing vesicle count, since they are splatting and when they splat, the sucrose disperses and you can't see the vesicle in focus, and the vesicle that was there will no longer be detected. I ran each frame through the program and put it together to get an output movie of detected vesicles and plot of the vesicle count through time. I found that as expected, there was a decrease in vesicle count through time, and unexpectedly, cleaning the slides with plasma did not make the vesicles splat faster.

Suha Na Javeed
City College of New York

Mentor: Michelle Juarez

Injuries occur as the consequence of various events and permit the occurrence of infections via pathogens. Meticulous regulation is necessary to execute simultaneous processes of activating wound repair and preventing infection. The inability to properly activate wound repair can lead to excessive cellular production and scarring. An insufficient immune response can cause harm to a greater degree to the tissue. The importance of research in wound repair cannot be emphasized enough because a patient can suffer due to a variety of wounds, ranging from surgical damage to chronic conditions. *Drosophila* contains a stable genetic system that serves as a tool to study processes that involve wound repair. The model organism, *Drosophila*, can be used to improve genetic conditions for tissue restoration. In the terminal stage of *Drosophila* embryonic development, the epidermis permits wound response analysis through its *in vivo* system in a protocol used by Dr. Juarez. The objective of these studies serves to understand the processes that conduct tissue damage control and limit infection development. The central hypothesis of this experiment is that wound repair and survival likelihood can be enhanced by limiting damage signals to affected regions only. If the spread of the damage to tissue can be limited to the site of injury, then the process of wound repair can be accelerated. The validity of this hypothesis can be tested by testing two specific aims. The first aim is to understand the cellular process that limits genetic damage to the site of injury. The second aim is to determine the role of each wound response activator and inhibitor during wound repair. The applicant will study *Drosophila* genetics and plans to generate a set of evolutionarily conserved genes that control the localization of a transcriptional response to an epidermal tear. In response to the first aim, comprehension of the process in which DNase II genes, which can serve as agents of DNA degradation, are transcriptionally controlled at the site of injury will facilitate the development of molecular mechanisms that would limit the spread of injury. To accomplish the second aim, newly built transgenes, which are prepared through the transfer of genetic materials into an organism, will be used to evaluate the functions of wound activator and wound inhibitor genes after injury and microinjection. Wound activators, i. e. plasminogen activators, participate in activities related to tissue remodeling. Wound inhibitors, such as plasminogen activator inhibitor-1 (PAI-1), restrict tissue degradative activity. Overall, wound activators and inhibitors collaboratively exhibit regulatory functions which balance reparative and destructive cellular activity. Molecular cloning techniques and fluorescence microscopy will be used to achieve these two aims. The usage of a transcriptional activation reporter provides a new methodology to unveil unknown constituents of the process that stops the spread of tissue damage and influences other aspects of wound repair. The findings from the model *Drosophila* are powerful because the findings from experiments with *Drosophila* can be translated into useful information for mammalian organisms.

Acute Lymphoblastic Leukemia and the contribution of Pax5 to genomic stability

Rachel Nation
The City College of New York

Mentor: Patricia Cortes

The RAG Recombinase is an enzyme that plays an essential role in lymphocyte development. Our preliminary data suggests that Pax5, in addition to working as a transcription factor during B cell development, functions as a regulator of the RAG recombinase allowing for controlled V(D)J recombination. Thus, when mutated, Pax5 would induce the deregulated activity of the RAG recombinase. Published work demonstrates that in patients with acute lymphocytic leukemia Pax5 mutations are associated with the deletion of CDKN2A/B. We propose that the expression of Pax5 mutant induces the deregulated increase in RAG activity, causing CDKN2A/B deletion. The proposed hypothesis will be investigated using a human proB cell line, in which we will first confirm both regulation of the RAG recombinase by Pax5 and the effect of Pax5 mutants in controlling RAG function using exogenous recombination substrates. After the regulatory role of Pax5 on RAG functions is established, increased deletion of CDKN2A/B in presence of Pax5 mutant will be explored. The methodology to introduce wild type and mutant DNA into the human proB cell line was established with an optimized transfection efficiency of 25%. Preliminary experiments were also performed to set up conditions for the analysis of RAG function. Together our finding will contribute to explain the phenotype of genomic instability observed in patients with ALL that carry Pax5 mutations.

THE ROLE OF TGF- β /SMAD3 SIGNALLING PATHWAY IN THE DIFFERENTIATION OF PREADIPOCYTE CELL LINES

Chi Nguyen
Colgate University-NIDDK/NIH

Mentor: Youngjae Bahn

The rise of obesity and type II diabetes is posing serious challenges to global public health (Zimmet 2017). Obesity is characterized by the accumulation of white adipose tissues (WAT) which serves primarily as energy. On the other hand, brown adipose tissues (BAT) specialize in energy expenditure, creating an energy homeostasis counterbalance (Gesta et al 2007). White and brown adipocytes share a common developmental origin from mesenchymal stem cells, but WAT differentiation is driven by the expression of PPAR γ and C/EBPs while the expression of PRDM16 in Myf5+ precursory cells induces brown fat differentiation (Park et al 2008).

In this study, we investigate the role of TGF- β /Smad3 signalling on brown-fat specific gene markers PRDM16 in HIB-1B and 3T3-L1 preadipocytes. TGF β pathway was enhanced through drug treatment with TGF- β 1, or suppressed through SB-431542 drug or Smad3 KD/KO cell lines. HIB-1B cells were treated with TGF- β 1 and harvested at 4 different time points. In HIB-1B cells treated with TGF- β 1 for 48 hours, PRDM16 mRNA significantly increased compared to non-treatment control. Moreover, mRNA level of PRDM16 increased over time, peaking at 8 hours before decreasing to the baseline level. mRNA level of Smad3-target genes Smad7 and Serp1 was lower in non-treated cells than in treated cells at 48 hours. Meanwhile, the level of phosphorylated Smad3 in treated condition was higher. Interestingly, the levels of protein expression for PRDM16 were similar between the two conditions, despite difference in mRNA level, suggesting another mechanism regulating PRDM16 post-transcription. In 3T3-L1 Smad3 KD adipocytes, PRDM16 mRNA level significantly increased compared to the wildtype. Overall, the study establishes the negative effect of TGF- β /Smad3 pathway on PRDM16 mRNA expression in both HIB-1B and 3T3-L1 preadipocyte cell lines. Future direction aims at reproducing the experiment under optimized differentiation conditions and observe the physiological development of the two cell lines *in vitro*.

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Entrainment of Visual Steady State Responses Is Modulated by Global Spatial Structure

Thomas Nguyen
Binghamton University

Mentor: Vladimir Miskovic

Abstract: Periodic modulation of stimulus luminance or contrast elicits visual steady state responses (VSSRs) that are often used in cognitive neuroscience research studies. These VSSR responses originate largely from striate and extrastriate regions when the signals are recorded at the scalp surface. A wide variety of visual inputs have been used to drive the VSSR, ranging from narrowband sinusoidal gratings to broadband, natural scenes. Here, we investigated differences in VSSR entrainment strength while varying the global spatial structure of the visual inputs. Specifically, we recorded high-density EEG in human participants while they viewed different categories of visual ensembles including visual spatial noise, sinusoidal gratings, and naturalistic scenes. We found greatest entrainment for visual inputs having second-order statistics ($1/f$ -2 spectral power drop-off) matching naturalistic inputs. Ongoing studies in our lab are examining the separate influences of image spectral amplitude and phase (higher-order statistics) on VSSRs. These studies will allow for a more comprehensive understanding how image statistics influence VSSR dynamics.

Poster Title: Early Life Stress and Sex-Specific Cognitive Impairments Comorbid With Stress-Related Mental Disorders

Luke Noreña
Brown University

Mentor: Kevin Bath

Early life stress (ELS) has been found to show similar cognitive impairment associated with many stress-related mental disorders such as major depressive disorder (MDD). The comorbid cognitive impairment found from early life stress and these stress-related mental disorders are suggestive of frontal lobe dysfunction, especially among females. The following study looks at the effects of early life stress on both male and female p65 ELS mice. The research looks at how it affects the development of different depressive-like behaviors and how it influences cognition in the frontal lobe. Data was collected from various behavioral and molecular assays including a sucrose preference paradigm, an attentional set shifting task, and qPCR analyses. The qPCR analyses showed significant decreases in parvalbumin (PV) and GAD67 gene expression in the orbitofrontal cortex but not the medial prefrontal cortex for female p65 mice possibly causing the difficulty of rule reversal learning in the attentional set shifting task. Future studies aim to look at more stress associated behaviors and pathologies in the frontal lobe, using immunohistochemistry and getting PV cell counts while also continuing to look at both male and female p35 ELS mice.

EXOSOMAL MIR-21 LIKELY MEDIATES HUMAN MESENCHYMAL STEM CELL PARACRINE EFFECTS ON HUMAN ENGINEERED CARDIAC TISSUE CONTRACTILE FUNCTION

Kasoorelope Oguntuyo
Harvard University

Mentor: Kevin Costa

Delivery of human mesenchymal stem cells (hMSCs) is an emerging therapy for heart disease. However, stem cell delivery benefits are generally modest and transient, warranting further investigation into the underlying hMSC-cardiac interactome. We recently demonstrated the exosomal fraction of the hMSC secretome to be largely responsible for increasing contractility and associated calcium handling gene expression in human engineered cardiac tissues (hECTs). However, the key exosomal cargo responsible for these effects remains largely unresolved. The aim of our study was to identify effects of lead candidate miR-21 exosomal cargo on our hECT three-dimensional in vitro contractility assay. After baseline contractile function testing, hECTs were treated with 30 picomoles of either miR-control or miR-21 mimic. hECTs were functionally assessed for five days post-treatment, and then snap-frozen for subsequent molecular characterization. Treating hECTs with miR-21 mimic recapitulated a wide range of effects observed with hMSC exosomes on hECT developed force and expression of calcium handling genes. More specifically, treating hECTs with miR-21, but not miR-control, statistically increased developed force five days post-treatment compared to baseline. These findings were corroborated by molecular characterization, as treating hECTs with miR-21 led to significantly increased mRNA levels of LTCC, SERCA2a, and b/a myosin-heavy chain ratio, while the BAX/BCL2 ratio significantly decreased. These findings suggest that exosomal miR-21 is a lead candidate responsible for hMSC paracrine mediated increase of myocardial contractility. Ongoing work involves the development of efficient cryostorage techniques for these cardioactive exosomes and their cargo.

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PODOCYTE-SPECIFIC OVEREXPRESSION OF KRUPPEL-LIKE FACTOR 15 ATTENUATES PODOCYTE INJURY

Jesse Pace
Stony Brook University

Mentor: Sandeep Mallipattu

Podocytes are terminally differentiated epithelial cells that play a critical role in the filtration barrier. Podocyte loss contributes to the development of several primary glomerulopathies including minimal change disease and focal segmental glomerulosclerosis. Kruppel like factor 15 (KLF15) is a kidney enriched zinc finger transcription factor that plays a central role in regulation of podocyte differentiation. Using murine models, we show that podocyte specific overexpression of KLF15 improved renal function in two different injury models: a transgene model of HIV associated nephropathy (Tg26) and chemical model of adriamycin induced nephropathy. KLF15 overexpression in Tg26 mice resulted in increased expression podocyte differentiation markers including nephrin, synaptopodin, and podocin, as measured through immunofluorescence staining and qPCR. Furthermore, KLF15 overexpression mice displayed reduced proteinuria and reduced expression of inflammatory and fibrotic markers compared to Tg26 mice. RNA sequencing identified 600 genes that were differentially expressed between the Tg26 and Tg26 with KLF15 overexpression, many of which have known roles in the cell cycle, FSGS and podocyte differentiation. However, majority of the differentially expressed genes lacked predicted KLF15 binding sites, suggesting that another transcriptional regulator may be involved. Interestingly, enrichment analysis identified differentially expressed Wilms Tumor 1 (WT1), a zinc finger transcription factor, as a predicted transcriptional regulator of several differentially expressed genes. Based on predicted KLF15 binding sites and published WT1 ChIPseq data, we suspect that KLF15 and WT1 act together to regulate podocyte differentiation. Our findings indicate that WT1 may act as a transcription factor directly regulated by KLF15 that is an essential mediator of the therapeutic effects of KLF15 overexpression in kidney injury.

Imari Parham
Xavier University of Louisiana

Mentor: Terry Watt

Lysine deacetylases (KDACs) are enzymes that modify proteins and they serve an important role in several major systems within the body. KDACs remove acetyl groups from lysines and complications with this function can contribute to several diseases such as asthma, diabetes, and some cancers. Mass spectrometry is being used to identify acetylation sites in cytoskeletal proteins (actin and tubulin). Purified proteins are digested by a protease into identifiable peptides, and the addition of an acetyl group to lysine is determined by changes of mass that correspond to acetate. The identification of these acetylation sites allows for us to have a basic understanding of the acetylation in cytoskeletal proteins. After the acetylation sites are established, we will manipulate KDACs using KDAC selective inhibitors and CRISPR genome editing to understand which KDACs are responsible for deacetylation of these proteins. We are currently performing genome editing with CRISPR to introduce mutations that will alter the behavior of KDACs produced in that cell. The cytoskeletal proteins will then be extracted from the cells and acetylated sites compared with proteins from unmodified cells to determine which sites each KDAC deacetylates.

DENTAL PULP REGENERATION USING NOVEL SELF-ASSEMBLING PEPTIDE SCAFFOLDS

Saloni Patel

NJIT

Mentor: Vivek Kumar

Dental pulp regeneration is a sought after alternative to root canal procedures and fillings which result in devitalized teeth. This project aims to create an injectable multidomain peptide (MDP) injectable hydrogel scaffold that promotes dental pulp regeneration and will create a revitalized tooth *in vivo*. The research works to develop a self-assembling peptide (SLD) which will encourage cell adhesion and promote dental pulp regeneration following extirpation of infected pulp. This peptide serves as a scaffold and a drug to promote growth following the pulppulpal extraction. Such a peptide and has the potential to work with growth factors in a hydrogel to promote dental pulp regeneration in the dental carie.post-extirpation of a carious lesion.

Scanning electron microscopy (SEM) and Infrared spectroscopy (IR) were performed on varying concentrations of peptide with either the calcium ions or ϵ -polylysine present. SEM, scanning electron microscopy, results not only showed regions of nanofibers formed within the gel but also guided the research project in seeing which sample of SLD had the optimal molecular structure. The IR, infrared spectroscopy, spectrum displayed the characteristic amide I band and antiparallel β -sheet formation. The IR results were used to show consistency among the different hydrogel samples. Further characterization was completed through running AFM and rheology on the hydrogel samples.

The hydrogel of SLD was tested in fibroblast 3T3 cells to test for biocompatibility of the peptide *in vitro*. The cells were stained using a live/dead assay after 48 hours to count the number of viable cells following the treatment and results expressed vitality following the stain examination.

Following the characterization and *in vitro* analysis of the peptide, procedures for an *in vivo* assay through a rat subcutaneous surgery were finalized and completed. The MDP was injected into a rat dorsal subcutaneous model. Histology was performed after one week to test for biocompatibility and the host response of the MDP.

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THE ROLE OF DOPAMINERGIC TERMINAL ACTIVITY IN NAc AND DMS DURING ADVERSIVE FEAR EXTINCTION

Katherine Pizano
Princeton University

Mentor: Ilana Witten

Dopamine (DA) neurons have long been implicated in encoding prediction errors in reward-based learning. However, the role of striatal DA neurons in aversive learning remains largely unknown. We hypothesize that DA terminal activity within the striatum, specifically the NAc and DMS, helps encode a teaching signal which enables extinction learning. To test this hypothesis, we use an aversive fear conditioning behavioral paradigm, along with calcium imaging, to record from striatal DA terminals during extinction. This allows us to analyze how these subpopulations of neurons correlate with conditioned response of freezing to aversive associations. Our preliminary findings suggest DA activity in the NAc and DMS during CS (tone) offset is, in fact, correlated with the aversive action (freezing) within the current trial. Moreover, a decrease in DMS terminal activity at the CS onset also correlates with freezing during extinction trials. Together, both these signals may help drive the conditioned aversive response by preventing extinction learning from occurring. Our next steps will include using optogenetics to manipulate and record from these DA terminals. This will provide causal evidence as to whether dopamine is involved in reinforcing aversive actions or updating the CS-US association. Understanding dopamine's role during extinction of aversive stimuli can help better our understanding of psychiatric disorders like PTSD, which show strong evidence for the involvement of the DA pathway.

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Derian Pugh
Xavier University of Louisiana

Mentor: Alison Goate

The microtubule - associated protein tau (MAPT) gene encodes for the protein Tau, which is abundant in the neurons of the central nervous system. Tau is responsible for stabilizing microtubules, which supply essential nutrients to the cell, and it participates in cell division. Tau can aggregate to form neurofibrillary tangles that ultimately end up disintegrating this vital transport system, resulting in cell death. Diseases that are associated with defective and aggregated Tau are referred to as Tauopathies. Exons 2, 3, and 10 of the MAPT gene are alternatively spliced in human brain. Altered splicing of these exons is hypothesized to be a contributing factor to the development of Tauopathy. In an effort to understand MAPT expression, splicing, and regulation in the human brain, we altered the expression of a candidate splicing factor, RBFOX1, to functionally validate its effect on the regulation of MAPT in vitro. We transfected HEK293T cells with iBac-MAPT, a construct that contains the entire MAPT gene. We evaluated the effect of RBFOX1 overexpression and knockdown on RBFOX1 and total MAPT expression by qRT-PCR and changes in MAPT exon 2 and exon 10 splicing by PCR. We validated the overexpression of RBFOX1 in vitro, but were not able to validate its knockdown due to very low endogenous expression in HEK293T cells. Further investigation and optimization of the iBacMAPT construct is required. We plan to test other candidate splicing factors in the future to better characterize and understand the regulation of alternative splicing, and how this may be associated with Tauopathies such as Alzheimer's disease (AD), frontotemporal dementia (FTD), and progressive supranuclear palsy (PSP).

Analysis of DNA hydroxymethylation in human glutamatergic and GABAergic neurons using OxBS MethylationEPIC method

Violetta Rucinski
The City College of New York

Mentor: Stella Dracheva

Methylation of cytosines in DNA is a well-known epigenetic mark that primarily occurs at CpG dinucleotides and is involved in many developmental and regulatory processes. However, recently an unexpected complexity of DNA methylation was revealed, as it was found to be a combination of “true” methylation (mC) and hydroxymethylation (hmC). Importantly, the effects of mC and hmC on gene expression can be opposite, and thus many previous results should be reconsidered. This is especially true in neuroscience, as the levels of hmC are much higher in the brain than in other tissues, and recent work has implicated hmC in neurodevelopment and memory formation. Much research is still needed to better understand hydroxymethylation and its significance. In particular, very little is known about differential distribution and roles of hmC in various brain cell types.

DNA methylation is commonly detected using bisulfite (BS) treatment, followed by sequencing or microarrays. Novel methods such as OxBS sequencing allow discriminating between hmC and mC. Considering costs of BS/OxBS-Seq, an attractive approach is to combine OxBS with microarrays. The recently introduced MethylationEPIC array probes >800,000 CpG positions including promoters, CpG islands and regulatory elements.

In current study, we used flow cytometry to separate nuclei of glutamatergic and GABAergic neurons from human postmortem brain using a protocol developed in the lab. We then used BS/OxBS-MethylationEPIC to complement and validate the whole-genome BS/OxBS sequencing study, also performed in our lab. In agreement with the results of BS/OxBS-Seq, we observed higher average levels of DNA hydroxymethylation in glutamatergic compared with GABAergic neurons. We detected numerous regions of differential hydroxymethylation between the two neuronal cell types, and described the distribution of hydroxymethylation across genomic features.

The current work provides insight into neuron-subtype-specific DNA hydroxymethylation and paves way for future studies in human subjects with neuropsychiatric diseases.

“Development of hepatocellular carcinoma and TP73 expression: Does DNA methylation play a role?”

Savannah Samuel
Xavier University of Louisiana

Mentor: Zaki Sherif

TP73 is a member of the TP53 family of proteins that is currently actively being studied for its role in tumor promotion or suppression in human cancers. As a transcription factor, similar to TP53, the TP73 protein is involved in cellular responses to stress and development. The gene maps to chromosome 1p36 that is rich in tumor suppressor genes but is frequently deleted in neuroblastoma and other tumors. Alternative gene splicing has produced many isoforms of the TP73 protein but the biological validity and the true nature of some of the variants have not been yet determined. In this study, we wish to investigate the role of TP73 in the tumorigenesis of hepatocellular carcinoma. We first established primary cell lines from the hepatic tissues of a young (HEPC-y) and old (HEPC-o) healthy individuals to use as controls in our study. We determined the level of *TP73* gene expression in HepG2, SNU449, SNU475 human Hepatocellular carcinoma (HCC) cell lines by RT-PCR. We also analyzed the TP73 protein level in normal and cancer patients' tumor tissues by immunohistochemistry. Our findings were quite surprising. TP73 was only expressed in the tumor cells but not in the healthy liver cells. Moreover, methylation-specific PCR results show that *TP73* gene expression is regulated by DNA methylation.

NVU-ON-CHIP DEVICE FABRICATION

Roy Samuel
New Jersey Institute of Technology

Mentor: Sagnik Basuray

The neurovascular unit (NVU) is comprised of neurons, astrocytes, pericytes, the blood-brain barrier (BBB) and their cellular interactions. The BBB, a barrier formed by endothelial cells that lines cerebral capillaries, regulates substance exchange between the blood and brain. This selectively permeable layer is implicated in drug delivery, since it precludes the passage of therapeutic nanoparticles involved in treating neurological disorders from the bloodstream into the brain. We are fabricating an NVU-on-a-chip device to supplement traditional, more expensive, in vivo models that often fail to precisely mimic NVU behavior. In future studies, this model could test the effectiveness of drug delivery to the brain.

We have optimized device construction by using electrodes composed of modified common glass slides with gold coated interdigitated electrodes, a permeable membrane, and polydimethylsiloxane (PDMS). The device is constructed using plasma-treated PDMS with an etched 300 micrometer channel placed over the glass slide with a gold-etched electrode interface, which is also plasma treated. The membrane to be used for cell seeding must also be plasma treated. It is then placed in a vial of a solution with 5% 3-aminopropyltriethoxysilane in an attempt to create an irreversible bond between the coated membrane and the plasma-treated PDMS. The membrane, a barrier between the aforementioned channel, is placed in between the PDMS layers. This small area in between the channel is the surface on which RBECs and nervous tissue are seeded on opposite sides, effectively mimicking the NVU.

We plan to analyze the NVU cell-membrane interface with electrical impedance spectroscopy that models the cellular tissue as an equivalent electric circuit. Readings will reveal seeded tissue integrity, BBB resistance and capacitance, and paracellular resistance and transcellular resistance differences. Following the fabrication of this device, a more complex device will be constructed to determine the mechanistic pathway of treatments in drug-induced conditions.

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INVESTIGATING THE BASOLATERAL AMYGDALA'S ROLE IN AUDITORY CUED FEAR RETRIEVAL THROUGH THE USE OF DREADDS

Miranda Scarlata
Vassar College

Mentor: Hadley Bergstrom

Post Traumatic Stress Disorder (PTSD) is one of the most prevalent psychiatric disorders in the world. Pavlovian fear conditioning is the leading preclinical model for studying neuronal circuits likely dysregulated in PTSD. The basolateral amygdala is a key network node necessary for acquiring and expressing conditioned fear memories. One outstanding question is the role of the BLA in the expression over memory over remote timeframes. Using Designer Receptors Exclusively Activated by Designer Drugs (DREADDs), a method for remotely controlling the neuronal activity within brain regions with high spatial specificity, mice were microinjected in the BLA with either the pAAV-CaMKIIa-hM4D(Gi)-mCherry (experimental) viral vector or a pAAV-CaMKIIa-EGFP (control) vector 21 days prior to fear conditioning. 1 day after fear conditioning, the designer drug CNO (3.0 mg/kg) that activates the hM4D(Gi) expressed receptor was intraperitoneally injected 60 minutes before cued fear memory retrieval. The brain was then processed for anatomical verification of viral spread using the fluorescent promoter for visualization. Fluorescent imaging revealed robust expression of control eGFP and some receptor expression of experimental mCherry. When stereotaxic coordinates [D/V: -0.4; M/L: $\pm$ 3.1; A/P: -1.82] were used to locate the BLA, the injection sites appear in the ventral hippocampus (vHC) instead of the BLA. In the cued fear memory retrieval paradigm, experimental mice showed significantly reduced freezing levels compared to control mice. While we were unable to confirm BLA's role in cued fear memory, we were able to demonstrate that the vHC is implicated in fear memory retrieval. Once the BLA is targeted via developing new, more rostral, stereotaxic coordinates, DREADDs can be an effective tool to further exploring the BLA's potential role in PTSD and alcoholism comorbidity through a chronic alcohol exposure paradigm altering auditory cued fear memory generalization.

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ADENOSINE A_{2A} RECEPTOR (A2AR) INHIBITS OSTEOCLAST DIFFERENTIATION AND PROMOTES OSTEOBLAST FORMATION BY REGULATION OF AXON GUIDANCE PROTEINS

Lopa Shah
NYU Medical Langone Center

Mentor: Bruce Cronstein

Bone remodeling is influenced by effective communication between osteoblasts and osteoclasts. Two novel targets have been expressed as essential regulators of bone homeostasis: Adenosine and Semaphorins. The axonal guidance proteins semaphorins (Sema) play an important role in communication between osteoclasts and osteoblasts. Sema4D, secreted by osteoclasts, binds to its receptor PlexinB1 on osteoblasts to inhibit osteoblast differentiation and function. Sema3A, produced by osteoblasts, binds to PlexinA1/Neuropilin-1 to both inhibit RANKL-induced osteoclast differentiation and stimulate osteoblast differentiation and function. Because stimulation of adenosine A_{2A} receptors (A2AR) diminishes osteolysis, we asked whether A2AR activation, which regulates both osteoclast and osteoblast function, modulates bone homeostasis by regulating both osteoclast and osteoblast expression of semaphorin 4D and semaphorin 3A.

Sema3A/PlexinA1/Neuropilin1 and Sema4D/PlexinB1 expression were studied by RT-PCR and Western Blot in primary bone marrow-derived OC and in primary bone marrow derived OB in the presence/absence of CGS21680 and ZM241385, 1 μ M each. RANKL and Osteoprotegerin (OPG) levels were studied by RT-PCR in OB. Sema4D expression and secretion are increased in osteoclast derived cells in the presence of RANKL alone or in the presence of ZM241385 (60 $\pm$ 4% increased vs. basal, $p < 0.001$, $n = 4$); this increase is reverted in the presence of the A_{2A}R agonist. Sema3A mRNA increases during osteoblast differentiation (3.5 $\pm$ 0.5 fold increase) and CGS21680 enhances it (8.7 $\pm$ 0.2 fold increase, $p < 0.001$, $n = 4$). PlexinA1 mRNA is enhanced by CGS21680 (4.5 $\pm$ 0.4 fold increase compared to 1.75 $\pm$ 0.2 for RANKL, $p < 0.001$, $n = 4$).

Our results demonstrate that A2AR activation diminishes secretion of Sema4D by osteoclasts and enhances secretion of Sema3A by osteoblasts leading to an increase in osteoblast differentiation and function, and, in combination with the suppressive effects of A2AR on osteoclast differentiation and function, diminishes bone osteolysis. These results suggest that targeting axonal guidance proteins directly or via stimulation of adenosine A2AR may be a novel approach for bone remodeling.

Ian Smith
Texas Tech University

Mentor: Robert Cox

Smoke exposure is known to influence germination of a wide variety of species. However, the influence of smoke on germination of grasses is poorly studied. In order to investigate the influence of smoke on grasses native to North America, we tested five species for germination response to smoke exposure. Species tested were: sideoats grama (*Bouteloua curtipendula*), blue grama (*Bouteloua gracilis*), big bluestem (*Andropogon gerardii*), buffalo grass (*Bouteloua dactyloides*), American basket flower (*Centaurea americana*), and different varieties of big sagebrush (*Artemisia tridentata*). Seeds were treated with one of four dilutions of smoke-water and distilled water: 0:1 (no smoke), 1:10 (high smoke), 1:100 (medium smoke), and 1:1000 (low smoke). Seeds were soaked in the smoke treatment for 20 hours, then placed in a germination chamber. Multiple germination chamber will be needed because some of the seeds required different growing conditions to mimic their time of germination. We monitored the seeds daily for germination. We hope to find that germination is inhibited in some species and promoted in others at different levels of smoke exposure. We perceive this happening because of past test performed on similar species to the ones that we are studying. It is our hope that we exhibit that different levels of smoke exposure could have a significant effect on germination of many grass species in North America.

IN VITRO MODEL OF ONCOGENESIS IN A PATHOGENIC BRCA2 MUTANT

Zoe Steinsnyder
Memorial Sloan Kettering Cancer Center

Mentor: Vijai Joseph

Mutations in *BRCA2* increase the risk for several human cancers. It's various functions include: maintaining genomic stability, repairing double stranded DNA breaks via homologous recombination (HR), cell cycle progression, and DNA replication. The *BRCA2* 6174delT mutation is seen in 1.5% of Ashkenazi Jews (AJ) and .026% of the general population. It increases the risk of breast cancer in 43% of women by age 70.

Here, we aim to understand the initiation and progression of oncogenesis in a *BRCA2* mutant background as well as potential resistance mechanisms related to current pharmacological treatment. To that end, four isogenic cell lines derived from the Human Mammary Epithelial (HMLE) cell line were exposed to seven different treatments over a period of time. Two cell lines used were *BRCA2* wild type (WT) and the remaining were 6174delT mutants, engineered via CRISPR/Cas9 genome editing. One of each cell line was further modified to respond to inducible estrogen receptor (ER) expression. The treatments for both ER and Non-ER cells lines were: normal passaging, γ irradiation, PARP inhibitors (Olaparib & Niraparib) and Aldehydes (formaldehyde & acetaldehyde). ER cells had an additional treatment of β -estradiol to determine if increased proliferation due to estrogen exposure affects genome stability in the *BRCA2* mutant state.

Treatments were carried out twice a week. Genomic DNA from each passage is being analyzed for genomic aberration using Short Multiply Aggregated Sequence Homologies (SMASH). SMASH sequencing detects genomic instability and copy number variations. We expect to identify signatures of genomic instability and mutations that confer resistance to the treatments employed.

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Don Stimpson
Rensselaer Polytechnic Institute

Mentor: Brigitte Arduini

Transcription factor FOXD3 has been linked to metastatic behavior, proliferation, and invasiveness in cancer cells, although the research done on this phenomenon so far is limited. FOXD3 overexpression in human embryonic stem cells (hESCs) and in other developmental contexts has been shown in past studies to promote epithelial to mesenchymal transition (EMT). Strikingly, the opposite appears to be the case in cancer cell lines. We have created transgenic cell lines that allow us to selectively overexpress or knock down FOXD3 expression in a line of human neuroblastoma (SK-N-AS). We find that the morphological response to this overexpression in neuroblastoma cells appears to be reciprocal to the response of hESCs in several key respects. Time lapse visualization shows overexpression decreases mobility and increases cell clumping in SK-N-AS. Immunofluorescence experiments suggest cytoskeletal changes occur with respect to BETA-ACTIN, as opposed to BETA-TUBULIN in hESCs. These results provide insight into the possible roles of FOXD3 in normal human development and in cancer metastasis.

INCREASED VOCAL, NOT FACIAL, EMOTION RECOGNITION PREDICTS POSITIVE EVALUATION BY PEERS IN YOUTH WITH AUTISM SPECTRUM DISORDER

Amanda Stoerback
Stony Brook University

Mentor: Matthew Lerner

Youth with Autism Spectrum Disorder (ASD) experience characteristic deficits in the development of social relationships. In addition, they are known to experience deficits in emotion recognition across both facial and vocal domains. While numerous theoretical models posit that emotion recognition is essential for positive peer regard, the degree to which this is empirically so – and whether it differs by domain – in ASD remains strikingly understudied. The present study examined the relationship between emotion recognition ability and peer regard in youth with ASD.

55 youth ($M_{age} = 12.42$, $SD_{age} = 2.87$) with ADOS-2-confirmed ASD completed the Diagnostic Analysis of Nonverbal Accuracy (DANVA-2) to quantify facial and vocal emotion recognition abilities. They then participated in a ninety-minute social group session in groups of 5-9. Upon completion, they engaged in a sociometric interview to evaluate the degree to which they were liked and made friends in the group.

Bivariate correlation revealed that errors made in vocal emotion recognition correlated with being the most liked person in the group ($r = -.349$, $p = .011$) such that youth who made fewer errors, were more often rated by peers as the most liked person in their group. This relationship was driven by errors made in child voices ($r = -.358$, $p = .009$), rather than adult voices ($p = .052$). There was no significant relationship between facial emotion recognition and any sociometric variables (all $ps > .192$).

These results indicate that, contrary to many theoretical models and the premise of several interventions, facial emotion recognition has little to do with the sociometric status of youth with ASD. Instead, vocal emotion recognition is more salient specifically in relation to being the most liked person in a group and may play a role in improving how youth with ASD are perceived by peers upon first impressions.

INVESTIGATING AMYLOID FORMING ACTIVITY IN ELK FECES

Margaret Tyer
Hampshire College

Mentor: Cynthia Gill

Chronic wasting disease (CWD) is a transmissible spongiform encephalopathy, also known as prion disease, endemic to white-tailed deer, elk and other cervids located in western United States. CWD is the only prion disease present in both captive and wild animals and its notable horizontal transmission suggests environmental contamination from cervids, such as from sheddings, may contribute to the spread of disease. I investigated two aspects of CWD and sheddings in research conducted with the Prion Research Center of Colorado State University to ask (1) Is there is a correlation between of prion forming activity in tissue and fecal levels in infected elk? (2) Is prion forming activity conserved in elk feces that are dried and exposed to UV light (sunlight)? Using an *in vitro* amplification assay known as real-time quaking induced conversion (RT-QuIC), we have preliminary data indicating the rates (high, medium, and low) of prion formation within elk tissue correlate with the rate of amyloid conversion in elk feces. Additionally, we have found that amyloid forming activity is conserved in dried, UV-exposed feces at rates comparable to that of wet, unexposed feces.

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LACK OF CHRONIC NEUROINFLAMMATION IN THE ABSENCE OF FOCAL HEMORRAGE IN A RAT MODEL OF LOW-ENERGY BLAST-INDUCED TBI

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Mentor: Miguel Gama-Sosa

Exposure to blast overpressure waves from detonated explosives can cause blast-related traumatic brain injury (TBI). Depending on the blast energy and the duration of exposure, blast waves can damage the brain microvasculature and disrupt the blood-brain barrier, triggering neuroinflammation. Other complications associated with blast-related TBI include epilepsy, seizures, PTSD, and cerebral vasospasms. Even though blast-related TBI is estimated to affect a large portion of the veteran population, the pathology of blast-related TBI is not completely understood.

Our study focused on the effects of repetitive, low-energy blast exposures in the rat model. Six weeks following three 74.5-kPa blast exposures, there were no significant changes in microglial activation, a marker for neuroinflammation. At 40 weeks post blast exposure, there were no cytokine differences observed between control and blast exposed animals. Interestingly, one animal demonstrated chronic microglial activation surrounding a focal hemorrhage 16 weeks post blast exposure. These findings suggest that a focal hemorrhage following blast-induced TBI may trigger chronic neuroinflammation. However, in the absence of focal hemorrhaging, chronic neuroinflammation is not a general feature of low-level blast injury.

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VALIDATION OF NOVEL 5' END RNA SEQUENCING TECHNIQUE

Samvida Venkatesh
Princeton University

Mentor: Zhiyuan Hu

RNA sequencing enables a quantitative analysis of the cellular transcriptome and differential gene expression. Existing techniques for both bulk and single-cell (sc) RNA sequencing are primarily 3' selective and expensive. A new 5'-end RNA-sequencing technique developed in the lab, labeled 'EndSeq', is able to sequence both bulk and single-cell data to 1/10th the depth of accepted techniques while still capturing gene expression. This abstract highlights the methods used to validate EndSeq against accepted RNA sequencing techniques in the field.

We identically processed bulk FASTQ files generated by bulk-EndSeq, SmartSeq2, and RNA-Seq, and sc-EndSeq and Nextera. This included trimming, mapping reads to the genome, and generating the counts matrix of genes. Additionally, EndSeq Unique Molecular Identifiers (UMI) counts were de-duplicated. We then performed quality control in R.

For both bulk and sc-EndSeq, we confirmed that there was significant 5' end gene coverage compared to other techniques. We showed that EndSeq had low total counts (depth), but identified similar number of features as other techniques. Finally, we demonstrated that the percentage of exons, introns, and unmapped regions were comparable for all the methods.

We demonstrated reasonable correlation (0.50) between gene expression from bulk-EndSeq and the other poly-A tailed-RNA sequencing method SmartSeq. We verified that EndSeq correlates to the "gold standard" RNA-Seq technique just as well as SmartSeq does. Finally, we checked that a significant proportion of the differentially expressed genes identified by EndSeq are also identified by other techniques.

We validated sc-EndSeq by clustering data from 9000 single T-cells isolated from different tissues. Cells clustered by tissue type, verifying that EndSeq picks up important differential gene expression. Additionally, we were able to isolate populations of activated CD8+ T-cells.

We show that low-cost, low-depth 5'-end RNA-sequencing with EndSeq matches accepted techniques, and captures differential gene expression in bulk and single-cell data.

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BLAST BRAIN INJURY RESULTS IN METABOLIC DYSFUNCTION

Phuong Vo

Icahn School of Medicine at Mount Sinai

Mentor: Christoph Buettner

Background: Soldiers and law enforcement officers risk exposure to explosive blasts in the line of duty, where even non-contact injuries may affect their health. There is evidence that blast-related traumatic brain injury (TBI) leads to physical and mental health complications. Blast-related TBI has been associated with pituitary dysfunction, and post-traumatic stress disorder has been associated with a 40% increase in risk of developing diabetes. TBI caused by blast exposure in armed conflict has been studied in relation to several stress and psychiatric disorders; however systemic metabolic dysfunctions have not been researched.

Methods: Long-evens rats were anesthetized and placed in a compression-driven shock tube to simulate the effects of non-contact blast injuries. The animals had their body mass tracked during the course of the study. After they had recovered from the blast, their glucose metabolism was studied using glucose tolerance (GTT) and insulin tolerance tests (ITT). At the end of the study the animals were sacrificed, and had their fat pads removed and weighed. Blood samples were collected and plasma triglyceride and insulin levels were measured.

Adipose and liver were used to

Results: The animals had no differences in body mass ($p=0.41$) and no differences in any of the fat pad masses. Despite no differences in body mass or fat mass, blast animals had impaired glucose tolerance during a GTT compared to the control animals ($p=0.01$) using a repeated measures ANOVA. The blast animals also showed reduced insulin sensitivity compared to control animals during the ITT ($p=0.03$) using repeated measures ANOVA. There were no differences in liver triglyceride levels between BLAST and control animals ($p=0.44$) using a one way ANOVA. In addition, there were no differences in plasma insulin between BLAST and control animals ($p=0.26$).

Conclusion: The blast exposure led to impaired glucose tolerance and insulin resistance despite similar body mass and fat masses. Blast-induced traumatic brain injury may lead to disruption in the brain's ability to regulate glucose metabolism, as the changes seem to be independent of changes in adiposity or body mass. The exact malfunctions are difficult to determine as blast injuries show varied distribution of injuries within the brain. This discovery requires further study to mitigate the risks of metabolic diseases to individuals exposed to blast injuries.

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The Functions of Circulating Tumor Cells and ctDNA in the Early Diagnosis and Real-time Monitoring During Cancers Advancement

Yuqiao Wang

The Cooper Union for the Advancement of Science and Art

Mentor: Zhiyuan Hu

In traditional cancer pathology, tumor imaging and a biopsy sample taken from a sector of a patient's tissue have long been the marks of the disease progression. However, the scarcity of tissue, inability to track the development of cancer real-time, and the failure to resolve the heterogeneity of tumor cells throughout the body urge the alternatives to traditional biopsy. Circulating Tumor Cell (CTC) has become a proper candidate of a "liquid biopsy" for its homogeneity, prevalence, and ease of detection. In this review, addressed are the fruit of applying CTC test to monitor the advancement of cancer, the relevance between CTCs concentration and a patient's expected lifetime, as well as techniques of CTCs collection and quantification. Such progress provides a handy tool to understand the behaviors of cancer cells during metastasis and develop corresponding treatments subject to each patient's individual status. This research received guidance from Dr. Zhiyuan Hu from the National Center of Nanoscience and Technology, Chinese Academy of Science.

Jesse Woodbury
Morehouse College

Mentor: Rajiv Saigal

Spinal cord injury (SCI) involves both a primary trauma and secondary injury. Secondary injury involves multiple pathophysiological mechanisms that can further compromise sensory and motor function. Inflammation is a central component of secondary injury. Dexamethasone is a glucocorticoid steroid used for its anti-inflammatory properties. Use of steroids after spinal cord injury is controversial due to systemic side effects. Intravenous steroids after SCI have been associated with increased rates of infection, sepsis, myopathy, and GI bleeding.

Polymers and biomaterials have been explored for local controlled drug release in order to achieve therapeutic efficacy while reducing systemic side effects. A recent study showed that prophylactic epidural methylprednisolone was modestly effective in improving recovery from experimental spinal cord injury in a rodent model [1]. Other studies have demonstrated the electrical stimulation of dexamethasone from polypyrrole microreservoirs promotes osteogenic differentiation in a bone tissue engineering model[2].

Here we sought to test whether electronically controlled release of dexamethasone could decrease inflammation in an in vitro model of central nervous system inflammation. We first tested whether dexamethasone could be encapsulated and later released via electronic control in polypyrrole (PPy), a model conductive polymer. We then activated BV-2 mouse microglia, in order to model post-SCI related inflammation. We tested biocompatibility and efficacy of electronically-controlled release of dexamethasone from PPy thin films in mitigating free radical production.

Laura Xiong
Hofstra University

Mentor: Emily Mundorff

Haloalkane dehalogenases (HLDs) use the hydrolytic mechanism to catalyze the cleavage of the carbon-halogen bond resulting in the formation of an alcohol, proton, and a free halide. Halogenated compounds are toxic byproducts of many industrial processes, which gives HLDs significant potential in biosensing, and bioremediation processes. A recently discovered HLD from *Caulobacter crescentus* (DccA) was found to be highly active towards brominated and chlorinated haloalkanes greater than three carbons in length. Many of the most important toxic halogenated compounds are smaller chlorinated compounds such as 1,2,3-trichloropropane, 1,2-dichloroethane, and 1-chlorobutane. We present the results of our ongoing directed evolution efforts to improve DccA's activity towards chlorinated haloalkanes and the identification of activity on smaller haloalkanes, and present our progress of library screenings of test combinations of mutations.

The reproductive impairment observed in male mice with adipose tissue-specific deletion of p110 α is associated with altered testicular mRNA expression of steroidogenic proteins.

Leon Yang
Stony Brook University

Mentor: Maricedes Acosta-Martinez

The adipose tissue modulates mammalian reproduction through the cross-talk between adipose factors called adipokines and the hypothalamic pituitary-gonadal axis. Adipokines such as leptin act centrally in the hypothalamus to regulate gonadotropin hormone release and peripherally in the gonads to regulate sex hormone production. In addition to its role in the central regulation of gonadotropin release, the phospholipid enzyme PI3K mediates insulin effects on glucose uptake and lipolysis. Deletion of PI3K catalytic subunit p110 α in adipose tissue (aP2-Cre/p110 α flx/flx, α -/- hereafter) results in increased adiposity, glucose intolerance, and liver steatosis. In addition to these metabolic impairments, α -/- males displayed delayed onset of puberty, whereas adult males exhibited hyperandrogenemia and were unable to sire pups in proven fertile females. Because hyperleptinemia was observed in both peripubertal and adult α -/- mice we investigated the testicular gene expression of markers of sex-steroid production. At postnatal day 30, testicular leptin gene expression was increased, whereas expressions of the cholesterol transporter StAR and of P450 cholesterol side chain cleavage enzyme were decreased in α -/- animals. The mRNA levels of leptin and of 17-beta dehydrogenase 3, an enzyme important for testosterone production, were significantly higher in the testes of adult α -/- males. The mRNA levels of ER α , an important regulator of intratesticular steroidogenesis, were lower in the testes of adult and peripubertal α -/- males. We propose that chronic hyperleptinemia contributes to the negative impact that disrupting PI3K signaling in adipocytes has on puberty onset, steroidogenesis, and fertility in male mice.

ELUCIDATING THE MOLECULAR FUNCTION OF THE COMMON 6174DELTA BRCA2 TRUNCATION MUTATION

Frederick Yen
City University of New York, Hunter College

Mentor: Ryan Jensen

Cancer is a disease driven by the accumulation of genetic mutations. Mutations in the BRCA2 gene increase the risk of hereditary breast and ovarian cancer by 70% to 40%, respectively. The 6174delT frameshift mutation in BRCA2 is the most frequently mutated allele in Ashkenazi Jewish women associated with a high lifetime risk for developing aggressive breast and ovarian cancer. While the 6174delT mutation is clearly linked to cancer predisposition, the altered molecular mechanisms resulting from BRCA2 protein truncation leading to tumorigenesis remain to be defined. In order to unmask the role of the 6174delT mutation, we are studying the genetic and biological impact of expression of this truncated protein in human BRCA2 knockout cells.

The BRCA2 mutated C-terminal truncated allele was generated by PCR, cloned into the pCMV1 mammalian expression vector containing an N-terminal 2XMBP (tandem Maltose Binding Protein) tag, and stably expressed in human DLD1 BRCA2 knockout cells. Multiple single cell clones were analyzed for 2XMBP BRCA2 6174delT expression by western blot and RT-PCR analysis.

Our future plans are to define the consequences of BRCA2 6174delT protein expression in a BRCA2 null background to shed light on the molecular pathogenicity of this frequent mutation leading to cancer. Our approach will encompass studying DNA damage signaling pathways, cellular localization of the truncated protein and protein binding partners of BRCA2 by confocal microscopy and proximity ligation assay, and analysis of DNA replication in response to DNA damage stress using single molecule DNA fiber combing assays. Our goal is to determine the molecular functions of BRCA2 that become compromised upon expression of this truncated allele.

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