SURF PROJECTS 2019

FACULTY MENTOR:
Salah-uddin Ahmed, Ph.D., Associate Professor, *ASPET member*

RESEARCH PROJECT:
Identifying novel therapeutic targets in the pathogenesis of rheumatic diseases

DESCRIPTION OF PROJECT:
The Ahmed lab is involved in translational studies aimed at identifying the unknown mechanisms that govern pathogenesis of rheumatic diseases such as rheumatoid arthritis, osteoarthritis, and gout. In particular, our group has been investigating the posttranslational and epigenetic mechanisms that facilitate rheumatic disease propagation. Using synovial cells, tissues, and serum samples from rheumatoid arthritis patient population, we conduct genomics and metabolomics studies to identify some key mediators that promote chronic inflammation and tissue destruction in rheumatoid arthritis. Based on this information, our group in collaboration at international level with computational biologists and medicinal chemists has been designing novel compounds to be tested for their efficacy in vitro and in pre-clinical models of rheumatoid arthritis and other autoimmune diseases.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
The SURF student is assigned a role and/or participation in some ongoing or new projects where they not only get the chance to learn about the inflammatory signal transduction pathways that promote rheumatic diseases, but also get a chance to sharpen their technical skills on running independently procedures such as ELISA, Western blotting, and In-gel zymography. After the training, the SURF fellow will be capable of summarizing his/her progress in the form of poster presentation. The work will be further expanded and presented at one of the national scientific meetings in which SURF students typically serve as co-authors. These studies have also resulted in the publication of manuscripts in the past.

FACULTY MENTOR:
Zhaokang Cheng, Ph.D., Assistant Professor

RESEARCH PROJECT:
Novel role of cell cycle regulators in the heart

DESCRIPTION OF PROJECT:
The widely-used anthracycline family anti-cancer drug doxorubicin can cause serious cardiotoxic side effects by inducing apoptotic death of cardiac myocytes. Dr. Cheng has previously identified the cyclin-dependent kinase inhibitor p21 as a critical protective molecule against cardiomyocyte apoptosis. Most recent findings from Dr. Cheng's laboratory revealed that p21 inhibited apoptosis by repressing cyclin-dependent kinase 2 (CDK2). These results support a novel function of the cell cycle regulators p21/CDK2 in promoting cell survival and conferring chemoresistance in the heart. The proposed project will determine the role of CDK2 downstream

signaling pathway in doxorubicin-induced apoptosis, and further explore the therapeutic potential of CDK inhibition against doxorubicin-induced cardiotoxicity.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
Upon joining the lab in early summer, the SURF fellow will work with his/her direct mentor in Dr. Cheng's group on the proposed project. The SURF fellow will receive expert training in a plethora of research skills including cell culture, Western blotting and immunofluorescence staining. Both the direct mentor and Dr. Cheng will be available to help the SURF fellow with research-related questions, difficulties, and future directions.

FACULTY MENTOR:
John D. Clarke, Ph.D., Assistant Professor

RESEARCH PROJECT:
Multifactorial approach to precision medicine

DESCRIPTION OF PROJECT:
Toxicities from xenobiotic exposures are a significant burden on human health. These exposures occur both intentionally (drugs and dietary supplements) and unintentionally (environmental contamination and naturally occurring toxins), and there are several factors in individual variability that determine who is at greater risk for these toxicities. This project is focused on precision medicine and investigates how perturbations in two or more factors in drug absorption, distribution, metabolism, and excretion affect drug exposures and toxicities. These factors include genetics, diseases, and co-exposures, which are investigated using cell culture and rodent models. Particular attention is given to drugs that are dependent on drug transporters and/or metabolism enzymes for elimination.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
After initial training, the fellow will be expected to work in an independent manner on all standard laboratory techniques (cell culture, Western blotting, etc.). The fellow will have the opportunity to work with lab staff on non-routine procedures (animal experiments, small molecule analysis, etc.) with the expectation that the fellow will be able to work in a semi-independent manner towards the goals of the project. The fellow will participate in the weekly lab meetings and meet with Dr. Clarke individually as needed, but no less than once every two weeks.

FACULTY MENTOR:
Travis Denton, Ph.D., Assistant Professor

DESCRIPTION OF PROJECT:
Projects in the lab focus on small molecule, organic synthesis-based, medicinal chemistry. We also focus on the analytical determination of small molecules in biological matrices (analytical biochemistry). Projects currently ongoing in the lab are multiple, and all focus on trying to make life easier for people who are tormented by devastating neurological diseases.

Project 1: Treatments for ALS, Alzheimer's disease, and other neurological disorders. In this project, we are developing small molecules to enhance cellular autophagy in an effort to fight neurodegenerative diseases. Project 2: Treatments for myc-induced T-cell leukemia (cancer). In this project, we are developing phosphonic acid bioisosteres of fundamental α-ketoacids to inhibit newly identified pathways important for the progression of a number of cancers. Project 3: Treatment of Alzheimer's disease by targeting the α7 nicotinic acetylcholine receptor (nAChR). In this project, we are synthesizing small molecules to target the α7-nAChR, the only nicotinic receptor in the brain of Alzheimer's patients that is not down regulated. Through this approach, we hope to enhance the cognitive function of people with this disease. Project 4: Development of a synthetic strategy and the preparation of biomarkers for human inborn errors of metabolism.

Degree of independence expected of summer fellow:
For Projects 1-3, the SURF student will synthesize, purify, and characterize new molecules that will be tested for biological activity by our collaborators. For Project 4, the undergraduate researcher will prepare the compounds using organic chemistry techniques as well as aiding in the development of extraction and ultra-performance liquid chromatography tandem mass spectrometry techniques for the quantification of these biomarkers. The SURF fellow will receive training from members of the laboratory so that they can perform procedures independently by the end of the summer. Results will be discussed with the mentor and presented in a poster at the end of the training period.

FACULTY MENTOR:
Philip Lazarus, Ph.D., Professor and Chair of Pharmaceutical Sciences, *ASPET member*

RESEARCH PROJECT:
The overlapping metabolism of opioids and cannabis: Potential consequences for toxicity and withdrawal among opioids users.

DESCRIPTION OF PROJECT:
As the U.S. population struggles with the ever-increasing burden of the ballooning opioid crisis, the interaction of cannabis and opioids is becoming an emerging area of research. Cannabis and opioids are metabolized by many of the same phase I and phase II enzymes in the liver, which could potentially influence the effects of either or both of these compounds. Compounds in both classes undergo rapid hydroxylation or oxidation by hepatic cytochrome P450 (CYP450) enzymes, followed by glucuronidation via the uridine 5'- diphospho-glucuronosyltransferase (UGT) class of enzymes, and finally excretion. The inhibition of these shared enzymes by cannabis and its metabolites has the potential to greatly influence opioid plasma levels in patients currently taking opioids as part of a pain management strategy. Preliminary research in our lab indicates that the cannabinoid (-)-trans-Δ9- tetrahydrocannabinol (THC) and three of its major metabolites (11-OH-THC, THC-COOH and THC-COO-glucuronide) are indeed inhibiting a number of CYP and UGT enzymes. Of particular interest is the inhibition of CYP2D6 and UGT2B7, which act to metabolize codeine, morphine and

hydrocodone. **The goal of the present proposal will be to fully characterize this inhibition by a panel of major liver metabolizing enzymes using THC, cannabidiol (CBD) and their metabolites.** Given the public perceptions and misconceptions surrounding the efficacy of cannabis, and its potential uses in pain management, opioid withdrawal, and as a part of many patient's general health practices, a better understanding of how these bioactive compounds are influencing metabolic systems within the human liver are necessary. This knowledge will aid in understanding how cannabis fits into the public health crisis surrounding opioid overdose and opioid use disorder, and provide a baseline understanding of how cannabis is metabolized, and how those active metabolites are influencing liver function in patients currently taking opioids

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
Students will be trained in the enzyme activity assays and analytical techniques needed to accomplish the experiments described above. The student will work directly with a graduate student trainee and have bi-weekly meetings with the mentor, as well as attending bi-weekly laboratory meetings. Once trained, the student will be able to work fairly independently, and will report on progress at meetings independently.

FACULTY MENTOR:
Kathryn Meier, Ph.D., Professor, Pharmaceutical Sciences, *ASPET Member*

RESEARCH PROJECT:
FSH as a potential therapeutic target in prostrate cancer

DESCRIPTION OF PROJECT:
The Meier lab is committed to identifying new signal transduction targets for cancer therapy. In particular, our group is investigating the role of follicle-stimulating hormone (FSH) in prostate cancer. FSH is a peptide hormone that is important in reproduction, but is also produced by other cell types including human prostate cancer cells. FSH stimulates growth of prostate cancer cells that express the FSH receptor. Using established prostate cancer cell lines, we will investigate which cellular signaling pathways are responsive to FSH, and which forms of FSH stimulate cell growth. The ability of other G protein-coupled receptors to exert positive or negative effects on FSH response will also be tested. The results of this study are anticipated to provide insight into how the FSH receptor can be targeted for the treatment of prostate cancer.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
The SURF fellow will be assigned a "mini-project" for the summer, and will be trained in the techniques needed to perform the work These techniques will include cell culture, cell proliferation assays, and immunoblotting. Results will be analyzed using statistics software. The SURF fellow will be able to perform these experiments semi-independently by the end of the summer. Weekly lab meetings will be held in which the fellow and other lab members will report on their results and receive feedback and suggestions. At the end of the summer, the fellow will present his/her work in a poster presentation. Projects conducted by previous SURF students have resulted in co-authorships on published abstracts and/or manuscripts.

FACULTY MENTOR:
Senthil Natesan, Ph.D., Assistant Professor

RESEARCH PROJECT:

Membrane-facilitated allosteric modulation of GPCRs

DESCRIPTION OF PROJECT:

Recent advances in structural biology of GPCRs, along with biophysical and computational studies suggest that amphiphilic and lipophilic molecules may gain access to binding sites of these receptors by first partitioning into the membrane and then reach the sites via lateral diffusion through the lipid bilayer. The highly ordered structure of the lipid bilayer may guide a lipophilic or amphiphilic drug molecule to a specific depth within the bilayer, affecting its local concentration near the binding site, and influencing its binding kinetics. Additionally, the constraints of the lipid bilayer including its composition and biophysical properties might play a critical role in "pre-organizing" ligand molecules in appropriate orientation and conformation to facilitate receptor binding. Despite its conspicuous involvement in molecular recognition processes, the critical role of membrane in ligand-binding mechanism to lipid-exposed transmembrane binding sites remains poorly understood. This project aims to gain mechanistic understanding of the structure-membrane interactions relationship (SMIR) of several approved drugs. Knowledge of SMIR will not only provide useful insights to receptor binding kinetics, but also enhance our ability to take advantage of the apparent membrane contributions in rational design of drugs targeting transmembrane proteins with improved efficacy and safety.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:

The SURF fellow will learn basic to intermediate level computational techniques including building 3D structures of small molecules, geometry optimization, charge calculations, preparation of protein-ligand complex structures, performing MD simulations using NAMD, calculating binding free energy etc. As these techniques require extensive initial training, the fellow will work very closely with Dr. Natesan and his postdocs in the beginning and will become semi-independent towards the completion of the project. The student will have plenty of opportunity to interact with the mentor and his group members to discuss the progress and future directions.

FACULTY MENTOR:
Mary Paine, R.Ph., Ph.D., Associate Professor, *ASPET member*

RESEARCH PROJECT:
Natural product-drug interactions: elucidating mechanisms and causative ingredients using translational research methods

DESCRIPTION OF PROJECT:
Patients often supplement their prescribed drug regimens with herbal and other natural products (NPs), which are readily available and typically perceived as safe. In parallel, patients may not inform their health care providers, and health care providers may not query their patients, about NP usage. These practices raise concern for potentially harmful interactions between NPs and conventional drugs. Mechanisms underlying these understudied interactions can be pharmacokinetic (what the body does to the drug or NP), pharmacodynamic (what the drug or NP does to the body), or both. Common pharmacokinetic mechanisms include inhibition of drug metabolizing enzymes by an NP, leading to enhanced systemic concentrations of the object drug and potentially, harmful effects. Projects involve (1) in vitro experiments using human-derived tissue to recover key enzyme kinetic or binding parameters of constituents in a given NP, (2) applying these parameters to mathematical models to make in vivo predictions, and/or (3) determining the pharmacokinetics of the NP constituents and object drug using plasma and urine obtained from a clinical interaction study.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
Dr. Paine will work with the student to develop a project that includes one or more of the three aforementioned facets of our translational approach based on the student's interests and skillset, and an experienced member of the research team will work with the student directly. The student will meet with Dr. Paine regularly to monitor progress. In addition to participating in various SURF activities, the student will interact daily with group members and participate in weekly group meetings. The student is expected to become semi- or fully independent.

FACULTY MENTOR:
Boyang (Jason) Wu, Ph.D., Assistant Professor

RESEARCH PROJECT:
Molecular Mechanisms of Enzalutamide Resistance in Prostate Cancer

DESCRIPTION OF PROJECT:
This laboratory currently centers on prostate cancer research, with particular interests in exploring the molecular mechanisms underlying the development of drug resistance to next-generation antiandrogens, including enzalutamide and abiraterone acetate, in advanced prostate cancers. We recently identified aberrant upregulation of Sema3E/PlexinD1/NRP2 in established enzalutamide-resistant prostate cancer cell lines using RNA-seq approach. These molecules have been originally implicated in the control of axonal guidance and angiogenesis and recently shown to play roles in certain types of cancers. Using enzalutamide-resistant cancer cell lines and derived xenograft mouse models, we will define the functional and mechanistic roles of

Sema3E/PlexinD1/NRP2 in the development of drug resistance as well as in regulating cancer cell crosstalk with other types of cells in the microenvironment, such as nerve cells and endothelial cells. We will also determine whether targeting these molecules could reverse antiandrogen drug resistance and synergize with other therapies for improved treatment efficacy.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
At the beginning of the summer, the fellow will work closely with Dr. Wu's postdocs to learn basic cell and molecular biology techniques necessary to execute the project in the in vitro setting, including cell culture, DNA and RNA extraction, Western blot, RT-qPCR, ELISA, MTS assays, etc. Meanwhile, the fellow will be exposed to reading scientific papers assigned by mentor, which are not only related to the project but also in a broad way to allow the fellow to learn the frontier research in prostate cancer filed specifically and cancer field in general. After obtaining required training for animal studies, the fellow is also expected to participate in observing animal experiments and assisting other lab members with animal experimental procedures, such as measurement of tumor growth and scoring body conditions of mice. After the training, the fellow will work semi-independently on his/her project. The fellow will report research progress at every biweekly lab meeting and give a research summary presentation to the lab at the end of summer.

FACULTY MENTOR:
Hui Zhang, PhD, Assistant Professor

RESEARCH PROJECT:
Suppression of anti-tumor immunity by chronic alcohol intake

DESCRIPTION OF PROJECT:
This laboratory is focused on how chronic alcohol consumption compromises anti-tumor immunity and on selecting appropriate targets counter the negative effects. They have shown that chronic alcohol consumption compromises CD8+ T cell function. CD8+ T cells are the key players in anti-tumor immunity and in controlling the survival of the tumor-bearing host. The function of CD8+ T cells are regulated by several immune regulatory cells including myeloid-derived suppressor cells (MDSC), tumor associated macrophage (TAM), regulatory T cells (Treg), NKT cells, and B cells. They also showed that chronic alcohol consumption increases MDSC and NKT cells, decreases B cells, and shifts the NKT cell cytokine profile from Th1 (anti-tumor) to Th2 (tumor-evasive). Chronic alcohol consumption does not alter TAM or Treg in the tumor-bearing mice. Two potential projects are examining the signaling pathways that are modulated by chronic alcohol consumption and tumor cells to 1) regulate B cell circulation and 2) alter the cytokine profile of NKT cells.

DEGREE OF INDEPENDENCE EXPECTED OF SUMMER FELLOW:
At the beginning the summer, the fellow will work closely with Dr. Zhang to learn some basic and specific procedures such as animal handling and molecular and cellular techniques. After the training, the fellow will work semi-independently on his/her project. There will be a lab meeting weekly to discuss progress and future directions.
