Annual CSU Summer Symposium at UCLA

Tuesday, August 9, 2022
1:00 - 3:00 p.m.
Geffen Hall Learning Studio

We would like to thank the following for their generous support of the CSU Summer Symposium at UCLA:

UCLA Graduate Programs in Bioscience

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CSU SUMMER SYMPOSIUM AT UCLA

Abstract Book

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Welcome to the annual CSU Summer Symposium and Graduate Program Fair at UCLA!

We are happy to have you join us for an afternoon featuring research presentations by students from CSU campuses, and information sessions on graduate training opportunities in STEM at UCLA. The aim of the Symposium and Fair is to promote scientific exchange and provide information on graduate educational opportunities as a way to strengthen interactions between CSU capstone research programs and UCLA graduate programs.

UCLA offers a wide variety of exceptional STEM graduate programs. The college, with Divisions of Life and Physical Sciences, is located on a single campus with the School of Medicine and hospital. This proximity provides a wealth of research training opportunities and promotes a collaborative and collegial culture of discovery and innovation that crosses traditional academic boundaries and spans disciplines, departments and schools. Our UCLA STEM community is built on core values of openness, inclusion, and respect that foster creativity and excellence by embracing a diversity of backgrounds, experience, ideas, and approaches.

We welcome CSU participants and extend thanks to our UCLA graduate program representatives.

Gregory S. Payne, Ph.D.
Director, Graduate Programs in Bioscience (GPB)
Senior Associate Dean of Bioscience Graduate and Postdoctoral Education, School of Medicine
Associate Dean of Graduate and Postdoctoral Education, Life Sciences

Diana Azurdia, Ph.D.
Director for Recruitment and Inclusion, Graduate Programs in Bioscience, UCLA

CSU SUMMER SYMPOSIUM AT UCLA

Schedule of the Day

1:00 – 1:20	Welcome
1:20 – 3:00	Graduate Program Tabling
2:00 – 3:00	Tabling continued with concurrent CSU Poster Session

Poster session will highlight CSU students planning to apply to UCLA who are participating in broadening participation capstone research programs such as MARC, IMSD, MBRS Rise, HMMI and others.

Light refreshments will be served.

1 Zoom Out of the Way

BREANA ACEITUNO, Alberto Guerra, Lauren E. Knox, Hyunbum Kim, Stefanie A. Drew

CSU Northridge, NIH MBRS RISE

With an increased use of virtual meetings due to the COVID-19 pandemic individuals have increasingly reported Zoom Fatigue symptoms (i.e. mental exhaustion, frustration, and irritability). Previous literature suggests that individuals who attend meetings in a virtual environment for extensive periods of time, have a difficult time staying focused;. Additional literature on environmental factors, such as household size, found that individuals from large household sizes tend to experience poorer academic performance. Studies on other environmental factors, such as lighting, found that the use of natural lighting enhances a students academic performance. However, little research has examined how physical environmental factors such as these relate to online learning or experiences in video conferencing meetings in general. As such, the current study sought to explore whether factors such as frequency of other individuals present during meetings, room of living space (i.e., kitchen, bedroom, office, etc.), and lighting during meetings are associated with Zoom Fatigue symptoms. We hypothesized that rooms that offered more privacy (i.e., bedrooms, offices, etc.) and having more privacy in general, in addition to more natural light, would relate to less Zoom Fatigue. Results of an ANOVA suggest that there were no significant relationships between room, lighting, and Zoom Fatigue. However, the relationship between how often other people were present during one's virtual meeting and Zoom Fatigue symptoms approached significance ($F(4, 420) = 2.191$, $p = .069$). These findings suggest that maintaining a private space during video meetings may be key to reducing Zoom Fatigue.

2 Purification of Full-Length NEMO and Structural Impacts of Conflicting Dimerization Sites

ADEN M. ALEMAYHU, Sally Luong, Matt Mealka, Tom Huxford

San Diego State University, NIH IMSD

NEMO is a subunit of the IKK kinase complex that is essential for the induction of transcription factor NF-κB via the canonical signaling pathway. It is a necessary component that is crucial to the activation of IKK. The function of the IKK multisubunit enzyme is to release NF-κB, a substantial transcription regulator involved in immune response, so that it may enter the nucleus to enable gene transcription. While much has been investigated regarding the structure and activities of the IKK complex, little is known about NEMO's structure and conformation as a dimer. My purpose with this project is to purify and produce full-length NEMO in order to conduct structural assays to determine the oligomeric states. While NEMO expresses a coiled-coil structure, there are four separate dimeric components of NEMO that may be competing for precedence in its overall conformation. To gain a better understanding of full-length NEMO's structure, we initially utilized digital structure generation through AlphaFold used in silico methods, before continuing with purification techniques. Once purifying the four dimeric pieces of NEMO, we incubated to full length to determine any conformational preference. In addition, we used HDX-MS both alone and in the presence of common NEMO-bound substances, such as ubiquitin, to look further into what is responsible for NEMO's dimerization tendencies. Following this, we sought to make two different tagged NEMO proteins and purify them together so that we could investigate the possible result of having three oligomeric states.

3 Role of miR-190 on Cell Fate Determination During Mitosis in Drosophila Neuroblasts

GERSON ASCENCIO, Cynnie Tam, Mathew de Cruz, and Blake Riggs

San Francisco State University, NIH Bridge to the Doctorate

Stem cells in the Drosophila brain, known as neuroblasts, generate most of the neurons in the brain by undergoing asymmetric cell division (ACD). ACD gives a product of two daughter cells with two separate identities facilitated by cell fate determinants such as aPKC, Prospero, and Numb. These determinants are partitioned asymmetrically to drive cell fate selection, with Prospero moving towards the basal cortex to promote cell differentiation. The mechanism of ACD is highly conserved across all multicellular organisms. However, the organization and regulation of partitioning cell fate determinants are poorly understood. microRNAs (miRNAs) regulate gene expression post-transcriptionally by silencing gene expression of targeted mRNA transcripts and play a role in regulating cellular functions such as development and differentiation. Here we hypothesize that miR-190 regulates Prospero during ACD. Our qPCR data highlighted Prospero, a transcription factor, as a potential target transcript for miR-190. We showed a shift from stemness to differentiation in neuroblasts and quantified the downstream effects of Prospero being derepressed under miRNA knockout conditions. Our data shows an increased Prospero expression which induces more differentiated progenitor cells of the neuroblast lineage. This data may suggest that miR-190 is a critical regulator in stem cell fate and differentiation and plays a vital role in maintaining the stem cell microenvironment. miRNAs have been discovered to play a role in neurogenerative diseases. Finding how mir-190 plays a role in ACD in Drosophila could lead us to discover new therapeutic agents for neurogenerative diseases.

4 Characterizing the Human Microglia Transcriptomic Response to Repeat Mild Traumatic Brain Injury (rmTBI)

NIMRAH ASHFAQ, Jonathan Hasselmann, Brian J. Cummings

CSU Fullerton, CSUF CIRM Bridges

Traumatic Brain Injury (TBI) is the occurrence of brain damage caused by an outside force, which results from events including car accidents, falls, sports injuries, and violent crimes. Of these injuries, mild traumatic brain injury causes concussions and concentration associated cognitive disfunctions, and its long-term effects are poorly understood. Microglia, the brain's immune cells, are critical in removing dying neurons, and supporting neuron survival, and their specific injury responses include migration to injury sites, and the release of inflammatory cytokines, making these cells viable in studying the extended effects of mild TBI. Currently, little is known about the human microglial response to repeat mild TBI. It is expected that human microglia will respond to injury resulting in dynamic changes in gene expression throughout the extension of the post-injury period. To examine these responses, human iPSC-derived microglia progenitors were transplanted into immune deficient neonatal MITRG mice, which are humanized for hCSF1, hCSF2, and thrombopoietin. Mice were aged for 8 weeks and given mild closed headed impacts every other day for a total of 5 hits. At 24 hours post-injury, human microglia were isolated from sham and rmTBI mice and underwent single cell RNA sequencing. Histological and transcriptomic analysis is ongoing for the 24-hour post-injury group. We expect a subpopulation of microglia that are unique within the rmTBI group, and that subpopulation is expected to express a distinct set of genes. Additionally, that microglia population and gene signature defining it are expected to change over time as the post-injury period progresses.

5 Extraction and purification of Pyocyanin

Katia Touahri, Catherine Grosdemange-Billiard, and Didier Lèévremont

CSU Dominguez Hills, NSF iREU

Pseudomonas aeruginosa is an aerobic Gram negative bacterium that commonly causes infections in immunocompromised individuals. It synthesizes pyocyanin, a redox-active secondary metabolite belonging to the phenazin group. This blue pigment, which possesses antibiotic activities against Gram positive bacteria is considered as a virulence factor involved in biofilm formation, electron shuttling, gene expression, and also as a quorum sensing signaling molecule. Disrupting bacterial virulence mechanisms e.g. pyocyanin production could be an attractive way to control P. aeruginosa. Since there are no experimental protocols published for identifying and quantifying pure pyocyanin, the aim of this project was to (1) establish an inexpensive and efficient protocol for the extraction and purification of pyocyanin, (2) Charaterize by nuclear magnetic resonance (H1, 13C, 2D experiments such as HMQC and MHMBC)

6 Structural Insight Into an Anti-tumor Antibody Bound to a MUC1 Glycopeptide

YAZMINE L. BEDOLLA, Angham Ahmed, and Cory L. Brooks

CSU Fresno, NSF LSAMP Undergraduate

While the cancer mortality rate in the United States continues to decline, cancer remains the second leading cause of death across the nation. Over the past five years, monoclonal antibodies have emerged as alternative pharmaceuticals and have been utilized as regimens for the treatment of cancer. In noncancerous cells, the glycoprotein mucin 1 (MUC1) serves as a physical barrier against invading pathogens and acts as a lubricant for epithelial cells. In malignant cells, however, MUC1 exhibits truncated and atypical glycosylation, leaving traditionally inaccessible epitopes exposed. Possessing high antigen affinity, tumor specificity, and an increased number of antibody binding sites, the MUC1 specific antibody, PankoMab, is a promising tool for antibody-based immunotherapies. We aim to elucidate the structure of PankoMab and its mechanisms of interaction with a MUC1 peptide to guide the future of antibody immunotherapies. To generate PankoMab Fab, the gene was cloned into the pcDNA 3.1 (+) vector and transiently transfected in Chinese Hamster Ovarian Cells (CHO cells). Protein expression was confirmed by SDS PAGE and purified using Nickel Affinity Chromatography. Purified PankoMab Fab will be complexed with previously purified MUC1 peptide and crystal screening will be carried out in 96 well plates. Once preliminary crystals appear, they will be optimized. X-ray crystallography will be performed on the antibody/antigen complex and diffraction data will be analyzed. Ultimately, a structural understanding of PankoMab Fab and its binding mechanisms to MUC1 may contribute towards the advancement of antibody-mediated immunotherapies.

7 Modeling Environmental Factors Effects on Valley Fever Incidence Rates

JACOB R. CARACCILO and Mario Banuelos

CSU Fresno, NSF LSAMP Undergraduate

Coccidiomycosis, also known as Valley Fever, is a disease caused and transmitted by the fungi Coccidioides immitis and Coccidioides posadasii. The disease affects the lungs and is endemic to arid regions within the southwest United States. While studies have been conducted to analyze disease severity and quantify risk factors, few focused on creating a mathematical model to study and quantify specific environmental parameters which affect transmission. Valley fever is not typically spread from person to person, so understanding environmental factors which affect transmission can help us better predict future occurrences of the disease. Using publicly available data from the last two decades, we construct mathematical models which consider factors such as air quality, water temperature, wildfires, and climate change in our study. Specifically, we use decision trees and deep learning to investigate patterns between those specific environmental parameters and case occurrences. With this research, our goals include better informed future predictions of case occurrences based on these environmental factors and identifying possible non-environmental factors which could serve as the basis of future studies.

8 Population Genetics of Two California Species of Checker Lily (Fritillaria)

Jacklyn Fajardo, Arshnoor Kaur, ANDREW CARDENAS, Chris Winchell, Katherine Waselkov

CSU Fresno, NIH MBRS RISE

Our project focuses on the genetics of two species of wildflowers called checker lilies, Fritillaria atropurpurea and F. pinetorum. Fritillaria is a genus that consists of approximately 140 perennial plants and is known for its huge genome (30-80 Gb of DNA). Two local California species of checker lily wildflowers, Fritillaria atropurpurea and F. pinetorum, are morphologically very similar but are currently listed as two different species. Aside from slight and inconsistent morphological differences, both species are generally found in different but contiguous parts of California. F. pinetorum is listed as rare by the California Fish and Wildlife Society. This research focuses collecting data about whether F. atropurpurea and F. pinetorum are the same or different species by utilizing the chloroplast DNA via DNA extractions, PCR, and gel electrophoresis. I have tested out a few different primers, and the results so far show that primers for the chloroplast region trnH-psbA amplify well in both species. Those DNA sequences were cleaned and then sent to Eurofins Genomics for Sanger sequencing. The sequencing alignment showed an indel of (TCTTA) from #352-357 bp for one population of F. atropurpurea, with the same insertion in three F. pinetorum populations. However, the insertion was not observed in 3 other populations of F. atropurpurea. The implications of this work could affect conservation decisions involving F. pinetorum: if these two species cannot be differentiated genetically, then it would imply that they are still exchanging genes and would not be two different species according to the Biological Species Concept.

9 Identification of Putatively Novel Natural Products from a Marine Derived Fusicolla sp. of Fungi

EDWIN O. CHAVEZ SANTANA, Nathan W. Williams, Ebonie Bennett, Jorge Hernandez Garcia, Shaz Sutherland, Erin McCauley

CSU Dominguez Hills, NIH MBRS RISE

Natural products (NP) play an important role in pharmaceutical drug development, they are secondary metabolites produced from living organisms and over 65% of all FDA approved pharmaceuticals are NPs, NP-derivatives, or their pharmacophore was inspired by NP chemical scaffolds. One of the major bottlenecks in NP research is the identification of previously known NPs. The objective of this research is to utilize an innovative tandem mass spectrometry (MS/MS) database called the Global Natural Products Social Molecular Networking (GNPS) platform to identify NP with novel chemical scaffolds. This research was initiated by culturing over 50 fungal strains and extracting the NP they produce. The extracts were analyzed in a MS/MS format so they could be run on the GNPS platform, there the MS/MS spectra were compared to a massive database of MS/MS spectra from known NP. Extracts with metabolites that showed minimal or no similarity to known NP in the database were prioritized for further study. One of these was an extract from a Fusicolla sp. of fungi. These putatively NP were purified using high-performance liquid chromatography and their structures were determined using MS in addition to 1D and 2D NMR spectroscopy.

10 Understanding The Experience Of Black Women During Prenatal Care In Minoritized Communities

AYSA M. COLLINS, Enrique Ortega

CSU Dominguez Hills, McNair Scholars

In recent years, the United States has seen an overall increase in maternal morbidity and mortality rates, notably in underrepresented populations. Non-Hispanic Black women are at the highest risk for maternal morbidity and mortality outcomes compared to other races and ethnicities. In 2019, the maternal mortality rate for non-Hispanic black women was 44.0 deaths per 100,000 live births. This was 3.5 times higher than the rate for Hispanic women and 2.5 times higher than the rate for non-Hispanic white women. The most recent research on maternal morbidity and mortality have shown that early and adequate prenatal care can considerably reduce the risk of preterm birth and maternal morbidity. Nonetheless, when examining factors contributing to poor maternal outcomes among non-Hispanic black women, education, and income alone do not correlate with the increases in poor maternal outcomes. Investigations have found that prenatal care alone does not account for the poor maternal morbidity and mortality outcomes reported among non-Hispanic black women. This study examines the five major risk factors for maternal morbidity and mortality associated with fetal deaths, the month prenatal care began, and patient demographics in California from 2014 to 2019 using the Centers for Disease Control and Prevention, National Vital Statistics System, CDC WONDER Online Database. This research will work to identify risk factors associated with prenatal care and maternal outcomes among non-Hispanic Black women. These outcomes include early evaluation of mothers for medical risk and the need for psychosocial treatment, cultural barriers, and educational resources.

11 Key Intermediate for Synthesis of Bis-Piperidine Alkaloids as Therapeutic Anticancer Treatment

KHAMYL T COOKSEY, Jasmine Hang, Qiao-Hong Chen, Erik J. Sorensen

CSU Fresno, NIH MBRS RISE

Due to their unique chemical structures, bioactivities, stereochemistry, and relative size for a significant class of alkaloids, bis-piperidine alkaloids must be explored. Isolated explicitly from marine sponges, these centralized 3,4'-linked bis-piperidine moieties adjoined with two aliphatic macrocycles have been demonstrated to display antiproliferative potency to various cancers. The proposition scheme of a library of bis-piperidine alkaloids and their evaluation of anti-cancerous potency was inspired by the intriguing integrated findings of novel research partially investigated. Through a developed eight-step subsequent scheme, the intermediate aims to be prepared through various optimized steps followed by Suzuki coupling for our small library of tetracyclic bis-piperidine alkaloids. Each subsequent compound for syntheses were characterized by their unique chemical structure displayed by their magnetic resonance spectroscopic shifts and infrared spectroscopic data.

12 Synthesis of a library of potential inhibitors of slc26a3 based on the thiazolo-pyrimidin-5-one scaffold

DANIEL CORNEJO, Ashley N. Welch, Marc O. Anderson

San Francisco State, Genentech scholarship recipient

Symptoms of constipation are extremely common; the prevalence is approximately 16% in adults overall and 33% in adults older than 60 years of age. Some of the problems caused by constipation are abdominal pain and extreme cases may lead to hemorrhoids. There are five different forms of laxatives, which relieve constipation, including bulk-forming laxatives, osmotic laxatives, stimulant laxatives, stool softeners, and rectal suppositories. Current forms of laxative medication function in a few different ways, one of the common ones is "bulk-forming" laxatives, which are high fiber compounds that absorb water and makes the stool larger causing bowel movement. These forms of treatment, like most treatments come with a few side effects which may be uncomfortable. Some of the side effects of laxatives include bloating, cramping, and dehydration. The blocking and opening of the chloride anion channel lead to various physiological consequences. A specific chloride transporter protein Slc23a3 is expressed in the large intestine. Through experimentation, it was seen that by inhibiting Slc26a3, mice were found to have induced diarrhea. This led to the hypothesis that Slc26a3 could be a novel drug target for the treatment of constipation. Through High-throughput screening (HTS), some molecules were shown to inhibit the function of Slc26a3. A specific scaffold (thiazolo-pyrimidin-5-one), was found to have an IC50 of 360 nM. By using medicinal and synthetic chemistry to optimize the substituents in this scaffold, we hope to (a) improve the potency of the inhibitor and (b) optimize drug-like features such as aqueous solubility and metabolic stability.

13 Design, Development and Computational Studies of Gemcitabine Analogues for the Treatment of Pancreatic Cancer

ALEKSYA K. DROBSHOFF, Ahmed. M. Awad

CSU Channel Islands, NSF LSAMP Undergraduate

Pancreatic cancer is the fourth leading cause of death in the United States. The dismal survival rate of pancreatic cancer is due to late detection at an advanced stage in the pancreas. The standard treatment at this late stage is chemotherapy with the drug Gemcitabine. The metabolism of Gemcitabine involves phosphorylation by deoxycytidine kinase to its diphosphate and triphosphate forms. The triphosphate form of Gemcitabine inhibits DNA polymerases by incorporation into DNA. The diphosphate form binds within the catalytic site of the enzyme ribonucleotide reductase (RNR) and irreversibly inhibits its activity by disrupting the catalytic pathway. Our research project involves the design and development of Gemcitabine analogues, the most common and effective modified nucleoside used in the chemotherapy treatment of pancreatic cancer. Our current and most tested modification made to Gemcitabine was adding a Poly Ethylene Glycol Amino chain to the 2' position on cytidine. This modification was proposed to act as a chelating site for the zinc ion, the metal ion required for the RNR function, which leads to a decrease in its activity. The previously tested enzymatic-target binding affinity and positive correlation to anticancer and antimicrobial properties gives the PEGA-nucleoside the potential to serve as a strong RNR inhibitor. We tested this molecule through a series of computational docking studies to test the binding affinity of our molecule with RNR. The results we have achieved demonstrate the positive potential of the PEGA modification as an anticancer nucleoside analogues leading to our transition into performing synthesis in the lab.

14 Arm-First Polymerization of Acrylate- and Acrylamide-Based Star Polymers Using RAFT

MELISSA S. GRIFFIN, Melanie Gonzalez and Madalyn R. Radlauer

San Jose State University, NIH MBRS RISE

Our lab is interested in understanding how a structured polymer scaffold directly bound to a transition metal catalyst can alter the activity of that catalyst compared to a free catalyst, specifically through a variety of secondary interactions provided by the polymer. Our approach mimics metalloenzyme structure by encasing a metal complex as an "active site" within a larger framework. This project focuses on the construction of the polymer scaffold, making star polymers with an arm-first method. We use reversible addition-fragmentation chain transfer (RAFT) polymerization to synthesize the linear polymer "arms" - that form the corona of the star polymer - and then covalently connect them through a reinitiation of the polymerization with cross-linkers as comonomers - to form the core of the star polymer. We chose RAFT because it is a controlled radical polymerization that tolerates a range of functionalized monomers. Our group started by examining styrenic star polymers and we are currently exploring star polymers with other monomers, particularly acrylates and acrylamides.We are investigating the significance of the arm lengths of the stars, the choice of cross-linker, and the solvent type used for the reaction. Characterization of these star polymers is done using NMR spectroscopy to determine linear polymer size by end-group analysis as well as by GPC to determine relative molecular weight and dispersity of our linear and star polymers.

15 Synthesis of Iodoenamine Precursor for Tetracyclic Bis-piperidine Alkaloids for Cancer Treatment

JASMINE HANG, Qiao-Hong Chen, and Erik J. Sorensen

CSU Fresno, NIH MBRS RISE

The American Cancer Society found that cancer is one of the leading causes of death. With high numbers of deaths per year, there are demands for more effective treatments. Natural products as a source of medicine have long been studied, especially for cancer treatment. Tetracyclic bis-piperidine alkaloids are derived from marine sponges and are characteristic for their unique chemical structures and potential to suppress cancer cell proliferation in three cancer cell models. However, the structure-activity relationship and in vivo animal studies have yet to be studied due to limited availability. The focus of this two month study was the creation of iodoenamine through a five step synthesis. This is the starting point of a long-term project to form a library of structurally diverse tetracyclic bis-piperidine alkaloids. The first three steps of the synthesis have been achieved, with each intermediate characterized using ^{1}H NMR and ^{13}C NMR. Due to poor yield and the volatility of one intermediate, several procedures were explored to improve the synthesis. The synthesis of iodoenamine is still in progress.

16 Investigating the Interaction of Kaposi's Sarcoma-Associated Herpesvirus Viral G Protein-Coupled Receptor and Angiogenic Proteins

EDUARDO HERNANDEZ, Jan Mikhale Cajulao, and Erica L. Sanchez

San Francisco State University, Genentech Foundation fellowship

Kaposi's sarcoma-associated herpesvirus (KSHV) is a cancer-causing virus and the causative agent for Kaposi's sarcoma (KS). KS is the most frequent type of cancer in males and children in the Mediterranean and sub-Saharan African countries. The virus encodes a constitutively active signaling molecule called the viral G protein-coupled receptor (vGPCR) which is critical for the initiation and progression of KS. KSHV establishes lifelong infection of the human host and currently there is no treatment. A previous study's proteomics screen revealed that vGPCR is suggested to interact with many human proteins. Among the interactions associated with KSHV are two understudied angiogenic proteins called endoplasmic reticulum membrane complex subunit 10 (EMC10) and mannoside acetylglucosaminyltransferase 5 (MGAT5). However, these vGPCR interactions with host proteins and the downstream implications have yet to be explored. This study aims to investigate the potential interaction between vGPCR and these specific angiogenic proteins. I hypothesize that KSHV vGPCR interacts with EMC10 and MGAT5, and that these interactions modulate angiogenesis. Immunofluorescence assays and western blot analysis were used to analyze the angiogenic protein expression in KSHV vGPCR-expressing cells. We successfully visualized EMC10 by IFA and detected it in vGPCR-expressing cells via western blot. Co-immunoprecipitation will also be utilized to validate the interactions. This study will add new insight into angiogenic proteins' effects on vGPCR signaling and roles of the understudied proteins EMC10 and MGAT5. Also, this study will provide information into understanding viral-host protein interactions affecting the modulation of angiogenesis and can lead to therapeutic targets for KSHV infection.

17 Identification of Novel Natural Products from a Marine Derived Geotrichum silvicola Fungal Strain

JORGE HERNANDEZ GARCIA, Edwin Chavez Santana, Shaz Sutherland, Nathan Williams, Erin McCauley

CSU Dominguez Hills, NIH MBRS RISE

The overall objective of this research was to identify novel natural products from marine derived fungi. To achieve this, over 50 taxonomically unique fungal strains were grown in five different media types. The natural products they produced were extracted and analyzed using liquid chromatography-mass spectrometry in a tandem (MS/MS) format. The MS-MS data was analyzed using the Global Natural Products Social Molecular Networking (GNPS) platform. The Spectral Library Search tool within the GNPS platform was used to compare the MS/MS data to a massive library of MS/MS spectra generated from structurally characterized metabolites. Extracts that had low similarity scores to metabolites in the GNPS library were hypothesized to contain putatively novel chemical scaffolds and were selected for further study. This led to the identification of a series of novel alkaloid natural products from a strain of Geotrichum silvicola. These compounds where purified from the crude G. silvicola using high performance liquid chromatography, and the structures were determined using high accuracy MS, 1D and 2D NMR spectroscopy, and circular dichroism spectroscopy experiments.

18 Achievement guilt and well-being among first-generation Latinx college students: Does familism influence this relationship?

SUMMER J. HERRERA and Rosa Toro

CSU Fresno, NIH MBRS RISE

Numerous first-generation college students (FGCS) experience a sense of achievement guilt, feeling responsible for leaving their family behind to pursue a greater opportunity than the rest of their family, throughout their college life. Due to the impact of the transition to college, achievement guilt is associated with increased depression and anxiety amongst FGCS. Current literature has studied the influence of the cultural value of familism, having a strong and supportive relationship with family members, on first-generation college students and its role in reducing depression and anxiety. It has been shown to help reduce depression and anxiety in students, which occur when having achievement guilt. The purpose of this study is to get a better understanding of the relationship between achievement guilt and well-being and the moderating influence of familism. It is expected that achievement guilt will be positively associated with stress, depression and anxiety. Also, when students have strong familism values, it will buffer the impact of achievement guilt. Participants included 256 (75% females) Latinx college students and completed an online survey. Preliminary results show emerging support for the proposed hypotheses. First, regression analyses indicated that achievement guilt was positively associated with stress, depression, and anxiety. Similarly, regression analyses indicated that familism was negatively associated with stress, depression, and anxiety. But moderation analyses indicated that familism did not act as a moderator. The findings bring awareness to the difficulties Latinx college students face when transitioning to college and the importance of providing resources to support this student population.

19 Exploring the Effects of Herbivory on Competition between Foeniculum vulgare and Two California Native Herbs, Eschscholzia californica and Achillea millefolium

SAMANTHA P. HUBBARD, Nicholas J. Torres, Joel K. Abraham

CSU Fullerton, Ronald E. McNair Scholars Program

Allelopathic species, or species that produce chemicals to inhibit competing species' growth, are prominent invaders, and the use of herbivory to control them may lead to higher production of these harmful chemicals. Foeniculum vulgare (fennel) is a notorious allelopathic invader in California, competing mainly with California poppy and California yarrow, so we want to observe how herbivory may affect these species when grown in direct competition. Upon putting these California natives into direct competition with fennel that has undergone herbivory, they should grow to have lower biomass and overall height than those paired with fennel that has been left alone. The plants were put into either intraspecific competition pots as controls or interspecific competition pots as the experimentals. As time went on, some plants didn't grow or some pots had excess of one species, and were removed. After this, herbivory was simulated on the plants. At the end of the experiment, the plants will be dried and their overall biomass will be measured to determine final effects that herbivory on fennel had on the natives grown with them. As of this point in time, data is being collected, though no clear patterns have emerged. As the experiment goes on and more data is collected and analyzed, the results will point in a more clear direction as to what herbivory on fennel suggests for the natives in direct competition with them.

20 Characterizing the Localization of SPD-1, and its Role as a Microtubule Bundler during Sperm Meiosis in Caenorhabditis elegans Males.

CUC M. HUYNH, and Diana S. Chu

San Francisco State University, NIH U-RISE

While male infertility brings significant challenges to human reproduction, little is known about the molecular mechanisms that drive male meiotic chromosome segregation required for efficiently generating healthy sperm. During mitosis and oocyte meiosis, chromosomes segregate by the pushing forces from the central spindle structure. The central spindle forms in the midzone area during anaphase after SPD-1 protein bundles microtubules while recruiting other midzone proteins. In Caenorhabditis elegans nematode, there is a unique unpaired lagging X chromosome present in males and interestingly, the central spindle formation is minimized during sperm meiosis. This led to the question of whether sperm meiosis relies on the central spindle for proper chromosome segregation due to the presence of the lagging unpaired X chromosome. Utilizing immunostaining and live confocal microscopy, I aim to determine the spatial-temporal localization of SPD-1 during anaphase of sperm meiosis. Results show that SPD-1 localizes and remains at the midzone region as the unpaired lagging X chromosome travels to one polar end during sperm meiosis. This suggests that SPD-1 stabilizes microtubule organization which is essential for accurate chromosome segregation during sperm meiosis. In addition, its interaction with other midzone proteins could help promote the completion of cell cleavage and regulate the final cell division process, cytokinesis. Taken together, this research will elucidate SPD-1 functions during sperm meiosis, especially whether it's involved in forming the central spindle which can further reveal the factors that cause male infertility.

21 The Extent of Hydrophobic Recovery on Nitrogen and Water Vapor Plasma Treated Silk Films

ASHLEY N. KEOBOUNNAM, Chase A. Lenert-Mondou, and Morgan J. Hawker

CSU Fresno, NIH MBRS RISE

Silk, derived from the bombyx mori cocoon, is a natural polymer that is widely studied for its mechanical strength, biodegradability, and non-immunogenic properties in biomedical applications. However, inducing specific interactions between silk and cells in vivo is challenging because of its hydrophobic nature. Therefore, modification must be performed to increase surface hydrophilicity. Plasma modification is a polymer modification method that is simple, economical, and solvent-free. Nitrogen and water vapor are both recognized plasma modifications that increase polymer surface polar functional groups. However, plasma-modified polymers often undergo hydrophobic recovery: rearrangement of modified polymer chains from a higher surface energy state to a lower one. This results in negating the plasma modification effects. The hydrophobic recovery of plasma-modified silk has not been previously studied. The goal of this study was to systematically evaluate hydrophobic recovery of nitrogen and water vapor plasma-treated silk films. Films were prepared by dropcasting. After drying, films were plasma modified using optimized plasma parameters. Interactions between plasma-treated silk films and water were examined using water contact angle goniometry. Untreated silk films displayed water contact angles of 69+/-1° while the nitrogen and water vapor plasma-treated films exhibited water contact angles of 35+/-1° and 25+/-2°, respectively, demonstrating a decrease in hydrophobicity. Treated films were aged up to 6 weeks under ambient conditions and analyzed after aging for 7 days, 21 days, and 42 days. Nitrogen and water vapor plasma-treated silk film water contact angles remained consistent throughout the 42 day aging period, suggesting minimal hydrophobic recovery occurred.

22 Comparing the Contrasting Ground States of Pyrethroids and Pyrethrins

KELLY KEPLER, Sean Duncan, and Kristi Closser

CSU Fresno, NIH MBRS RISE

Pyrethroid insecticides are synthetically derived based on the structure of pyrethrin compounds found in chrysanthemum flowers. Both pyrethroids and pyrethrins function as nervous system disruptors on small insects, however pyrethroids are known to exhibit significantly increased stability in contrast to pyrethrins. Pyrethroids are commonly used in both residential and agricultural pesticide management, remaining pervasive in water and soil ecosystems after application. Degradation products for pyrethroids are not well defined and identification could enable more targeted approaches in defining the concentrations of these eco-toxins, therefore a comparison with the lower stability pyrethrins was conducted. We studied the ground state conformation of pyrethroids, Lambda-Cyhalothrin and Cypermethrin, and pyrethrins, Cinerin II and Jasmolin II, using Density Functional Theory. Low energy structures have been isolated using B3LYP with increasing basis sets and accounting for environmental effects through the polarizable continuum model for solvation. Using these structures, the electron orbitals and absorption spectra were compared to determine potential differences in reactivity. Future work will focus on the determination of possible photodegradation pathways based on the excited state electronic structure to illustrate the aforementioned compound's contrasting stability.

23 Fluorescence Spectroscopic Analysis of Apolipoprotein AI Reconstituted High Density Lipoprotein

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High density lipoproteins (HDL) are protein-lipid complexes that aid in cholesterol efflux, a process in which HDL particles interact with ATP-binding cassette transporters (ABC) A1 and ABCG1 to remove excess cholesterol from macrophages. The major protein on HDL, apolipoprotein AI (apoAI), exists in lipid-free and lipid-bound states. When bound to phospholipids, it contains 10 a-helices (H1-H10) that wrap around the hydrophobic lipid tails. Our goal is to understand how the conformation of apoAI changes during cholesterol efflux. We hypothesize that residues 125-158 (helices H5 and H6) form a disordered loop that can accommodate lipid loading and changes in HDL particle size. To test this hypothesis, an apoAI double-cysteine mutant (L134C/A152C) was designed with cysteines positioned on the loop. The purified protein was labeled with N-(1-pyrene)-maleimide (NPM), a spatially sensitive fluorophore that has a distinct emission at ~460 nm when it is within ~10Å of a neighboring pyrene. The pyrene-labeled apoAI was reconstituted with phospholipids at different phospholipid:protein molar ratios (28:1, 70:1, and 100:1) to generate small (~7.8 nm), medium (~9.6 nm) and large (~10.5 nm) diameter HDL referred to as reconstituted HDL (rHDL). The pyrene-labeled rHDL was incubated with J774.1 macrophages undergoing cholesterol efflux, and fluorescence spectra compared before and after efflux. In the rHDL-bound state, apoAI undergoes conformational reorganization with a significant decrease in excimer emission. The data suggests that the conformational changes accommodate for lipid loading, but more studies are required to obtain further details about the conformational reorganization during lipid loading of HDL.

24 Image Recognition : Face Masks Detection

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During COVID-19, face masks prevent the spread of the virus the most. People often get COVID-19 because they don't wear masks, and it has increased the speed of spreading the virus. This project can check whether a mask is worn through an image. In particular, it is expected to be helpful in many places where there are many elderly and young children who are vulnerable to the virus. Dr. Chen (Mentor) and I intend to implement a computer vision system to detect whether or not a face mask is worn using publicly available image data through convolutional neural networks (CNNs) and VGG16. The focus is on statistically improving accuracy of captured images with face masks using machine learning. The project is carried out because it needs to minimize the spread of viruses necessary for public health.

25 Luminex Multiplex Assay for Bead-Based Detection of Rheumatoid Arthritis-associated Autoantibodies

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Immune checkpoint inhibitors (ICI) have been shown to improve treatment of various advanced cancers but can result in immune-related adverse events (irAE). ICI arthritis (ICI-A) is one such irAE that imitates rheumatoid arthritis (RA) in many ways. One characterization of clinical RA is testing anti-citrullinated protein antibody (ACPA) positive. ACPA epitope expansion occurs prior to the onset of clinical RA. This study examines and compares the degree of said epitope expansion in ACPA+ RA and ICI-A patients. Collaborators at the Hospital for Special Surgery (HSS) and Weill Cornell Medical College performed an enzyme-linked immunosorbent assay (ELISA) to distinguish ACPA positive and negative sera samples. ICI-A, early RA, and late RA samples were run on an 18-protein (51-peptide) ACPA panel using the Luminex FLEXMAP3D Instrument. The FLEXMAP3D instrument is Luminex's most advanced and versatile multiplexing platform and through the use of MagPlex-Avidin Microspheres, up to 500 analytes can be detected in a single assay. Samples were incubated with MagPlex-Avidin beads conjugated to the ACPA peptides via an N-terminal Avi-tagged Biotin prior to being run on the FLEXMAP3D instrument. Lower ACPA intensities and epitopes were observed in ICI-A patient serum compared to both early and late RA patient serum. ICI-A may represent an accelerated model of RA pathogenesis with ICI triggering an early transition into clinical disease. Through the comparison of the number of ACPA epitopes targeted in ACPA+ ICI-A and RA serum, this study brings us closer to earlier diagnosis, intervention, and possible prevention of RA.

26 How Do the MyoD Binding Regions Upstream and Downstream of the Acta1 Gene Influence Transcription of α-actin RNA?

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The transcription factor MyoD contributes to myogenesis (muscle development) by binding to conserved regulatory sequences typically found upstream of target genes. Interestingly, binding sites for MyoD have been found downstream as well as upstream of Acta1, one of MyoD's target genes. The role of the downstream binding regions is unknown. I hypothesize that MyoD binding both upstream and downstream of Acta1 is necessary for developmentally regulated, muscle-specific Acta1 transcription. Testing this hypothesis will increase our knowledge of longer-distanced, downstream transcriptional regulation, and will provide regulatory mutations to test for in ACTA1-related myopathies that are not yet ascribed to a coding-region mutation. I am performing transient plasmid transfections with normal and mutated Acta1 regulatory regions inserted upstream and downstream of a firefly luciferase reporter gene. The constructs are transfected into mouse pre-muscle and fibroblast cells, and a dual luciferase assay is performed at both myoblast (undifferentiated) and myocyte (differentiated) stages. I have determined the quantity of normalizer NanoLuc plasmid to use; have found that apparent NanoLuc activity is lower in growth versus differentiation conditions; and have observed an apparent effect of the specific plasmids present in the transfection mixture on NanoLuc activity. I address how these findings will inform in our data analyses. Normalized Acta1 promoter-driven firefly luciferase expression is higher in the presence of the upstream MyoD binding region than in its absence in myocytes, but not in myoblasts, consistent with previous reports. Procedures using additional constructs in both fibroblast and pre-muscle cells are underway. (Funded by RISE NIH-R25 GM061331)

27 Flat and Fearsome: a Newly Discovered Carnivorous "Palm Frond Sponge" from the Deep Northeast Pacific Ocean

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The deep ocean is a vast frontier, largely unexplored, with new species yet to be identified and described by science, including many sponges (Porifera). Describing and classifying sponges can be challenging because their morphology is often quite variable. Sponges are typically classified into taxonomic groups using morphology, careful analysis of spicules (hard skeletal elements made of silica or calcium carbonate), and, more recently, molecular techniques. This research aims to taxonomically classify the recently discovered "Palm Frond Sponge", a leaf-shaped carnivorous sponge. We collected and preserved 18 specimens from deep waters ranging from central California to northern Canada. We identified and measured 5 spicule types including (n=50 for each type): small and large oxeas (length: 71-288-1136 µm; width: 0.91-11-29 µm; min-mean-max), strongyles (length: 645-655-664 µm; width: 10-10-10 µm), forceps (32-53-62 µm), and palmate anisochelae (18-20-24 µm). Amphipods and other small crustaceans were observed within the sponge tissue, suggesting active carnivory at the time of collection. The suite of spicules identified confirm that this species is a Cladorhizidae (the family of carnivorous sponges). Our goal for this project is taxonomic identification and formal description with assignment of a scientific name if it is confirmed to be a new species, which we believe it is. Ultimately, this unknown species will contribute to a clearer picture of evolutionary relationships within the Cladorhizidae and will be the foundation of further investigations of taxonomic, evolutionary, and ecological relationships within both its family and genus.

28 Bioengineering a Scaffold for a Myocardial Fibrosis Model

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According to the American Heart Association, heart disease remains the number one leading cause of death in the United States with more than one person having a heart attack every minute. Collagenous scar tissue formation that occurs after a heart attack does not have the functional phenotypic characteristics of myocardial tissue leading to dysregulation of ventricular filling, electrical homogeneity, and oxygen diffusion within the myocardium ultimately leading to heart failure. The objective of the study is to construct a myocardial fibrosis model by investigating methods to modulate fibroblast proliferation and myofibroblast activation. To prepare the engineered scaffold, fibroblasts were seeded within 2.5mg/mL of Rat Tail Collagen Type I gel. Cell viability was observed on polydimethylsiloxane (PDMS) molds with and without grooves. Cells were stained using immunostaining of F-actin and α-smooth muscle actin. Quantitative polymerase reaction (qPCR) was used to detect GAPDH, α-smooth muscle actin, and cyclin D1. Preliminary data presents significant cell viability and proliferation when fibroblasts were cultured within a Collagen Type I gel in a PDMS substrate without grooves. Results suggests that fibroblast activation and proliferation can be influenced through different micro-topographical environments. A model of fibrosis progression can demonstrate a promise for the development of therapeutic treatments through the modulation of scar tissues to support cardiac repair and prevent heart failure.

29 Identification of Putatively Novel Natural Products from a Marine Derived Fusicolla sp. of Fungi

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Natural products are secondary metabolites produced by living organisms. They play an important role in the current medical system accounting for over 65% of all FDA approved pharmaceuticals. The overall goal of this research is to identify fungal natural products with novel chemical scaffolds they can be screened in various biological assays. To initiate this research, over 50 taxonomically unique fungal strains were grown in different media types and the metabolites they produced were extracted. The extracts were analyzed in a liquid chromatography (LC)-tandem mass spectrometry (MS/MS) format so they could be run on the Global Natural Products Social Molecular Networking (GNPS) platform. The GNPS platform contains a massive database of MS/MS spectra from known natural products. The MS/MS spectra of the fungal extracts was compared to spectra in the database using the Spectral Similarity function of the platform. Extracts with metabolites that showed minimal or no similarity to known compounds in the database were prioritized for further study. One of these was an extract from an Fusicolla sp. of fungi. These putatively novel compounds were purified using high-performance liquid chromatography and their structures were determined using MS in addition to 1D and 2D NMR spectroscopy.

30 Identifying Genomic Regions in Rockfish Associated with Speciation and Ecological Divergence

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Marine rockfishes (Sebastes) are part of an adaptive radiation, making this genus a unique model to study speciation mechanisms in temperate fishes. Rapid speciation in rockfish has resulted in high morphological, and ecological diversity. A lack of geographic isolation among closely related rockfish species points to ecological divergence (along a depth gradient) as a potential driver of speciation. In this study we aim to identify genomic regions associated with depth divergence. We will focus on the closely related sister species, S. chlorostictus and S. rosenblatti. Their geographic range spans from Washington to Baja California; though, in terms of depth, S. chlorostictus populates the epibenthic region (60-240 m) and S. rosenblatti is found deeper in the mesobenthic region (100-490 m). We compared the distinct sister taxa through whole genome sequencing and performed a genome-wide analysis. We used next-generation sequencing (NGS) for both species (S. chlorostictus and S. rosenblatti) and conducted several analyses, including admixture analysis, principal component analysis (PCA), and genome-wide differentiation (Fst). PCA was done with 15,738 single nucleotide polymorphisms (SNPs) and resulted in distinct clusters, demonstrating the two species are reproductively isolated and there is no hybridization or admixture. A genome-wide scan for differentiation (Fst) found a large region on chromosome 18 which we are further characterizing to identify the function of genes in this region. By comparing closely related species of rockfishes that are ecologically divergent, we can provide a deeper understanding of diversification mechanisms of rockfishes and other marine organisms.

31 Innate Immune Protein C1q Modulation of Endothelial Wound Healing

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Atherosclerosis is a chronic inflammatory disease that is very common and is a major cause of death in the USA. This disease causes damage to the endothelium that lines blood vessels and leads to infiltration of monocytes/macrophages and low density lipoproteins (LDL) into the arterial wall. LDL can then become oxidized also known as oxLDL, which is pro-inflammatory. Innate immune protein C1q is known to be produced by macrophages in atherosclerotic lesions, and binds oxLDL. C1q has beneficial effects on macrophage functions but the effect of C1q on endothelial functions in atherosclerosis is not well known. The aim of our study was to test C1q modulation of endothelial migration. We tested the hypothesis that C1q bound to oxLDL will increase endothelial wound healing compared to oxLDL alone. To test this, a wound healing assay was performed. A wound was generated in a confluent culture of human aortic endothelial cells (HAEC) using 3-well culture inserts and a sterile pipette. Cells were treated with oxLDL with and without C1q in 1% serum media and monitored for 24-48 hours using live cell imaging. 10% serum was used as a positive control. The average width of the gap was calculated at different time points for each treatment. Data showed that the presence of C1q substantially increased wound healing, even above levels seen in our positive control. These data suggest C1q may have a beneficial effect on the damaged endothelium in atherosclerosis.

32 Optimization of HIV-1 p17 Expression in E. coli

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The objective of this research is to understand the interaction between heparan sulfate proteoglycans, found on the surface of human cells, and the HIV-1 p17 protein. Heparan sulfate receptors are known to influence growth factor signaling, cell adhesion, and enzymatic catalysis on human cells. Thus, this interaction may be one of the deciding factors for disease progression in humans infected with HIV-1. In order to understand the interaction between the HIV-1 p17 and heparan sulfate, the HIV-1 p17 protein needs to be expressed and isotopically labeled for characterization with nuclear magnetic resonance (NMR). A construct using a pET-16b plasmid was used and the expression with this construct was optimized as follows. Transformed E. coli was grown in M9 media and monitored via UV-vis spectrophotometry. The E. coli cells were harvested and lysed to extract the soluble protein and the HIV-1 p17 was isolated from the other soluble proteins via affinity chromatography. Gel electrophoresis was used to monitor samples taken throughout the expression protocol to optimize the yield of purified protein. The growth conditions and purification conditions were systematically modified to maximize protein yield and develop reliable methods for separation of the monomer and trimer forms of HIV-1 p17. In this stage of the project the Arixtra penta-saccharide is used as a heparan sulfate model. Future NMR studies will provide useful information on interactions that HIV-1 p17 has with the immune system of the animal's cells to deregulate their functions.

33 Are You the Culprit?: The Impact of Workplace Incivility on Black LGBTQ Members

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There is little research on how workplace incivility impacts Black LGBTQ individuals. The goal of this study is to investigate how workplace incivility affects stress levels of Black LGBTQ members and their likelihood of reporting such incidents to human resources. The negative stereotypes surrounding their Black identity, gender, and/or sexuality can affect their rates and reasons for reporting such that they will be less willing to report these negative experiences to organizational authorities due to backlash. Data collection has been completed for a sample of 125 Black LGBTQ workers residing in the United States. Participants completed a survey about their experiences with various types of workplace incivility, reporting of these acts, and their levels of stress within the last month. Quantitative analyses reveal a positive correlation between workplace incivility and perceived stress. Qualitative responses for reporting incivility and evaluation of the reporting process will be analyzed for Black women, gay Black men, and Black bisexuals specifically. False and harmful stereotypes can be a reason why Black queer people will be less likely to report their experiences with workplace incivility. The idea of the angry Black woman, intersectional fears about predation from gay men and criminality for Black men, and promiscuity for Black bisexuals may explain why these groups either avoid reporting incivility or believe these stereotypes disrupt the process. Investigating this overlooked population will bring awareness to their experienced stress and drive further interest in analyzing how these negative effects accumulate over time and spill over into non-work domains.

34 Oil Slick Detection on Oceanic Surfaces Using Hydrocarbon Remote Sensing

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The use of petroleum and oil products in the United States alone amounts to roughly 19.7 million barrels of oil per day. With this large volume of material being extracted, refined, transported, refined, and utilized by the end user, the chances of oil spills due to human error, faulty equipment, or natural disasters are extremely high. Early detection of oils spills are critical to minimizing their adverse environmental impacts. Numerous detection methods are currently in use but have drawbacks that limit their use on a large-scale 24 hour basis. The ideal option is remote sensing, whereby an optical system monitors the ocean surface and triggers an alarm when oil is detected. In this work, we propose to develop a fluorescence based method. Using a telescope and a photodiode, we are able to receive optical signals from up to 60 meters away from the light source. Varying levels of light intensity are experimentally generated and compared to the results of computer simulations. Multiple outputs of light intensity are generated and independently controlled, demonstrating that fluorescence occurs. The intent for this project is to a develop a method of early detection tht would lead to the development of sensitive, dedicated (and lower cost) oil sensors and to future patent application and/or funding proposals to support that work.

35 Removal of endotoxins from 2G8 antibodies

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The purpose of this experiment is to show the process of preparing Thermo Scientific NAb Spin columns for purification of antibodies from endotoxins that it during production. This purification removes a certain percentage of endotoxins. Endotoxins can affect organ systems so they must be removed to negligible amounts. The sample had an endotoxin level of 12,000 eu/ml. The sample used was 2G8. Anti-1, 3 beta glucan antibody [2G8] has been shown to be effective in models of fungal diseases including vaginal and systemic Candida infection, invasive Aspergillosis and Cryptococcus Neoformans. The Detoxi gel resin made of Polymixin B ligand immobilized and ethanol (25-45%). To regenerate the Detoxi-Gel Resin wash with five resin-bed volumes of 1% sodium deoxycholate, followed by 3-5 resin-bed volumes of pyrogen-free buffer to remove the detergent. Regenerate the resin before each use. Equilibrate the Detoxi-Gel Resin with 3-5 resin-bed volumes of pyrogen-free buffer. Apply sample to the column. Add pyrogen-free buffer and collect the flow-through. With a gravity-flow column, the sample will emerge from the column when 90% of the bed volume has been collected. 42.56% or 0.3405 mg was recovered from 0.8 mg sample. The Endotoxin Units per milliliter was 0.15 eu/ml. The FDA regulates the acceptable level of endotoxin contamination to be 0.5 eu/ml which means our sample is ready to administer to mice as a treatment. This method is not perfect as 58% of the sample is lost during purification. The goal in the future is to increase the amount of the sample recovered.

36 Pharmacokinetic Modeling Utilizing Feed Forward Neural Networks

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CSU Channel Islands, NSF LSAMP Undergraduate & NSF Bridge to Doctorate

The selection of optimal drug dosages is crucial to every phase of the drug development process. Central to understanding these dosages is the study of pharmacokinetics or how a drug is affected by the human body after consumption. Pharmacokinetic modeling is used to understand the relationship between a drug's dosage and its consumer's blood plasma concentration levels, but it can be challenging due to complex data and the inherent variability within and between human subjects. Pharmaceutical companies manage these challenges through the use of traditional statistical modeling to identify the covariates that could influence the dose-concentration relationship. Although this approach suits small sample sizes and sparse data sets well, it requires researchers to specify a structural pharmacokinetic model a priori, which can be cumbersome to adjust when dealing with highly variable and massive data sets. In this research, we focused on utilizing neural networks as an alternative method to discover the dose-concentration relationship from a sample of 612 patients. In particular, we sought to predict the area under the concentration curve and maximum concentration given patient biometric data and initial drug dosage as the network's input features. These desired outputs are vital in determining the extent of exposure and the highest concentration the drug will achieve in the patient's bloodstream. After training several single-layer neural networks, we were able to evaluate their performance to determine the best neural network architecture and determine whether neural networks can replace parametric models as the standard in pharmacokinetic modeling.

37 Polymer-bound Metal Complexes for Enzyme-like Catalysis

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Our group aims to synthesize an enzyme-like catalyst with a system that mimics the active site of biological enzymes, including the metal and both the primary and secondary coordination spheres, the latter through the use of polymer-based scaffolding. The primary coordination sphere includes the direct bonds between the enzyme's polypeptide backbone and the catalytic metal center. The secondary coordination sphere provides additional interactions between the polypeptide backbone and things bonded to the catalytic metal. The secondary coordination sphere involves steric and noncovalent interactions both of which can improve reactivity, selectivity, and specificity. Most synthetic enzyme mimics only mimic the primary coordination sphere and while these mimics can teach us about the structural behavior of the enzymes, few achieve enzyme-like catalysis. We expect that the addition of the polymer-based scaffold as a mimic of the secondary coordination sphere will influence the reactivity at the metal center. In this project, we are targeting a mimic of dopamine β-monooxygenase. We have successfully synthesized literature-based copper complexes and modified these complexes to include vinyl groups that allow for direct copolymerization with alkene monomers such as acrylates and acrylamides. We are currently studying the copolymerization of our vinylated ligand precursors with commercial monomers using reversible addition-fragmentation chain transfer (RAFT) polymerization and subsequent metalation of the resultant polymers to form our desired metallopolymers. We will initially study the catalytic behavior of these metallopolymers via a variety of oxidation reactions and compare the reactivity and selectivity with non-polymer-bound analogues that we have synthesized to use as benchmarks.

38 Targeting Enhanced Fluorescence with Azetidine Groups in Nucleobase Analogues

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Fluorescent nucleobase analogues are used to study the structure and dynamics of DNA/RNA folded motifs, lesions, modifications, and binding complexes with proteins. Brightness, photostability, and relatively long absorption and emission wavelengths greater than 400 nm are key enablers of these studies, especially with current interest in single-molecule fluorescence. Prior studies in our lab have led to a new tricyclic pyrimidine nucleoside analogue that exhibits brightness greater than any other FBA when incorporated in oligonucleotides, either single-stranded and duplex. This C-linked 8-(diethylamino)benzo[b][1,8]naphthyridin-2(1H)-one nucleoside, which we named ABN, is also among the most red-shifted fluorescent nucleobase analogue in duplex DNA, Φem, 540 = 0.50-0.53 when base paired with adenine. As seen in Janelia Fluor dyes, replacing a N,N-dimethylamino in a fluorophore with azetidine greatly improves quantum yield and photostability. This slight modification increased fluorescent lifetime while preserving extinction and emission profiles. I hypothesize that substituting an azetidine ring for the diethylamino group of ABN will result in improvements in quantum yield and photostability. This new fluorescent nucleobase analogue would be beneficial in its utility for labeling, imaging, and detection of DNA/RNA modifications. Details on the design of this new FBA and progress towards its synthesis, characterization, and applications will be presented.

39 Purification of the Manganese Oxidizing Protein, MopA-hp, Found in Marine Bacterium Erythrobacter sp. SD-21.

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Manganese (Mn) in both terrestrial and marine environments can be oxidized from soluble Mn-II into insoluble Mn-III/IV by bacteria. The Mn oxides play an important role in the fate and biogeochemical cycling of vital elements such as carbon, sulfur, and iron. The Mn oxidation mechanism employed by bacterial enzymes is unclear, but with the potential to be utilized in the field of bioremediation. The marine bacterium Erythrobacter sp. SD-21 is known to produce a Mn-oxidizing protein, MopA (250-kDa) – a peroxidase cyclooxygenase. Previous purification attempts of the peroxidase domain, MopA-hp (120-kDa), have provided heterologously expressed samples with incomplete purification or no activity. The purpose of this project is to derive an active and pure sample of MopA-hp through different purification methods. The current purification protocol involves gravity nickel-affinity chromatography. Such a method has provided an active but not very pure sample of MopA-hp. Additional purification by anion exchange chromatography was investigated. The 120-kDa MopA-hp is identified by SDS-PAGE. The Mn oxidation activity is quantified through a colorimetric leucoberlin blue assay. A considerably pure sample of MopA-hp failed to oxidize Mn. A pure and active sample of MopA-hp will allow future studies on the mechanism of Mn oxidation implied by MopA-hp.

40 Production of CRISPR-Cas9 editing tools for the purpose of treating hemophilia A patients with inhibitors

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Hemophilia A is due to a deficiency in the FVIII gene. Current treatments include multiple weekly injections of recombinant FVIII (rFVIII). However, 30% of individuals with severe Hemophilia A develop inhibitors to rFVIII. Once a patient develops inhibitors, the current treatment options are no longer viable, and current gene editing protocols have excluded this population. Therefore, we are proposing a strategy in which FVIII is shielded from preexisting circulating inhibitors via incorporation into one of two platelet specific loci (the von Willebrand locus and the platelet factor 4 locus). We hypothesize that these loci can be edited to treat hemophilia A patients with inhibitors because FVIII can be sequestered inside of platelets isolated from circulating inhibitors until required for hemostasis. Towards this aim, we have selected two sets of sgRNAs using several in silico tools. Each guide was then incorporated into a Cas9 expression plasmid and confirmed by sequencing. The cutting efficiency of one set of sgRNAs was tested in K562 cells and found to range from 7% to 20% after 72 hours. In addition, we have designed and produced DNA repair templates containing 500bp arms of homology to enhance homology directed repair at the loci. These homologous regions flank a proof of principle transgene (GFP) to be expressed bicistronically via a viral T2A element. Together these two components, the DNA repair template and the sgRNA/Cas9 expression plasmid, will be nucleofected into hematopoietic cells. If successful, this strategy will provide treatment for a patient population that is currently overlooked.

41 Probes for Metal-Insulator Phase Transition in Half-Filled One-Dimensional Fermi-Hubbard Model

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Computer simulations provide valuable insight into investigating the behavior of quantum many-body systems. The objective of our computational experiment is to perform a simulation of a collection of fermions in a crystalline solid from the fundamental equations of quantum mechanics. Using Quantum Monte Carlo simulation, we computed quantum metric tensor and localization length in the ground state of the one-dimensional Fermi-Hubbard model, as probes for conductivity. We compare these values with the exact values of the charge gap of the system, for different values of interparticle interaction strength. We perform calculations for lattices up to 650 sites, at half-filling, using both periodic and open boundary conditions. We assess and discuss the sensitivity of the mentioned probes in a metal-insulator phase transition. We will also discuss and compare the preliminary results for two-dimensional lattices. This is a relevant study in a strongly correlated system as most current approaches rely only on response functions which involve the excited states of the system, thus making it a formidable task for theoretical and even computational approaches. *This material is based upon work supported by the CSU-LSAMP program funded by NSF under grant #HRD-1826490, CSU Office of the Chancellor, and Fresno State.

42 Photoperiod alters ovarian mRNA expression of genes in the retinoic acid pathway

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Cyclic ovarian function requires the orchestration of multiple signaling pathways; however, most vertebrate ovaries do not cycle continuously due to seasonal pauses in reproduction. While the endocrine regulation of seasonal ovarian change is well understood, how changes in ovarian function impact other signaling pathways remain unknown. Because the retinoic acid (RA) pathway is involved in ovarian cell proliferation, differentiation, apoptosis, and oocyte maturation, we hypothesized that the genes in the RA pathway would be differentially expressed in ovaries that are cycling, non-functional, and returning to function. To address our hypothesis, we used ovaries from seasonally-breeding Siberian hamsters who were exposed to 16-weeks of long-days (16h light:8h dark; LD; cycling ovaries), or short-days (8L:16D; SD; regressed ovaries), or 16-weeks of SD followed by 2, 4, or 8-weeks post-transfer to LD (PTw2-8; recrudescing ovaries). Real time PCR expression showed that retinoic acid receptor-γ and retinoid X receptor-β, both of which bind the RA ligand, were present in LD ovaries and decreased significantly with SD exposure. Expression of these receptors was restored to LD levels in the PTw2, 4, and 8 groups. In contrast, mRNA expression of RA-degrading enzyme Cyp26b1 increased significantly in SD as compared to the LD group, with expression returning to lower LD levels in the recrudescing groups. Our results suggest that the RA signaling pathway is active in cycling Siberian hamster ovaries, with decreases in RA binding concomitant with increases in RA degradation in regressed ovaries, and restoration of RA binding occurring as photo-stimulated ovaries return to function.

43 Discovery of Novel Natural Products with Selective Cytotoxicity Towards a Pancreatic Cancer Cell Line

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Natural products are secondary metabolites produced by living organisms. They have played an important role in traditional medicine for thousands of years and continue to be an essential part of the current healthcare system, as over 65% of all approved therapeutic drugs are either natural product, natural product derivatives, or their pharmacophores are inspired from natural products. The success of these compounds and their derivatives as therapeutic agents is largely due to their high structural diversity and specific biological targets. Pancreatic cancer is a solid tumor cancer and over the last 50 years there has been no significant progress made to increase the 5-year survival rates of patients diagnosed with pancreatic cancer. The overall goal of this research is to identify novel chemical scaffolds from a large library of marine derived fungi that exhibit selective activity towards a pancreatic (PANC-1) cancer cell line. This was achieved by building a fungal library. The fungal library was grown by culturing 50 unique fungal strains in five different types of media and extracting the biosynthesized natural products. The metabolites were extracted and used to generate Distinct Testing Units (DTU). The DTU plates were screened in a biological assay to identify the extracts that contain compounds that exhibit selective cytotoxicity towards a pancreatic cell line. Extracts that exhibited cytotoxicity towards the PANC-1 cell line and that have a novel chemical scaffold were given priority.

44 Identifying Novel Chemical Scaffolds from Marine Derived Fungi using Mass Spectrometry based Molecular Networking

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Natural products are secondary metabolites produced by living organisms. They have played an important role in traditional medicine for thousands of years and continue to be an essential part of the current healthcare system, as over 60% of all approved therapeutic drugs are either natural products, natural product derivatives, or their pharmacophores are natural product inspired. The overall objective of this research was to identify novel natural products from marine derived fungi. This was achieved by culturing 50 taxonomically unique fungal strains and extracting the biosynthesized natural products they produced. The extracts were analyzed using liquid chromatography-mass spectrometry (LC-MS) in a tandem (MS/MS) format. The MS/MS data was analyzed using Global Natural Products Social Molecular Networking, a program that allows for rapid identification of known chemical scaffolds. This enabled the identification of clusters of compounds from different fungal extracts that had putatively novel chemical scaffolds. The natural products present in these extracts were purified using high performance liquid chromatography, and structurally elucidated using high accuracy MS as well as 1D and 2D NMR experiments.

45 Identification of Novel Pyrrolo[4,3,2-de]quinoline Natural Products from the Marine Sponge, Zyzza fuliginosa

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Marine sponges are a reservoir of immense chemical diversity. The marine sponge Zyzza fuliginosa is a rich source of families of compounds that contain fused A/B/C tricyclic pyrrolo[4,3,2-de]quinoline moieties. This includes the makaluvamines and damirones, compounds that have been shown to have potent cytotoxicity towards a variety of solid tumor cell lines. The objective of this research was to identify novel pyrrolo[4,3,2-de]quinoline compounds from extracts of Z. fuliginosa. To achieve this multiple extracts of Z. fuliginosa along with standards of nine makaluvamines and three damirones, were analyzed using LC-MS in a tandem (MS 2) format. The MS 2 data was analyzed using the Global Natural Products Social Molecular Networking platform. Metabolites whose MS 2 fragmentation patterns showed high similarity scores to the makaluvamines and damirones, but that had unique molecular formulas based on high accuracy MS were selected for further investigation. These putatively novel pyrrolo[4,3,2-de]quinoline compounds were purified from the crude Z. fuliginosa extracts using HPLC and structurally elucidated using 1D and 2D NMR experiments. This led to the identification of a series of novel pyrrolo[4,3,2-de]quinoline compounds.