Commonwealth Health Research Board
FY 2019/2020 Grant Award Abstracts

Matthew Buczynski, Ph.D., Virginia Polytechnic Institute and State University
Evaluation of a 12/15-LM receptor as a target for Non-Opioid Pain Therapeutics

Project Summary: The opioid crisis has reached epidemic proportions in the United States, and in 2016 Governor McAuliffe declared opioid addiction in the Commonwealth as a Public Health Emergency [1]. Rural western VA (where Virginia Tech is located) reports some of the highest per capita opioid abuse in the country. Thus, non-opioid therapeutic alternatives to NSAIDs (Nonsteroidal Anti-inflammatory Drugs, e.g. ibuprofen) for the effective management of chronic pain are essential to limiting opioid overuse. Our published studies identified a novel class of signals (12/15-lipoxygenase metabolites, 12/15-LMs) that contribute directly to (NSAID)-insensitive nociceptive behaviors in multiple pre-clinical pain models, and our preliminary results identified a novel receptor for 12/15-LMs. In this proposal, we plan to characterize the 12/15-LM receptor, and screen potential lead compounds that block receptor activity. Ultimately, our goal is to enable drug discovery efforts for novel analgesics with minimal abuse potential and mitigate risks of opiate misuse, diversion and addiction.

Charles Clevenger, M.D., Ph.D., Virginia Commonwealth University
HDAC6 as a Therapeutic Target in Breast Cancer

Project Summary: We recently identified a growth regulatory role in breast cancer for the histone deacetylase, HDAC6. HDAC6 functions as a "signaling switch" for cancer by removing acetyl groups from proteins. We discovered that HDAC6, acting through the complex of HGMN2/H1.2/Stat5, regulates the growth and progression of breast cancer. Through these actions, we hypothesize that HDAC6 is controlling breast cancer gene expression and growth at a global level, functioning as an "oncogenic node". These findings are significant in that small molecular inhibitors of HDAC6, have proven safety in Phase I trials (in myeloma patients), and therefore can be rapidly translated to breast cancer patients. To test our hypotheses, we propose two specific aims to assess the nuclear function of HDAC6 in breast cancer:
Aim #1. **To assess HDAC6 regulation of global gene expression in breast cancer.**
Aim #2. **To translate the therapeutic potential of HDAC6 inhibition into mouse models of mammary cancer.**

Paul Dent, Ph.D., Virginia Commonwealth University
Novel anti-sarcoma therapies

Project Summary: Votrient (pazopanib) is approved for the treatment of sarcoma. We discovered that the seizure medication Depakote (sodium valproate) synergized with pazopanib to kill sarcoma cells on a plastic dish and when the cells were growing as a tumor. Cells that had been exposed to [pazopanib + valproate] had evolved and activated an enzyme called c-MET, which on a plastic dish made the cells chemotherapy resistant. **Aim 1. Determine whether inhibition of c-MET promotes [pazopanib + valproate] lethality against sarcoma tumors.** Treatment of sarcoma cells with valproate and more so [pazopanib + valproate] rapidly reduced the levels of proteins called PD-L1, IDO-1 and ODC that block anti-tumor immune responses, and increased levels of the class I MHC protein MHCA, which enhances anti-tumor immune responses. **Aim 2. Determine using mouse sarcoma tumors the impact of [pazopanib + valproate] has on the anti-tumor response of anti-PD-1 and of anti-CTLA4 checkpoint inhibitor immunotherapies**.

Commonwealth Health Research Board
FY 2019/2020 Grant Award Abstracts

Brent French, Ph.D., University of Virginia
Bioengineering of Cardiac Regeneration In Situ after Myocardial Infarction

Project Summary: Heart failure currently afflicts some 5.7 million Americans, and by 2030 this number will increase by 46%. The single most common cause of heart failure is heart attack (or myocardial infarction) which results in the irreversible loss of cardiac muscle. Current statistics show that ~790,000 people in the US have heart attacks each year. Of those, about 114,000 will ultimately die from heart failure. The overarching goal of this project is to combine recent advances in cardiology, radiology and gene therapy to demonstrate that cardiomyocytes can be genetically-reprogrammed to divide and replace the heart muscle lost during heart attack. This is important because the adult heart has essentially no capacity to repair itself after a heart attack. Instead, injured cardiomyocytes are replaced by scar tissue to prevent the heart from rupturing. If successful, this research will show that gene therapy can regenerate muscle tissue after heart attack instead of scar.

Li Jin, Ph.D., University of Virginia
Could we treat acute back/leg pain with nanoparticle fullerene instead of steroid?

Project Summary: Low back/leg pain is the single leading cause of disability worldwide. The use of fluoroscopy-guided lumbar epidural injections of steroids to reduce local inflammatory response induced by disc herniation has increased dramatically. However, side effects of epidural steroid injection have been reported widely. Nanoparticle C_{80} has potent anti-inflammatory and anti-oxidative stress effects. We hypothesized that local administration of C_{80} relieves the back/leg pain and retard disc degeneration. Due to the long-lasting activity of C_{80} and its excellent cell membrane-penetrating abilities, we anticipate that nanoparticles C_{80} replace steroids for acute back/leg pain. The proposed three-aim study will delineate the beneficial effects of C_{80} on back/leg pain both *in vitro* and *in vivo*. The preliminary work strongly suggests that predicted outcomes are highly feasible. The success of this proposal is ensured by a strong multidisciplinary collaboration among Drs. Jin (biologist, UVA), Dorn (nanofullerene expert, VT) and Li (a spine surgeon, UVA).

Peter Kasson, M.D., Ph.D., University of Virginia
Rapid identification of entry inhibitors and neutralizing antibodies for emerging viruses

Project Summary: Zika virus is an emerging health threat in the Americas, causing a febrile illness with severe neurological after-effects and birth defects. Virginia has mosquitos capable of transmitting Zika and similar viruses and had over 100 cases of Zika in 2016. Early evidence suggests that some antibodies to Zika can be protective while others may actually increase the severity of infection. This proposal develops a means to discover antibodies and drugs that inhibit Zika entry in a fashion that is readily adaptable to other emerging viruses. We will accomplish this via an assay using fluorescence microscopy in microfluidic flow cells to measure viral entry. This capability will help bolster the public health defense in Virginia and elsewhere against emerging viruses by allowing rapid testing of potential antibodies and antiviral drugs.

Commonwealth Health Research Board
FY 2019/2020 Grant Award Abstracts

James Landers, Ph.D., University of Virginia
Diagnostic Assay for On-Site Detection of Bordatella pertussis

Project Summary: *Bordetella pertussis*, the causative agent of whooping cough, infects millions of individuals worldwide each year and continues to be the world's leading cause of vaccine preventable deaths. In recent decades, there has been an alarming resurgence of reported pertussis cases. A major step to address this problem is for *B. pertussis* to be detected rapidly during a suspected outbreak, enabling initiation of treatment, limitation of transmission and reduction in mortality. However, current methods require patient samples to be sent to centralized laboratories for analysis and results are typically not available in time to support epidemiologic intervention. Instead, physicians and healthcare officials use presumptive antibiotic treatment until diagnostic results are available, thereby putting many individuals at risk unnecessarily. To address this challenge, we are developing a portable microfluidic device, "lab-on-a-CD", to screen for the presence of *B. pertussis* DNA and allow for robust identification of infection in 20 min or less.

Alberto Musto, M.D., Ph.D., Eastern Virginia Medical School
Role of CD40L in Limbic Epileptogenesis

Project Summary: Temporal lobe epilepsy (TLE) or limbic epilepsy, the most common forms of epilepsy in adults, has no cure or effective treatment in a subset of patients. CD40 ligand (CD40L), small protein involved in immune and inflammatory process is upregulated after seizures. The overall objective is to define if CD40L interaction with its receptor CD40 participates in an aberrant neuronal circuitry organization during development of epilepsy or epileptogenesis. Therefore, blocking CD40L-CD40 interaction should limit seizures and neuronal damage, improving survival rate and prevent TLE.

Nagaraja Nagre, Ph.D., Eastern Virginia Medical School
Exploring the potential role of cannabinoid receptor type-2 activation in protection against bacterial pneumonia-induced lung injury

Project Summary: Bacterial pneumonia is a major risk factor for developing acute lung injury (ALI). Although mechanical ventilation remains the last resort of treatment, it carries risks of lung cell injury, high mortality, and morbidities. *Pseudomonas aeruginosa* is an opportunistic pathogen causing a wide range of acute and chronic infections and is a major cause for Ventilator-Associated Pneumonia (VAP). The ineffectiveness of conventional antibiotics therapy among severe pneumonia-induced lung injury patients appeals for novel options of treatment. One such candidate is Cannabinoid receptor-2 (CB2R) that is predominantly expressed in immune cells. Synthetic agonists like endocannabinoids (that do not generate undesired psychotic effects) can be used to activate these CB2Rs leading to the display of anti-inflammatory functions. Considering the unique stance of CB2R as a potential novel therapy for bacterial pneumonia, the hypothesis that CB2R activation can ameliorates bacterial pneumonia induced lung inflammatory/injury (using a well-validated mouse model) will be tested in this project.

Commonwealth Health Research Board
FY 2019/2020 Grant Award Abstracts

Swati Palit Deb, Ph.D., Virginia Commonwealth University
Targeting mutant p53-dependent checkpoints of genome duplication in lung cancer

Project Summary: The American Cancer Society estimated number of new lung cancer cases this year alone is 234,030 in the US, and 5,860 in Virginia, of which at least 60% of patients will not survive, suggesting extremely poor efficacy of current lung cancer treatment. Gain-of-function (GOF) mutations of tumor suppressor p53 are very frequent (up to 70%) in lung cancer and establish resistance to chemo- or radiotherapy and are essential for oncogenesis. Accordingly, the tumorigenic ability of human lung cancer cells lines is drastically reduced or eliminated when endogenous mutant p53 is disabled. **In a recently published study (highlighted by the Journal of Clinical Investigation),** we demonstrated that GOFp53 activates checkpoint signaling to establish its oncogenic activities**.** Here we propose to determine the mechanism by which GOFp53 activates checkpoint signaling to establish dependency in lung cancer cells and evaluate the therapeutic efficacy of GOFp53-induced checkpoint signaling inhibitors, which has not been explored.

Bhaumik Patel, M.D., McGuire Research Institute
Development of a Selective Non-Saccharide Glycosaminoglycan Mimetic for Colon Cancer

Project Summary: Complete cure of cancer is never achieved for most advanced colorectal cancer, in part, due to the inability of the standard chemotherapy and other targeted drugs in eradicating the 'seeds of cancer', also called cancer stem cells (CSCs). We have demonstrated, for the first time that specific short sequence of heparin (HSO6) selectively eliminates CSCs. But, HSO6 cannot be a candidate drug as it is very difficult and expensive to purify it. However, we have succeeded in synthesizing a non-sugar mimetic of HSO6 – G2.2 which is easy to make, homogenous, and rather inexpensive. Using primary human CSCs, innovative animal models, and advanced in vitro methods to study stem cells, we will determine the efficacy and toxicity of G2.2 as well as its potent analogs against colon CSCs in conjunction with FDA approved colon cancer therapies. This, in our opinion, is a major step towards achieving complete cancer cure.

Liya Qiao, Ph.D., Virginia Commonwealth University
Role of TrB.T1 in Bowel and Urinary Bladder Comorbidity

Project Summary: The pain-sensing neurons in the sensory ganglia are surrounded by satellite glial cells (SGCs) and in the spinal cord by astrocytes, which form glial networks, and receive and pass pain signals from one type of neurons (e.g. those that sense pain of the colon) to another (e.g. those that sense pain of the urinary bladder), causing comorbidity of the bowel and the urinary bladder. TrkB.T1 is the sole high affinity receptor in SGCs and astrocytes for brain-derived neurotrophic factor (BDNF), a critical pain-related molecule in the occurrence of bowel and urinary bladder comorbidity. However, deletion of BDNF is lethal and cannot be used as an intervention target. We aim to use various novel genetic tools to provide evidence that TrkB.T1 is involved in bowel and urinary bladder comorbidity (Aim 1), and TrkB.T1 is a safe and unique therapeutic target in treatment of bowel and urinary bladder comorbidity (Aim 2).

Commonwealth Health Research Board
FY 2019/2020 Grant Award Abstracts

Jason Reed, Ph.D., Virginia Commonwealth University

A new approach for detecting IGH translocations in hematologic malignancies

Project Summary: Blood cancer represents a large group of different malignancies and represents roughly 10% of all cancers diagnosed in the United States each year. The complexity and implications for treatment, of blood cancer diagnosis create a critical need for molecular methods which can be applied in less specialized medical settings such as community hospitals. To address this need, we will employ very simple 'DNA barcoding' approach to detect chromosome rearrangements in blood cancers. This method will be as accurate, but much quicker and substantially less costly than, all existing alternatives. This technology can significantly improve outcomes for patients in underserved populations.

Martin Wu, Ph.D., University of Virginia

Are persister cells culprits of recurrent Clostridium difficile infections?

Project Summary: *Clostridium difficile* infection (CDI) causes mild to life-threatening diarrhea. It poses a major healthcare burden to the global population primarily affecting individuals treated with antibiotics. The biggest challenge facing CDI is the high rate of treatment failure or recurrence, which has increased remarkably in the past two decades. Persister cells (dormant or slow-growing bacteria) are known to survive antibiotic treatment. However, whether they are a major cause of recurrent CDI remains unclear. We hypothesize that persister cells play an important role in recurrent CDI. Specifically, we aim to 1) determine whether the presence and abundance of persister cells are significant risk factors for CDI recurrence, 2) determine the genetic basis of persistence by sequencing genomes of the persister cells. This study will be the first to quantitatively determine whether persister cells are significant risk factors for recurrent CDI and therefore has the potential to shift the paradigm in therapeutic strategies.