UNIVERSITY of WASHINGTON

Undergraduate Research Symposium May 19, 2017 Mary Gates Hall

Online Proceedings

Poster Session 1

MGH 241, Easel 140

11:00 AM to 1:00 PM

Analysis of Coordinated Kinase Activity during Gap Junction Disassembly

Joshua Michael (Josh) Dawson, Junior, Extended Pre-Major
Mentor: Joell Solan, Public Health Sciences
Mentor: Paul Lampe, Pathobiology

Gap Junctions (GJ) are specialized portions of cell membranes that connect adjacent cells and are critical for synchronous electrical signaling in the heart. Therefore, better understanding of GJs can lead to more effective treatment for individuals with arrhythmia. GJs are a collection of channels that pass small molecules and metabolites between cells. They are composed of connexin proteins, the most common being Connexin 43 (Cx43). Kinases phosphorylate Cx43 to control the assembly and disassembly of GJs. Specifically, Protein Kinase C (PKC) and Extracellular Signal-Regulated Kinase (ERK1/2) phosphorylate Cx43 during assembly and disassembly and are activated during cardiac ischemia. PKC phosphorylates amino acid S368 on Cx43, which changes channel permeability, while residues S279/282 can be phosphorylated by ERK1/2 which results in channel closure. Lastly, it is known that S262 is phosphorylated under conditions which lead to GJs disassembly. However, both PKC and ERK1/2 have been implicated as the S262 kinase, in part because typical reagents that activate PKC also result in activation of ERK1/2. My study was designed to clarify which of these kinases phosphorylates S262 by quantifying phosphorylation of Cx43 at S368, and S262 in the presence of PKC and ERK1/2 kinase activators and inhibitors. By examining S262 phosphorylation using different combinations of these reagents, we should be able to discern which kinase phosphorylates S262 or, at least, whether this event correlates with phosphorylation on S368. The data indicated that ERK1/2 is the kinase involved in S262 phosphorylation due to the greatly diminished phosphorylation activity observed when ERK1/2 inhibitors were introduced; and that the PKC activator, TPA, in reality plays a role in stimulating ERK1/2. I now want to find out which phosphorylation event is the most important aspect for gap junction disassembly between MAPK and PKC and perhaps if the two kinases work together.

Session 1E

Excitations: Art and Visuality

Session Moderator: Rebecca Cummins, School of Art + Art History + Design

MGH 238

12:30 PM to 2:15 PM

* Note: Titles in order of presentation.

Spiritual Vinyl Spectrographs

Alexis M. (Alexis) Neumann, Junior, American Music, Comparative History of Ideas
Mentor: Joel Ong, Department of Computational Arts
Mentor: Phillip Thurtle, Comparative History of Ideas

Frequencies are heard vibrating inside of our minds, bodies and souls, and these sensations influence spiritual experiences of the world. My project visualizes the religious experiences of sound by dripping paint onto vinyl records in order to illustrate the beauty and extrasensory splendor of religious sounds. The introduction of audio recording gives religious music new implications and availability because the religious practitioners do not need to be physically present to engage in the sound. Some argue that this makes spirituality more present throughout our every day lives, whereas others reason that the novelty and intimacy of live performance in religious settings is more powerful, and that this experience is often lost because of the convenience of recordings. By creating a physical mixed media spectrograph of the frequencies of the recorded religious music, my work intends to visualize the audial, sensorial experience of the religious practitioner. Religious artwork that highlights the divine, beatific aspects of sound and music encourages respect and appreciation of the charm of other religions and promotes open-mindedness because there is a similar yet unique beauty in each auditory experience of these spiritual sounds.

Poster Session 2

MGH 241, Easel 142

1:00 PM to 2:30 PM

Further Investigation into the Influence of UGT1A4*3 Genotype on The Glucuronidation of 25-Hydroxyvitamin D_3

Huanbin Huang, Senior, Physics: Biophysics
Mentor: Kenneth Thummel, Pharmaceutics
Mentor: Tim Wong, Pharmaceutics

Uridine diphosphate glucuronosyltransferase 1A4 (UGT1A4) is a phase II drug metabolizing enzyme that plays an important role in the conjugation of various xenobiotics and endogenous compounds. In the context of vitamin D metabolism, conjugation of 25-hydroxyvitamin D3 (25(OH)D3), the main circulating form of vitamin D, may function as a pathway to help facilitate the enterohepatic circulation of vitamin D. Allele variants of the UGT1A4 gene are reported to contribute to inter-individual differences in enzyme activity, with the UGT1A4*3 variant gene product exhibiting increased glucuronidation activity. Previous work in our lab evaluated the polymorphic glucuronidation of vitamin D using human liver microsomes isolated from liver samples that were procured by St. Jude Children's Research Hospital and University of Washington. The study found that carriers of the UGT1A4*3 allele showed increased 25(OH)D3 glucuronidation activity. While the samples used in this previous work was limited to a smaller subset of livers with the UGT1A4*3 genotype, we sought to further our investigation to include gene expression data obtained through RNA-Seq analysis and UGT1A4 protein quantification in LC-MS data. In addition, the current analysis was expanded to include a significantly greater portion of both the St. Jude and UW liver banks. From this study, we hope to gain greater insight into the impact that the *3 allele may have on the RNA and protein expression of UGT1A4. In addition, by assessing the relationship between UGT1A4*3 genotype and 25(OH)D3 glucuronidation, we may develop a better understanding of factors that may contribute to the variability in the biliary excretion and/or plasma levels of vitamin D glucuronides.

SESSION 2C

USING SPECULATION, POETICS, AND ART TO UNDERSTAND BIOLOGICAL RELATIONSHIPS

Session Moderator: Phillip Thurtle, Comparative History of Ideas

MGH 231

3:30 PM to 5:15 PM

* Note: Titles in order of presentation.

Noise Pollution Effects on Whales

Viviana Carolina (Viviana) Castillo Contreras, Senior, Oceanography
Mary Gates Scholar
Mentor: Phillip Thurtle, Comparative History of Ideas
Mentor: Rebecca Cummins, School of Art + Art History + Design
Mentor: Tyler Fox, Human Centered Design & Engineering, College of Engineering, UW
Mentor: Joel Ong

What happens when the form in which whales communicate, locate food and find each other, is clouded by large ships, sonar technology and increased ice cracking noise in their environments? This presentation considers what the world of whales looks like beneath the ocean. I address this by taking a closer look at the auditory world of whales as a direct result of the lack of visual abilities due to limited light. I have focused on the chaos whales feel as a result of noise inserted within the depths of the ocean. With an increase in technological advances we see an increase in the number of ships and consequently of noise in the ocean. Underwater noise pollution is also attributed to ice fractures and collisions. These events do not occur at one singular time, they occur simultaneously for unknown periods of time and unpredictable areas. Understanding how animals feel and what they think is a difficult task, but we can better understand them by taking a closer look at their behaviors as the noise in the ocean becomes more abundant. My intention is that I will be able to formulate the chaos heard underwater into a humanized perspective and begin to understand these creatures as the hearing, feeling mammals they are.

SESSION 2C

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MGH 231

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Data Day: Mindful Tracking of Changes in Emotions
Gerlene Ragmac (Gerlene) Ragsac, Senior, Comparative History of Ideas
Mary Gates Scholar
Mentor: Phillip Thurtle, Comparative History of Ideas
Mentor: Rebecca Cummins, School of Art + Art History + Design
Mentor: Tyler Fox, Human Centered Design & Engineering, College of Engineering, UW
Mentor: Joel Ong

I view energy as strongly intertwined with emotions, and that they are powerful forces that fluctuate and influence decisions. Thus, I chose to track instances where I felt a change in emotion in conjunction with its duration, what thought caused it to emerge, and if there was an attempt to deliberately shift moods. By developing my own codes and keys, I was able to monitor these fairly accurately on paper. However, I wanted to present this data in ways that differed from "traditional" portrayals of information. Hence, I used yarn and glitter in glass bottles to mimic bar graphs and liquid samples, respectively. These materials also served to emphasize the therapeutic nature within my project. The ideas of tracking something and its presentation were heavily inspired by duration exercises, which were done as assignments for the Summer Institute in the Arts & Humanities 2016, as well as Dear Data. The former required us to keep track of the passage of time for a minute, an hour, and 24 hours; the latter was a collaboration between Giorgia Lupi and Stefanie Posavec where they exchanged weekly postcards over the course of one year. Each week's postcard included a theme of what they would both monitor in their daily life, a visual representation of the data they collected, and a key to decipher the image. Initially, my desire for this project was to encourage others to engage in mindful practices in order to experience the benefits it may have in their everyday life. However, by making this project deeply intertwined with something so personal as emotions, I also hope to challenge people to contemplate the relationship between emotions and data and the emphasis placed on pursuing objective results.

POSTER SESSION 3

Balcony, Easel 106
2:30 PM to 4:00 PM

Development of a Doxorubicin-Loaded Cyclic Polymer Drug Delivery System
Karl Theodore (Karl) Manner, Senior, Bioen: Nanoscience & Molecular Engr
Mary Gates Scholar, UW Honors Program
Mentor: Suzie Pun, Bioengineering
Mentor: Yilong Cheng, Department of Bioengineering

The most common form of cancer treatment is the use of small-molecule chemotherapeutic agents. However, due to non-specific distribution and uptake, these drugs have a host of complications that limit their efficacy and usage. Polymer nanocarriers have been demonstrated to improve drug pharmacokinetics, enhancing therapeutic benefit of the drugs. The feasibility of cyclic micelle-like polymer nanoparticles for use as a delivery vehicle for doxorubicin (abbreviated as DOX), a potent chemotherapeutic drug, is discussed here. It is hypothesized that cyclic polymers will afford a more efficacious pharmacokinetic release profile than their linear counterparts. A linear polymer is functionalized with "click"able moieties at each end, which are then joined to create a single cyclized polymer. The polymer is decorated with poly(ethylene glycol) (PEG) chains to reduce immunogenicity, and carboxylic acid groups to electrostatically complex with the positively-charged DOX molecule. Upon addition of DOX, the polymer self-assembles into unimolecular micelle-like structures. When the construct accumulates in a tumor microenvironment due to the enhanced permeability and retention (EPR) effect, the low pH of the tumoral interstitium protonates the carboxylic acid groups, releasing DOX locally. Using a unimolecular micelle-like approach, less polymer can be used to deliver the same therapeutic payload, relative to other similar approaches that utilize pluralities of potentially toxic polymers to form micelle structures. The polymer's average molecular weight is around 11530 Da and the polydispersity of the reaction is 1.067. Gel Permeation Chromatography, Nuclear Magnetic Resonance, Fourier-Transform Infrared Spectroscopy, Ultraviolet-Visible Spectroscopy and Dynamic Light Scattering are used to confirm chemical identity, drug loading efficiency, particle size, and polydispersity. Cytotoxicity studies conducted with MDA-MB-231 invasive ductal carcinoma cells *in vitro* demonstrate the feasibility of this construct.

POSTER SESSION 3

Commons West, Easel 39
2:30 PM to 4:00 PM

The Sound of Light: Transduction as a Way of Knowing
Nathan Christopher Mahr, Senior, Comparative History of Ideas
Mary Gates Scholar
Mentor: Tyler Fox, Human Centered Design & Engineering, College of Engineering, UW
Mentor: Phillip Thurtle, Comparative History of Ideas
Mentor: Rebecca Cummins, School of Art + Art History + Design
Mentor: Joel Ong, Department of Computational Arts

What would it mean to be able to hear light? How could this new perspective on light illuminate the fundamental intricacies of light energy? In Gilbert Simondon's musings on individuation he argues that the individual is never given in

advance but is constantly coming into being through a process of interacting with its milieu and realizing potentials out of huge pool of possibilities. This work uses Simondon's focus on individuation and life as a never ending process of development to understand how energy exists through a similar process of change and potentials. Utilizing solar panels hacked into speakers, this installation seeks to employ the construction of a new relationship to light as a methodology for thinking about how energy emerges. The solar panels in the installation transduce light energy into electrical energy which is then fed into speakers for the participants to hear. Energy is most essentially a process of change, disruption and movement. It demonstrates to us the myriad of potentials which lie within and between us and energy. Through the use of transduction of energies this installation allows us to peer into the minutiae of our relationship with energy and begin to conceive of how we absorb, alter, collaborate and connect with it on a daily basis.

POSTER SESSION 3

Commons West, Easel 22

2:30 PM to 4:00 PM

Optimization of CRISPR-Activation to Engineer Metabolic Pathways

Anika Smita (Anika) Patel, Senior, Biochemistry
UW Honors Program
Mentor: Jesse Zalatan, Chemistry
Mentor: Chen Dong, Chemistry

Bacteria are widely used in metabolic engineering because they can synthesize chemical products from cheap, renewable carbon sources. A major current problem, however, is that toxic intermediates can build up when bacteria express heterologous biosynthetic pathways. In principle, we can control metabolic gene expression levels so that downstream enzymes quickly consume toxic intermediates. To achieve this goal, we use a programmable gene regulatory tool called the CRISPR-Cas system. To optimize gene activation activity of CRISPR-Cas in bacteria, we sought to identify modular transcriptional activation domains that could be recruited to specific genes to activate transcription. This strategy has been previously used successfully in eukaryotic cells, but with only limited success in bacteria. We used CRISPR-Cas to recruit a number of potential synthetic transcriptional activators to specific loci in *E. coli*, and identified candidates that can activate a fluorescent reporter gene. We also used protein-engineering approaches to optimize the gene activation activity of our CRISPR-Cas system. The transcription factor screening and protein engineering we did gave an 80-100-fold increase in activation compared to basal transcription levels. To make this new tool useful for practical metabolic engineering, we plan to use this system to dynamically respond to metabolic stresses. This tool could potentially be used to optimize biosynthesis of useful polymer precursors such as lactate and p-aminostyrene. Production of these types of cheap, biosynthetic precursors will help us move towards a greener industrial future.

POSTER SESSION 3

Commons East, Easel 75

2:30 PM to 4:00 PM

Analysis of Optical Properties of Transition Metal Dichalcogenides

Essance Leigh (Essance) Ray, Senior, Bioresource Science and Engineering
NASA Space Grant Scholar
Mentor: Xiaodong Xu, Physics, Material Science and Engineering

Semiconductor materials are essential components for modern electronics, making them one of the most useful tools in the modern world. In recent years, scientists have discovered single layered, two-dimensional (2D) materials that exhibit semiconducting properties. These 2D semiconductors can be used to create light emitting diodes (LEDs), field-effect transistors (FETs) and are promising candidates for a new generation of high performance solar cells. The optical response of these 2D semiconductors are highly sensitive to the crystal purity; impurities within the crystal lattice can obscure their unique optical properties. Here, we use photoluminescence spectroscopy to characterize the optical properties of tungsten diselenide (WSe2) produced using a new high pressure growth technique at the National Institute for Materials Science and Engineering in Japan. We analyze the spectrum of emitted light to determine the crystal quality and compare the degree of circular polarization in the sample photoluminescence, a key indicator of crystal quality, with other samples found in the literature. We find the sample of WSe2 to be of good quality with few impurities, making this method a step forward in the production of high quality WSe2 crystals.

POSTER SESSION 4

MGH 241, Easel 160

4:00 PM to 6:00 PM

Influence of Single Nucleotide Polymorphisms in SULT2A1-mediated Sulfonation of 25-Hydroxyvitamin D_3

Sara Soofian, Senior, Public Health-Global Health
Mary Gates Scholar, UW Honors Program
Mentor: Tim Wong, Pharmaceutics
Mentor: Kenneth Thummel, Pharmaceutics

Vitamin $D_3$is an essential hormone involved in the regulation of calcium and phosphate homeostasis. The major circulating form of vitamin D_3 is 25-hydroxyvitamin D_3 (25(OH)D_3)

which is the biomarker often used as a measure of vitamin D status. In addition, the sulfonated form ($25(OH)D_3$-sulfate) has also been found to be a major metabolite, often circulating in plasma at concentrations comparable to, and at times exceeding, levels of the unconjugated form. Sulfate conjugation occurs primarily in the liver and is catalyzed by the enzyme sulfotransferase 2A1 (SULT2A1). In this study, we evaluated allele variants of the SULT2A1 gene in an effort to better understand factors that may contribute to the inter-individual variation of plasma vitamin D sulfate concentrations. Using liver samples obtained by St. Jude Children's Research Hospital and the University of Washington, our analysis revealed the presence of three common single nucleotide polymorphisms (SNPs). Of the three SNPs, only one SNP (rs296361) was found to be significantly associated with SULT2A1 mRNA and protein expression as well as vitamin D sulfonation activity. Specifically, we found that homozygous carriers of the mutant allele had significantly reduced expression of SULT2A1 both at the RNA and protein level as well as significantly reduced sulfonation activity. The results of this study provide valuable insight into the role that genetics plays in the disposition of vitamin D, and allows us to better understand factors that may impact inter-individual differences in plasma vitamin D levels.

POSTER SESSION 4

MGH 241, Easel 134

4:00 PM to 6:00 PM

Macrophage *Mmp28* Expression is Regulated by Type 1 Interferons

Joe Volk, Senior, Biology (Molecular, Cellular & Developmental)
Mentor: Anne Manicone, Medicine
Mentor: Matthew Long, Pulmonary and Critical Care Medicine

Macrophages are specialized cells of the immune system that direct the pro-inflammatory response and its resolution. Matrix Metalloproteinase 28 (MMP28) is an extracellular matrix proteinase expressed in alveolar macrophages during lung inflammation and is involved in macrophage recruitment and polarization. Our aim was to explore the regulation of *Mmp28* expression in macrophages. We hypothesized that LPS (lipopolysaccharide, a component of bacterial cell membranes which simulates bacterial infection) stimulation would increase *Mmp28* expression via a TLR4-dependent (Toll-Like Receptor 4) pathway involving Type I Interferon (IFN) signaling. We also hypothesized that Polyinosinic-polycytidylic acid (Poly(I:C), a synthetic form of double-stranded RNA which simulates viral infection) would increase *Mmp28* by a TLR3-dependent pathway. To test these hypotheses, we generated bone-marrow derived macrophages (BMDM) from wild-type mice. We exposed these cells to purified recombinant IFNα/β (two forms of IFN involved in antiviral responses), LPS, Poly(I:C), or control media; 4-6 h after treatment, we isolated RNA from the BMDM, synthesized cDNA, and used qPCR to measure mRNA levels of several inflammatory genes. We also included BMDMs from knockout mice for several proteins thought to be part of the *Mmp28* regulatory pathway. We confirmed that LPS and Poly(I:C) increased *Mmp28* expression in BMDM. By comparing knockout mice for two adaptor molecules shared by TLR4, we found that LPS-stimulated *Mmp28* expression was retained in *MyD88-/-* but decreased in *TRIF-/-* macrophages. Since both poly(I:C) and the TRIF-dependent response to LPS induce a type I IFN response, we tested if *Mmp28* expression was dependent on type I IFN by using *IFNAR-/-* (IFN Receptor) macrophages. We found that both TLR4 and TLR3 ligands require IFNAR for *Mmp28* expression. These data link type I IFN responses to regulation of MMP28 and development of experimental COPD and emphysema models of lung disease, and further our understanding of how MMP28 may regulate macrophage responses.

POSTER SESSION 4

MGH 241, Easel 161

4:00 PM to 6:00 PM

Single Nucleotide Polymorphisms in SULT1A1 and Their Impact on SULT1A1 RNA and Protein Expression in Humans

Gregory Mun Sum (Gregory) Tong, Senior, Biology (Molecular, Cellular & Developmental)
Mentor: Tim Wong, Pharmaceutics
Mentor: Kenneth Thummel

Sulfate conjugation is an important metabolic pathway which has been shown to play a vital role in modulating the biological activity and disposition of drugs and endogenous compounds. Studies have shown that sulfotransferases (SULTs) have a broad substrate selectivity that includes hormones, neurotransmitters and xenobiotics. SULT proteins are expressed ubiquitously in human tissue, but is largely expressed in the gastrointestinal tract, liver and bone marrow. Numerous SULT isoforms exist in humans and the most predominate of these isoforms in the liver is SULT1A1. In this study, we evaluate the impact of various single nucleotide polymorphisms (SNPs) present in the SULT1A1 gene using samples obtained by St Jude Children's Research Hospital and the University of Washington. Of particular interest is the effect these SNPs have on the RNA and protein expression of SULT1A1. In total, 17 SNPs within the SULT1A1 gene was identified. A subsequent analysis using RNA-seq data found that 7 out of the 17 SNPs were common variants with an allele frequency greater than 5%. To assess the effects of these SNPs on the protein expression of SULT1A1, samples of human liver cytosol was analyzed using a mass spectrometry based protein

quantification method. While this protein analysis continues to be an ongoing effort, we expect results from this analysis to be consistent with genotypic trends observed in our RNA-seq dataset. By evaluating the relationship between various SNPs in the SULT1A1 gene and their effect on SULT1A1 RNA and protein levels, we hope to expand on our knowledge of the impact SNPs have on the disposition of substrates that undergo sulfonation.