Fiscal Year 2014-2015 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
David Lee, Ph.D.	University of Miami	Addressing Racial/Ethnic Tobacco Health Disparities via Group Intervention	The importance of reducing tobacco-associated health disparities between cannot be understated. Racial/ethnic minorities are less likely to quit smoking, and tend to have elevated stress and depressive symptoms, which may contribute to cessation disparities. Cognitive behavioral therapy (CBT) for cessation addresses these concerns and has the potential to reduce/eliminate disparities. Our preliminary research found racial/ethnic differences in baseline perceived stress and depressive symptoms. Following CBT, these differences were no longer present. Moreover, compared to Whites, African Americans exhibited blunted hypothalamicpituitary-adrenal (HPA) axis functioning. This RCT will be the first to test the impact of CBT on smoking cessation disparities. Our specific aims are to: (1) Examine the effects of CBT on perceived stress and depressive symptoms in a racially/ethnically diverse sample; (2) test the efficacy of CBT for eliminating smoking cessation disparities; and (3) examine physiological distress as an underlying mechanism for the effects of CBT on racial/ethnic minority smokers (exploratory). We expect that CBT will eliminate racial/ethnic differences in stress and depressive symptoms, and smoking cessation compared to the general health education (GHE) control group. We also hypothesize that HPA functioning will mediate the effect of CBT on smoking cessation, particularly among racial/ethnic minorities. We will randomly assign African American/Black, Hispanic, or White smokers to CBT or GHE, and provide transdermal nicotine patches (TNP) to both intervention groups. Assessments will occur at the end-of-therapy (EOT), and 3, 6, and 12-months. Our primary abstinence outcome will be smoking cessation over the previous 7 days. We will also examine the effect of CBT on TNP adherence. This study has implications for eliminating disparities in psychosocial factors related to smoking cessation, and disparities in quitting success. Addressing stress and depressive symptoms through CBT may facilitate cessation, particularly among racial/ethnic minorities.

Fiscal Year 2014-2015 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Michael Campos, M.D.	University of Miami	Adverse Airway Effects of Inhaled Nicotine from Tobacco and E-cigarettes	In healthy persons, the lungs and airways are cleared from dust, viruses and bacteria to prevent disease development. Cigarette smoke impairs these host defense systems, allowing mucus build up, which is revealed by cough productive of phlegm and associated with frequent infections. This leads to diseases called chronic bronchitis and COPD. From a public health perspective, smoking cessation is therefore an important goal. To try to decrease nicotine craving during smoking cessation, tobacco-free nicotine delivery devices such as electronic cigarettes (ECs) are used. However, the safety of inhaled nicotine via ECs is unknown. In the present study, we will first use human cells that represent the airway surface in a dish and expose them to smoke to study the mechanism by which smoke components, especially nicotine, cause changes leading to increased sputum production. Our preliminary results show that the inflammatory molecule TGF-β1 is responsible for many of these changes and that inhibition of this molecule's signaling can prevent mucus build up. We will test whether nicotine directly or delivery via ECs causes changes similar to tobacco smoke in vitro. Next, we will examine whether changes observed in vitro also occur in vivo in human beings. We will test whether subjects who quit smoking with ECs show toxic effects from nicotine delivered to their airways or whether such a strategy is safe. Therefore, this translational research project will examine treatments to reverse smoking effects on the airway epithelium and will comprehensibly examine whether the delivery of nicotine via ECs has detrimental effects as well. The outcome of this project will not only be important for subjects with smoke-induced lung diseases, but will also provide a decision making basis for subjects and policy makers how to use and regulate nicotine delivery devices such as ECs.

Fiscal Year 2014-2015 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Vani Nath Simmons, Ph.D.	H. Lee Moffitt Cancer Research Center	Expanding the Reach of a Validated Smoking-Cessation Intervention: A Spanish-language Clinical Trial	Tobacco smoking is the leading preventable cause of cancer mortality. Pharmacotherapy and behavioral counseling have demonstrated independent and additive effects on smoking cessation rates; however, counseling is rarely chosen by smokers. Minimal self-help interventions, such as smoking cessation booklets, have very high potential reach, yet have shown low efficacy, with the exception of the extended self-help smoking interventions developed by our research team. Originally developed to prevent post-cessation relapse to smoking, these booklets titled, Forever Free, significantly reduced smoking relapse through two years of follow-up among individuals who had recently quit smoking and were extremely costeffective. Based on its efficacy and cost-effectiveness, we expanded the intervention to assist current smokers with initial smoking cessation as well as relapse prevention. Our recently completed National Cancer Institute funded trial of this intervention titled, Stop Smoking for Good, revealed high efficacy through the 24-month follow-up, further supporting the utility of extended self-help for promoting and maintaining tobacco abstinence. Availability of a validated Spanish-language version would enhance its public health impact by reaching the largest and fastest growing ethnic minority population of smokers. Although the current smoking prevalence among Hispanics (12.5%) is lower than non-Hispanic whites (18.1%), higher prevalence is observed among certain subgroups (e.g., Puerto Rican males, 35%). In Florida, the smoking prevalence among Hispanics (15.1%) is greater than the national prevalence, and it is higher among subgroups and within medically underserved communities. Prior work has demonstrated that Hispanic smokers face unique challenges such as lower awareness and acceptance of pharmacotherapies and less cessation assistance from health providers. This study goal is to expand the reach of our evidence-based, self-help intervention by developing and testing a Spanish-language version. This would represent an easily disseminable, low-cost intervention with significant public health impact for Hispanic smokers in Florida and elsewhere.

Fiscal Year 2014-2015 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Frederic J. Kaye, M.D.	University of Florida	First-of-its-Kind Intralesional Delivery of Oncolytic therapy for Limited Stage Small Cell Lung Cancer	Small cell lung cancer (SCLC) is a unique and highly aggressive subtype of lung cancer that rapidly spreads to distant organs and for which there have been no improvements in standard treatments for the past 3 decades. SCLC is also the subtype of lung cancer that is most tightly linked with tobacco use and will kill almost 26,000 patients in the U.S. in 2013. Therefore, there is a great need for new therapeutic strategies. We now propose a new team science project to exploit Myxoma virus (MYXV) as a novel viral-based therapeutic that is harmless for normal human tissues but targets and kills SCLC. Projects 1 and 2 will take advantage of our unique resources of i) a large collection of human SCLC tumor samples for testing and optimizing the killing of tumor cells in the laboratory and in specialized animal models, ii) a new genetically engineered SCLC mouse model for further confirmation of safety and efficacy testing, iii) our expertise in optimizing MYXV to enhance immune-mediated cell killing to maximize our ability to cure this disease, and iv) our promising preliminary data already showing efficient MYXV infection and cell killing of human and mouse SCLC in vitro and in vivo. Project 3 focuses on a pioneering clinical program in navigational and interventional bronchoscopy that is only available at the University of Florida and which allows for studying the efficacy of MYXV on fresh SCLC samples with plans for future investigator-initiated clinical trial using intralesional delivery of this therapy. We are making a strong effort to develop tobacco-related SCLC as an important topic for clinical and translational research in the state of Florida.

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Sheng Wei, MD	H. Lee Moffitt Cancer Center & Research Institute	Nanoparticlebased targeting of miR183 for immunotherapy of lung cancer	This application offers a new approach to lung cancer immunotherapy that has never been tried before. It is based on our novel observations of immune suppression in the tumor microenvironment. Circulating immune cells function to survey the body for abnormal cells and a type of immune cells, called natural killer (NK) cells are especially potent in seeking out tumor cells through multiple receptors that only bind unique proteins that appear on nascent tumor cells. These receptors utilize a key protein called DAP12 to anchor to the NK cell surface. However, we find that tumor cells produce a protein called transforming growth factor-b that can disrupt NK cell function by depleting DAP12. This is accomplished by activation of a microRNA, miR183 that specifically binds to the DAP12 gene to destroy it. Therefore NK cells cannot display their receptors on the cell surface and become blind to the surrounding tumor cells, allowing tumor cells to escape immune detection. We also find that nicotine from tobacco smoke, long associated with lung cancer development, can activate the same mechanism in NK cells. Without immunity, cancer cells can grow unchecked. This is the first report of a microRNA that controls immune cells in lung cancer and the targeting of this microRNA presents a highly innovative and new strategy to treat cancer. Before attempting this therapeutic approach in man, it is critical to first obtain proof of concept in mice. We have established a working model in immunodeficient NSG mice which can accept human tumors and human NK cells. Human NK cells, if implanted in mice the same time as the tumor, can readily eliminate the tumor, but if implanted after the tumor has grown for a week, an immunosuppressive environment is already established that renders NK cells impotent. This model recapitulates what is occurring in human cancer patients and will be used to test formulations of anti-sense miR183 to treat lung cancer. Nanoparticles, made of poly(lactide-co-glycolide) (PLGA) that have long been in medical use in dissolvable sutures and proven safe, will be used to deliver anti-sense miR183. These nanoparticles will then be injected into human tumor-bearing mice along with human NK cells to evaluate if NK cell recovery is obtained. To optimize the nanoparticles to reach NK cells in the tumor bearing mice, we will functionalize them with NK homing molecules and facilitate their entry into the NK cells by attaching cell penetrating peptides to the nanoparticles. If efficacy is demonstrated with these nanoparticles, this method would revolutionize the treatment of lung cancer. Moffitt Cancer Center is poised to conduct these trials because it has an outstanding immunotherapy research program and admits about 800 new lung cancer patients annually. Several advantages are associated with such nanoparticles. They can serve as off-the-shelf reagents and it can be administered not only to any lung cancer patient, but also to patients of other tumor types. It is well accepted that immune escape is a hallmark of all cancers and our product could be a universal anticancer agent.

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Vani N. Simmons, PhD	H. Lee Moffitt Cancer Center & Research Institute	Facilitating Smoking Cessation with Reduced Nicotine Cigarettes	Cigarette smoking remains the top avoidable cause of death and disease in Florida, responsible for most cases of lung cancer, as well as many other cases of cancer, heart disease, pulmonary disease, and diabetes. Despite the state-supported availability of multiple avenues for smoking cessation (e.g., telephone counseling, web-assisted cessation, in-person counseling, free nicotine replacement therapy), only a small percentage of smokers within Florida avail themselves of these treatment options, and approximately 18% of Floridian adults continue to smoke. Research over the past several decades has shown that long-term cessation rates with even the most intensive interventions rarely exceed 20-30%. Thus, it is vital that additional research be conducted to develop and validate novel methods for effective smoking cessation. It has long been understood that nicotine is the primary constituent in cigarettes and other tobacco products that supports the initiation and maintenance of an addiction to tobacco. The recent availability (via the NIDA Drug Supply Program) of research cigarettes with varying levels of nicotine creates a unique opportunity to evaluate the potential benefits of very low nicotine content (VLNC) cigarettes as a new tool for smoking cessation. In that vein, the proposed research will develop and test a novel smoking cessation strategy, based on theory and research concerning extinction. Specifically, this theory-driven intervention will be designed to extinguish the expectation of reinforcement from smoking via pre-quit smoking of VLNC cigarettes. In turn, this should result in a higher likelihood of successful quitting. First, our experienced research team and consultants will adapt and refine intervention materials, in order to provide smokers with clear and detailed instructions for smoking VLNC cigarettes prior to quitting, in a manner that will maximize extinction to smoking-related reinforcement. This intervention development process will involve expert review and recommendations, and will incorporate feedback from smokers (n=20) enrolled in a pilot study of the intervention. The pilot study will also examine the feasibility of the targeted intervention, along with two tapering schedules (immediate vs. gradual) for transitioning to VLNC cigarettes over a 4-week pre-quit period. Next, a randomized controlled trial (RCT; n=200) will compare effects of the targeted intervention vs. a standard intervention, in combination with the two tapering schedules, on smoking cessation outcomes. In addition, two behavioral paradigms will be administered to RCT participants, to determine intervention effects on smoking behavior in a short-term laboratory Smoking Analogue tasks and a 5-day abstinence challenge task. Finally, analyses will explore if the treatment works better for particular types of smokers, and if the treatment effects on smoking cessation outcomes are mediated by effects on related variables (e.g., decreased nicotine withdrawal or cravings to smoke). This information will be particularly useful for determining which smokers may benefit most from this smoking cessation approach, and for better understanding how this form of treatment works.

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Daiqing Liao, Ph.D.	University of Florida	Pharmacologic inhibition of acetyltransferase CBP/p300 as a new therapeutic approach for breast cancer	Although effective therapies exist for breast cancer (BC), many patients do not derive clinical benefits from current standard of care treatments such as endocrine therapies for estrogen receptor-positive (ER+ subtype), trastuzumab for HER2-enriched BC, and chemotherapies for the triple-negative (TNBC) subtype. Therefore, it is imperative to identify new therapies that increase response rate to current therapies and overcome drug resistance. ER depends on coactivators to activate gene expression underlying tumor growth. CBP (also called CREBBP) and p300 are key ER coactivators, and found to be overexpressed in the majority BC cases. By assisting ER and other oncogenic proteins, CBP/p300 are active in cancer cells of all BC subtypes, and thus pharmacologic inhibition of CBP/p300 should suppress the growth and progression of primary breast tumors of all BC subtypes. Candidate drug compounds that target CBP/p300 have been identified in our preliminary studies and will be tested for their effectiveness in suppressing tumor growth and metastasis of major BC subtypes. Importantly, we will determine whether the new agents can enhance therapeutic effects of endocrine therapy (e.g., tamoxifen), chemotherapies (e.g., docetaxel) as well as HER2-targeted therapies (e.g., trastuzumab). Patient safety is of paramount concern for any drug development effort. Therefore, the safety and pharmacologic properties of new CBP/p300 inhibitors will be stringently assessed to ensure that they will be suitable for clinical use. These novel agents, specifically targeting CBP/p300, should be effective for suppressing the growth and metastatic progression of primary breast tumors. The new inhibitors are small-molecule compounds and are thus suitable for various systemic treatments, such as via oral administration. Drugs that target CBP/p300 have not yet been developed, and thus new agents emerging from this project could have an unprecedented impact on treating BC, contributing to the ultimate goal of ending breast cancer

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Maria Jose Miguez, M.D., Ph.D.	Florida International University	Biobehavioral Intervention For Smokers Living With HIV	While substantial progress has been made in reducing smoking prevalence to 20% among the general population, smoking ranks among the top preventable causes of death and disease in Florida. Unfortunately, these statistics are even more grim for people living with HIV. The smoking rates among this population are two to four times higher, thus increasing the risks of numerous diseases and threatening health gains achieved with antiretroviral treatment. As a result, PLWH receiving ART lose more years of life to smoking than to HIV, with mortality tripling compared to the background population. Unfortunately, research on smoking cessation interventions for PLWH has been scarce and has provided disappointing results. Thus, the long-term goal of this program is to develop a tailored intervention that can be readily integrated into HIV treatment settings. Such research is critical given that Florida remains the epicenter of the HIV/AIDS epidemic in the United States. Our formative work indicated that people living with HIV receiving antiretroviral therapy exhibited significantly higher levels of plasma nicotine. These heightened levels may be hampering smoking cessation efforts and success rates. The overall objective of the transdisciplinary team of HIV/tobacco funded researchers is to test if tailoring nicotine replacement doses to temper these excessive levels will enhance the efficacy of the intervention. This will be accomplished by: 1) assessing pre-trial plasma levels, 2) determining participant genotype (which to our knowledge has not been used in cessation studies among PLWH) , and 3) providing tailored feedback to the participants based on the assumption that the higher the knowledge and perception of risk the higher the interest in modifying risky behavior(s). To test our proposed model based on proven smoking prevention and control methods, 500 PLWH ready to quit smoking will be enrolled in a double-blind, randomized clinical trial with intent-totreat design. We will be comparing a standard, well-validated, brief smoking intervention that follows NCI guidelines and will consist of brief advice + nicotine replacement therapy (NRT) versus the tailored one (brief smoking intervention + personalized doses of NRT). The primary outcome for this study will be the rates of smoking cessation, point prevalence abstinence (prior 7 and prior 30 days), and verified continuous abstinence 3-, 6-, and 12-months post scheduled quit day. Building on the strengths of the SHARC center funded by NIAAA, and our FILTERs center funded by the Florida Department of Health, the knowledge gained here could potentially contribute a more complete bio-behavioral model to the intervention field. The proposed study, if successful, will provide a new tailored, replicable, and manual-based intervention for people living with HIV. It can also provide much needed information about the key mediators and moderators of smoking cessation interventions in this vulnerable population. Our long term goal is to reduce the burden of one of the most devastating causes of morbi-mortality in our time and improve overall quality of life. The study could also pave the way for the future development of tailored smoking interventions for other populations (e.g., menthol users, older women, adolescents, older adults).

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Jong Park, Ph.D., M.P.H., M.S.	H. Lee Moffitt Cancer Center & Research Institute	Biobank for African American Prostate Cancer Research in Florida	Prostate cancer disproportionally affects men of African Ancestry (AA) who have much higher incidence and mortality rates than Caucasian men. In the state of Florida, approximately 2,000 AA cases were reported every year according to the Florida Cancer Data System (FCDS) of the Florida Department of Health. The relationship between risk of prostate cancer and smoking is a matter of debate. However, male smokers have higher level of blood male hormones, thus androgens, which may increase prostate cancer risk and progression. Recently, a large study with more than 20,000 prostate cancer patients found a consistent risk (11-22%) increase for prostate cancer among current smokers. Moreover, significant increase of risk (24-30%) for advanced prostate cancer, such as lethal cases, was observed among current smokers. We agree on the urgent need for a statewide biobank to support prostate cancer research among men of African Ancestry in Florida. It has not been initiated due to various reasons, such as limited resources to establish the infrastructure for collaborative data and biospecimen collection. Successful completion of this proposed project will lead to the development of an extremely valuable research asset for health disparity studies for prostate cancer. We anticipate this resource will contribute not only to generate important scientific findings but also allow researchers to leverage additional national funding, such as NIH, or DOD and ultimately lead to better strategies to reduce prostate cancer incidence and mortality. We will also investigate the effect of smoking on aggressiveness of prostate tumor by various mechanisms. For example, we and other investigators previously reported that smoking causes mutations in various tumor suppressor genes, influencing molecular pathways to change behaviors of tumor and increased cancer progression. We will address the shortcomings of these efforts with a systematic recruitment of all AA prostate cancer patients (n=6,000) who diagnosed between Jan 2013 and Dec. 2015 using a well-organized protocol, which was guided by FCDS and our research team.

Fiscal Year 2015-2016 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Jang-Yen Wu, Ph.D.	Florida Atlantic University	Granulocyte colony-stimulating factor (G-CFS) gene therapy for stroke	Granulocyte colony-stimulating factor (GCSF) is a FDA-approved drug for enhancing hemopoiesis. In addition, we as well as others have shown that GCSF has neuroprotective and neurogenesis properties in animal models of stroke and other neurodegenerative diseases, e.g., Parkinson's disease. Protein therapy using GCSF is attractive because GCSF is well tolerated after systemic delivery. However, its plasma half life is about 4 hours; moreover, there is potential for chronically elevating white blood cells during repeated delivery. Also, no reliable monitoring system is available to follow the progression of stroke treatment. Here we propose one alternative approach which is to administer well regulated GCSF gene delivered by a viral vector –namely a replication deficient adeno-associated virus (AAV) in conjunction with a non-invasive imaging system for monitoring delivery and expression of GCSF in the brain. Recently we have developed an innovative MRI technology which comprises a phosphorothioate-modified antisense (AS) oligo DNA (sODN) to hGCSF which is conjugated to a superparamagnetic iron oxide nanoparticles (SPION) known as hGCSF-AS-sODN-SPION probes and demonstrated that increased expression of hGCSF is associated with increased survival rate in BCAO stroke mice model (Gene Therapy, doi:10.1038/gt. 2015.81; 24July, 2015). Specifically, the following specific aims will be addressed: Aim 1 – We will deliver GCSF gene using AAV-CMV-hGCSF or AAV-CMVHRE-hGCSF vectors in mice BCAO stroke model as well as in cell cultures stroke (hypoxia) model and to determine the mode of neuroprotective function of expressed GCSF. Aim 2 - To further elucidate the mechanism of expressed GCSF in neurogenesis. The significance of this project is that we have already demonstrated the proof of concept of GCSF gene therapy for ischemic stroke. Furthermore, it is highly feasible that the proposed work will lead to securing a patent and that it may facilitate its eventual commercialization and clinical use for stroke treatments.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Helen M. Bramlett, Ph.D.	University of Miami	Whole Body Vibration Improves Stroke Outcome in Nicotineexposed Rats	Millions of smokers are disabled as a result of stroke and ischemic stroke accounts for almost 85% of total stroke cases. Ischemic stroke occurs when the blood supply to part of the brain is disrupted due to thromboembolic occlusion of a cerebral artery. Disruption of blood supply to part of the brain causes focal ischemia damaging the cortical region initially. To date, the only drug that has been approved to treat acute stroke is the clot-dissolving drug tissue plasminogen activator (tPA). However, tPA must be administered within 3 hours of the onset of an ischemic stroke, which makes it a viable treatment for less than 15% of stroke patients. Thus, new therapies for acute stroke with extended therapeutic windows are badly needed. Physical therapy and exercise have been shown to be beneficial for recovery but often are not an option for frail patients. Whole Body Vibration (WBV) mimics the internal forces exerted on by exercise, and can be effectively incorporated in any patient's treatment regimen. Although WBV has been previously shown to be beneficial in maintenance and increase of bone mass, in this study we want to test its direct application in the recovery from stroke. We hypothesize that WBV will significantly improve cognition, inflammation and neuron growth in nicotine exposed rats after stroke.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Christine Chung, M.D.	H. Lee Moffitt Cancer Center and Research Institute	Molecular Signatures of Immunotherapy Response and Improved Survival in Tobacco-related Head and Neck Cancer	Head and neck squamous cell carcinoma (HNSCC) remains one of the most devastating cancers affecting oral cavity, oropharynx, hypopharynx, and larynx that are critical structures for life's most essential functions such as eating, breathing, and talking. Common risk factors are tobacco and alcohol use and human papillomavirus (HPV) infection. The patients with tobacco-related HNSCC have the worst prognosis compared to the HPV-related HNSCC. Even within the HPV related HNSCC, patients have worse outcome if they have history of smoking compared to nonsmokers, demonstrating the devastating effects of tobacco use in cancer development, treatment resistance and cancer-related death. Recently immunotherapy has become a promising therapeutic option in HNSCC. Among the numerous immunotherapeutic agents, programmed cell death-1 (PD-1) inhibitors are the most advanced in development in HNSCC, particularly pembrolizumab and nivolumab. PD-1 is an important protein that regulates the immune cell functions which are critical in recognizing and eliminating the abnormal cancer cells. Activation of PD-1 can decrease this immune function by suppressing T cells. Thus, inhibiting PD-1 improves the ability of T cells to fight the cancer. These immunotherapy agents set themselves apart from chemo- and other therapies by their ability to induce long lasting clinical benefits leveraging the patient's own immune system; however, the efficacy is seen only in a limited number of patients. Only 13-18% of HNSCC responds to these agents, and long term toxicities have not been fully defined because it is very new treatment approach. It is imperative to identify patients who will truly benefit from these immunotherapy agents, to improve the current response to immunotherapy, and accurately assess the toxicities as we move towards more personalized therapies. In this project, we propose; 1) to identify predictive biomarkers to select the patients who will benefit the most from current PD-1 inhibitors based on their tumor genetic alterations that may trigger the immune response, 2) to determine whether combining cetuximab, a currently FDA-approved cancer drug, and nivolumab will improve the immune response and clinical benefits, 3) to determine changes in the tumor infiltrating immune cells induced by tobacco use, 4) to determine changes in the tumor genes and proteins induced by tobacco use in order to find new drug targets, and 5) to develop a smartphone-based assessment for real-time reporting of toxicities and tobacco use by the patients between the clinic visits. The findings from this project will have a significant impact in the health of Floridians by reducing mortalities through improving the selection of patients who will gain the most benefit from receiving PD-1 inhibitors, reducing mortalities by a novel combination therapy, and reducing morbidities of treatment-related toxicities and on-treatment tobacco use through development of a real-time assessment by patient reporting.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
W. Dalton Dietrich, Ph.D.	University of Miami	The Therapeutic Effect of P7C3- A20 on Stroke	Focal cerebral ischemia leading to stroke is a devastating condition that has few therapeutic interventions available except for early thrombolytic therapy or new catheter-based endovascular strategies. During severe cerebral ischemia, cells deplete their oxygen and energy reserves, leading to neuronal cell death and prolonged behavioral deficits including motor and cognitive impairments. There is therefore a major need to develop and test new pharmacological agents to protect neurons from irreversible cell death. In addition to cell death, several studies have reported an increase in the generation of new neurons in specific brain regions following focal cerebral ischemia. This cellular response is believed to potentially provide an endogenous repair mechanism that could improve outcome by replacing injured neurons. However, the majority of these newly formed cells undergo cell death. Therapeutic strategies that also protect these newly formed neurons for death would potentially promote functional recovery after stroke. The recently identified proneurogenic compound P7C3-A20 has been reported to inhibit neuronal cell death, enhance the formation of new neurons and improve cognitive function in several neurodegenerative models. The goal of this project is to determine for the first time whether treatment with P7C3-A20 at various periods after the focal ischemic insult would decrease overall brain pathology, reduce the death of the newly formed neurons and improves long term motor and cognitive function. Proposed studies will investigate the therapeutic window for treatment effects and clarify a potential causative role of new neuron cell survival in the improved outcomes. To conduct this study, a transient middle cerebral artery occlusion model in rats and mice will be used to examine sensorimotor and cognitive behavioral outcomes over chronic survival points. The generation of new neurons after focal ischemia will be examined in two distinct areas of the brain (subgranular and subventricular zones) that are known to demonstrate neurogenesis after injury. Special staining approaches will be used with novel tissue clearing technique combined with 3-dimensional reconstructions to resolve cellular responses to injury and treatment. Finally, a special transgenic mouse model (Nestin-TK-GFP) will be used to independently manipulate the degree of neurogenesis and determine a causal link to improved functional outcomes. We will make use of established research guidelines to reduce experimental bias and enhance reproducibility of findings. The major endpoints of the proposal are therefore to determine if P7C3- A20 is a viable therapeutic strategy after experimental focal ischemia and to clarify potential mechanisms of action including enhanced neurogenesis. Importantly, if found to be effective, this neuroprotective treatment could be combined with available endovascular approaches to maximize protection and recovery.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Jhanelle Gray, M.D.	H. Lee Moffitt Cancer Center and Research Institute	Targeting Immunosuppressive Cancer Associated Fibroblasts and Immune Checkpoints in NSCLC	Lung cancer causes more cancer deaths than breast, colon, prostate, and pancreatic cancer combined. It is an immunotherapeutically responsive cancer. Immune checkpoint inhibitors, including anti-PD1/PDL1 therapies, produce improvements in median overall survival from 12 to 24 months, with some durable responses. As dramatic as these results are, less than half of patients benefit. There are many other mechanisms that may suppress the immune system, including in the tumor microenvironment, which can lead to escape from tumor killing by immune cells. Combination strategies that interfere with the different immunosuppressive mechanisms operational within the tumor microenvironment are of interest in lung cancer immunotherapy. We recently discovered that the agent nintedanib (FDA approved for idiopathic pulmonary fibrosis; approved in Europe for combination with chemotherapy in lung cancer), which blocks multiple receptors including fibroblast growth factor receptors, has the potential to be repurposed as an anti-cancer immunotherapeutic, abolishing the immunosuppressive influence of cancer-associated fibroblasts (CAFs). CAFs are the most prominent cell type in the tumor stroma and differ from normal fibroblasts as they are continuously activated. At Moffitt (Antonia Lab), we developed a technique to grow out CAF cell lines made from human lung cancer tumors. In this model, T cells are strongly inhibited in the presence of CAFs due to expression of immune checkpoints and other immunosuppressive enzymes. Based on our preclinical work and the literature (which support our findings), we hypothesize that targeting immunosuppressive CAFs within the tumor microenvironment in combination with immune checkpoint blockade with nivolumab may translate into better tumor control. Our project is unique and innovative in that, while many studies are solely evaluating various checkpoint inhibitor combination strategies, we recognize the importance and have provided a novel, rational, feasible and scientifically sound approach to also target a unique source of immunosuppression within the tumor microenvironment. The significance of this project is that by immune suppression blockade, we are primed to increase the immune-mediated tumor responses, identify markers that can better predict tumor shrinkage while reducing waste and toxicity (precision medicine), and enrich patient treatment algorithms and ultimately improve outcomes for patients with nonsmall cell lung cancer. With these goals in mind and based on the above, we developed a phase IB/II clinical trial with serial blood collections and tumor biopsies to evaluate the combination of nintedanib with immune-checkpoint blockade (nivolumab). Objective #1. To determine the safety and efficacy of nivolumab plus nintedanib in advanced lung cancer. Objective #2. To correlate key markers obtained from serial tumor biopsies and blood collections with response to treatment. Objective #3. To correlate key markers obtained from serial tumor biopsies with resistance to treatment.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Zhihua Jiang, Ph.D.	University of Florida	Mechanisms for Tobacco Smoke to Modulate Aortic Aneurysm Development	Aortic aneurysm stands as the 15th leading cause of death in the United States. This aortic disease affects 5% of the general population, with the incidence being 3-5 times higher in smokers than in non-smokers. Furthermore, tobacco-use doubles the rate of aortic dilation and the risk of rupture. Although quitting tobacco has proven benefits of halting further escalation of the aortic aneurysm, yet around 42 million Americans continue to smoke. Currently, mechanisms underlying tobacco smoke-exacerbation of aortic aneurysms are poorly understood. Strategies capable of reducing or eliminating the deleterious effect of tobacco smoke on aortic aneurysm development remain unavailable. Studies for lung cancer and chronic obstructive pulmo-nary disease have generated rich knowledge about the impact of tobacco smoke on the biology of endothelial cells, smooth muscle cells (SMCs), and immune cells. A commonly held view is that tobacco smoke impairs the body's defense mechanisms via suppressing the function of the immune system, provoking dysfunction and death of endothelial cells, and accelerating proliferation of SMCs. However, this theory cannot explain the clinical presentation of aortic aneurysms where intense inflammatory infiltrates are located across the aortic wall and fewer SMCs are present in the tunic media. A large body of clinical and experimental evidence supports the concept that aortic aneurysm is an inflammatory disease. Recent advances in immunology have identified two different types of inflammation, with each type of inflammation driven by a distinct subset of immune cells and cytokines. Specifically, the type 1 inflammation is governed by a subset of Tcells, called type 1 T helper or TH1 cells whereas the type 2 inflammation is dominated by TH2 cells. Under physiological conditions, the function of TH1 and TH2 cells is well-balanced to maintain tissue homeostasis. Interestingly, epidemiological investigations have shown that compared with the general population, diabetic patients are two times less likely to develop aortic aneurysms while asthma pa-tients are at a 2 times greater risk of developing aortic aneurysms. A detailed characterization of the immune system of these patients has uncovered that diabetic patients have a TH1-baised immunity whereas asthma patients have a TH2-biased immunity. These studies have established a correlation of aortic aneurysm formation with the TH2-biased immune responses. As opposed to the traditional belief that tobacco smoke suppresses the immune function, emerging evidence suggests that it skews the TH1/TH2 balance toward a TH2 phenotype. This finding has provided a plausible explanation to the protective effects of diabetic condition and the detrimental effects of asthma on aortic aneurysm formation. It appears that tobacco smoke exacerbates aortic aneurysms via shifting the TH1/TH2 balance to a TH2 phenotype. In this project, we will use both genetic and pharmaceutical approaches to test this hypothesis. Results generated in this project will not only deepen the mechanistic understanding of tobacco ex-acerbation of aortic aneurysms but also provide information on the potential of FDA approved immune-modulators to eliminate the deleterious effects of tobacco smoke on aortic aneurysm development.

Fiscal Year 2016-2017 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Mandip S. Sachdeva, Ph.D.	Florida A&M University	Oral Nanotechnology in Triple Negative Breast Cancer	An estimated 1 million cases of breast cancer are diagnosed annually worldwide. Of these, more than 170,000 are described as triple-negative. Triple-negative breast cancer (TNBC) is defined by the lack of protein expression of estrogen receptor (ER) and progesterone receptor (PR) and the absence of HER2 protein over-expression. The complex nature of tumors represents a significant challenge to the health care system. Triple Negative Breast Cancer (TNBC) does not have a first line treatment. Development in this area will help many patients' especially the African American population which is disproportionately afflicted by it. Majority of the therapy is done in clinics by intravenous administration which involves repeated hospital visits and is cumbersome for patients. Development of an oral nanoparticle product of an already existing drug (Docetaxel which is given intravenously) in combination with another agent (Piperlongumine), which can significantly potentiate its activity in a synergistic manner against TNBC, will be of immense help to cancer patients allowing them to avoid the adverse effects involved with multiple parenteral injections and also avoid the need to go to the hospital. The ultimate goal in this proposal is to develop an oral nanoparticle capsule based formulation for the treatment of triple negative breast cancer with minimal toxicity and enhanced efficacy.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Gregg Fields, PhD	Florida Atlantic University	Inhibition of Tumor Cell Surface Proteolysis	Matrix metalloproteinase 14 (MMP-14)/MT1-MMP is a type I transmembrane cell-surface protease overexpressed in many tumors. The increased presence of MT1- MMP is associated with poor prognosis in patients with melanoma, small cell lung cancer, tongue squamous cell carcinoma, head and neck carcinoma, bladder cancer, and breast cancer, amongst others. Increased tumor cell production of MT1-MMP enhances tumor growth, invasion, and metastasis. Overall, the production of MT1- MMP correlates to poor prognosis in a number of tobacco-related cancers and the collagen-cleaving ability of MT1-MMP is critical to the progression of a number of tobacco-related cancers. A mechanistic examination of MT1-MMP at the cell surface would unravel the influences of cell surface binding partners on MT1-MMP activities, and set the stage for the development of unique MT1-MMP inhibitors. The present proposal seeks to utilize cutting-edge technologies to examine, on a molecular level, how a cell surface protease (MT1-MMP) functions in its native environment. In addition, the cell surface nature of MT1-MMP will be utilized to design novel inhibitors. The specific aims to achieve these goals are as follows: (1) quantitative analysis of MT1-MMP activity on the cell surface, including the modulation of activity by specific MT1-MMP domains and binding partners; and (2) development of inhibitors of MT1-MMP function based on one-bead-one-compound conformationally constrained libraries targeting secondary binding sites (exosites) within the enzyme. The present work will lead to a detailed, mechanistic understanding of cell surface proteolysis and the exploration of cell surface proteolysis inhibitors based on unique modes of action. Inhibitors will be characterized using three dimensional invasion models of melanoma.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Jennifer Permuth, Ph.D.	H. Lee Moffitt Cancer Center	The Florida Pancreas Collaborative Next-Generation Biobank: Reducing Health Disparities and Improving Survival for Pancreatic Cancer	Of all tobacco-related cancers in the United States, pancreatic cancer (PC) is the deadliest, with a five year relative survival rate of only 9%. PC just became the third leading cause of cancer deaths and will become the second leading cause around 2020. In contrast to breast, prostate, and colorectal cancer, incidence and mortality rates for PC are increasing. Florida ranks second in lives lost to PC each year. Striking racial disparities in PC incidence and mortality rates exist nationally and in Florida, with higher rates among African Americans (AA) compared to other racial groups. Reasons for these disparities remain unexplained. One factor that contributes to increased morbidity and mortality and diminished quality of life (QoL) in most PC patients is cancer cachexia, a metabolic condition characterized by stages of progressive muscle wasting, unintentional weight loss, and fatigue. Based on preliminary data generated by our team, we hypothesize that cachexia may influence racial disparities in PC such that AA may present with a higher prevalence of cachexia earlier in the disease process compared to Non-Hispanic Whites (NHW). We further hypothesize that biological correlates of cachexia represented by key imaging features and molecular markers may underlie the disproportionate burden of PC in AA and associate with worse outcomes. To reduce PC burden in Florida and simultaneously address racial disparities, we seek to expand upon an existing collaboration between Moffitt, the University of Florida, and the University of Miami known as the Florida Pancreas Collaborative (FPC). This infrastructure grant will enable teams from other Florida cancer centers that diagnose and treat a high volume of AA, Non-Hispanic White, and/or Hispanic individuals with PC to join forces with the FPC sites, with the goal of creating state resources to conduct basic, clinical, population-based, and translational science that will impact several racial and ethnic groups affected by PC. We aim to 1) prospectively build a robust 'next-generation biobank' that contains viable tissues, biofluids, medical images, and clinical and laboratory data, all derived from a racial/ethnically diverse cohort of PC patients, and 2) use the biobank to test the hypothesis that cancer cachexia may underlie racial disparities in PC. Immediate investment in this infrastructure is crucial since most PC research (including that related to cancer cachexia) has been performed using images, biospecimens, and data that do not represent racially diverse populations. In summary, this project will address a critical gap in PC research by capitalizing on Florida's large underserved minority PC population, an already established and productive multidisciplinary collaboration with new passionate partners, and proof-of-concept data. We will also foster a valuable state-wide resource for PC disparities research that will generate impactful findings related to cancer cachexia and enable Florida's researchers to compete for national funding to increase QoL and reduce PC burden, goals in line with the Florida Department of Health.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Nagi Kumar, Ph.D.	H. Lee Moffitt Cancer Center	Chemoprevention of Lung Cancer in Former Smokers	Although screening high risk populations using low dose Computed Tomography (LDCT) and smoking cessation programs are critical, former smokers on surveillance are eager to participate in chemoprevention interventions that can further reduce their risk for lung cancer. We and others have shown that curcumin (CUR) and omega 3 fatty acids (ω-3 FA)are effective at suppressing Stat3P and NF-κB signaling pathways- relevant to lung carcinogenesis- resulting in suppression of proliferation of human lung tumor lines and inflammation responses. More recently, strong evidence has emerged demonstrating the role and mechanism of ω-3 FA as specialized fat mediators, with anti-inflammatory, anti-proliferative and pro-resolving properties towards resolution of cigarette smoke-induced lung inflammation in former smokers. We and others have also shown that CUR when combined with ω-3 FA is bioavailable in the lung and produces a more robust antiproliferative effect in lung tumor tissue compared to when these agents administered independently. Based on this evidence, we hypothesize that a standardized formulation of CUR + ω-3 FA will target molecular pathways that are critical for lung cancers development, leading to a reduction in the overall size and density of nodules, in former smokers. We hypothesize that this will be mediated by reducing cell growth, inflammation and through pro-resolving effects in the lung and in the precancerous lesions or the nodules. We will test our hypothesis by using an experimental design and rigorously evaluating the safety, efficacy and validate the potential mechanism of a combination of ω-3 FA + CUR or placebo administered for 6 months in former smokers, age ≥55 years, with lung nodules detected during LDCT screening program. Results of the proposed trial may have immediate and significant benefit to former smokers and other high-risk populations towards lung cancer prevention.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Frederic J. Kaye, MD	University of Florida	Good Manufacturing Practice (GMP) Production to Allow Phase 1 Clinical Trial Testing Intralesional Delivery of Myxomavirus to Patients with Advanced Small Cell Lung Cancer	We have demonstrated efficient myxomavirus (MYXV) infection, late viral replication, and MYXV mediated cell killing of small cell lung cancer (SCLC) in vitro and in vivo with negligible effect on normal tissues. We have optimized the oncolytic viral backbone for efficient SCLC cell killing by targeted mutational inactivation of viral survival signals and we have confirmed dramatic SCLC cell killing in vivo using both immunodeficient human xenograft models as well as immunocompetent genetically engineered SCLC mouse models. We have also tested efficacy of myxomavirus cell killing in our mouse models combined with both anti-CTLA4 and anti-PD1 immunotherapy to enhance durable clinical response. We have recently published our experience with intralesional injection of different anti-cancer agents by navigational bronchoscopy directly into lung parenchymal and mediastinal/hilar chest tumor sites which is a unique UF College of Medicine resource (UF IRB protocol 2015400327). We also have a submitted manuscript under review with our preclinical data supporting this clinical trial proposal. We now propose a Phase 1 clinical trial to initiate a first-of-it kind direct intralesional study of the effectiveness of oncolytic virotherapy combined with antiPD1/CTLA4 immunotherapy in patients with advanced SCLC who have no other treatment options. This trial would be conducted, as a sponsored collaboration with DNAtrx in Houston and this funding opportunity would allow both completing GMP production and safety testing and would also allow a unique Investigator-Initiated clinical study for the State of Florida. We have already prepared a clinical trial synopsis for this Phase 1 study. Briefly, we would study safety of a dose escalation schedule using direct bronchoscopy injection of MYXV into biopsy proven sites of recurrent SCLC. Secondary aims would study the effectiveness of MYXV on injected tumor sites compared with adjacent non-injected tumor sites, and distant non-injected sites. Ultimately, we would study the effect of our optimal MYXV dosing on clinical outcome in patients with advanced SCLC who are receiving concurrent immunotherapy with anti-PD1 or dual anti-CTLA4/PD1 therapy.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Sergei Kusmartsev PhD	University of Florida	Dissecting the mechanisms of tumor-induced tolerance and immune suppression in bladder cancer	There is an urgent need for efficacious and well-tolerated therapies in metastatic urothelial bladder cancer, as even first-line chemotherapy is poorly tolerated in a large proportion of individuals. One of the most promising approaches for treatment of advanced bladder cancer is anti-PD-L1 therapy. It appears that high expression of PD-L1 may allow cancers to evade the host immune system. Programmed death-ligand 1 (PD-L1), which is expressed on many cancer and immune cells and is strong negative regulator of T-lymphocyte activation.. Blocking PD-L1 enhances the anti-cancer immunity. Many PD-L1 inhibitors are in development as immuno-oncology therapies and are showing good results in clinical trials. Recently published studies demonstrated that treatment of bladder cancer patients with anti-PD-L1 antibody results in high response rates, and importantly, that the likelihood of response can be increased by determining the PD-L1 status of tumor-infiltrating immune cells. In fact, previous biomarker analysis has focused on PD-L1 expression on tumor cells rather than tumor- infiltrating immune cells. More recent observations indicate that that PD-L1 expression of immune infiltrates on pre-treatment tissue positively correlated with outcomes. However, little known regarding mechanism(s) of that regulate PD-L1 expression in immune cells infiltrating bladder cancer tissues. Better understanding of such mechanism (s) could provide an opportunity to uncover the underlying pathways that cancer cells using to create the immune tolerance and evade immune surveillance. Furthermore, it will allow develop the novel modalities for treatment of advanced cancers. Recently, we found that bone marrow-derived myeloid cells, frequently infiltrating tumor tissues, upon contact with bladder tumor cells become macrophages with strongly up-regulate PD-L1 expression. These PD-L1-expressing macrophages exhibit strong immunosuppressive effects and able to eliminate the T lymphocytes through apoptosis. We also found that formation of PD-L1expressing macrophages mediated by tumor-produced glycosaminoglycan hyaluronan. Tumorderived hyaluronan specifically binds to its CD44 receptor expressed by bone-marrow myeloid cells, promoting development of PD-L1-expressing macrophages. Inhibition of hyaluronan synthesis in tumor cells with pharmacologic inhibitor or blockade of CD44 signaling in myeloid cells with antagonistic anti-CD44 antibody prevented formation of PD-L1-expressing macrophages. The overall goal of current proposal is to examine the specific mechanisms underlying hyaluronan-mediated formation of immunosuppressive PD-L1-expressing macrophages, regulation of PD-L1 expression and to investigate the therapeutic potential of targeting hyaluronan-CD44 link for the treatment of bladder cancer using an experimental tumor model and cancer patients. Two specific aims are designed to achieve a proposed goal. First, the mechanistic part of the study will be focused on the molecular mechanisms by which tumor-derived hyaluronan promotes formation of immunosuppressive PD-L1-expressing macrophages. In second part of this study we plan to evaluate the therapeutic anti-tumor activity of targeting hyaluronan synthesis and CD44 receptor in bladder cancer. Inhibiting of hyaluronan synthesis by tumors or targeting hyaluronan-mediated CD44 signaling could

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

			provide an attractive approach to break tumor-induced immune tolerance and unleash the anti-tumor immune response.
Principal Investigator	**Principal Investigator's Organization**	**Project Title**	**General Audience Abstract**
Linda Hayward, PhD	University of Florida	The role the gut-microbiome-brain axis in cardiovascular disease following prenatal exposure to nicotine	Smoking during pregnancy is a major risk factor for spontaneous abortion, prematurity, and low birth weight. Additionally, offspring of smokers have an increased incidence of chronic behavioral problems, obesity, and nicotine addiction. Although many of the problems associated with prenatal nicotine exposure have been documented, the mechanism(s) underlying these changes remain elusive. Emerging evidence now suggests that a common factor underlying many diseases may be an imbalance of the bacterial microbes in the gut or gut dysbiosis and associated changes in bidirectional communication between the gut and the central nervous system or dysregulation of the gut-microbiome-brain axis. At present little is known about how smoking influences the gut-microbiome-brain axis during pregnancy and nothing is known about how prenatal exposure to nicotine modifies the gut-microbiome-brain axis in adult offspring and whether sustained gut dysbiosis contributes to a life-long predisposition for obesity, cardiovascular disease, heightened anxiety, and/or nicotine addiction in the offspring. Our preliminary analysis of the fecal samples from 21 day old rats with prenatal exposure to nicotine demonstrates there are sustained changes in the gut-microbiome. This is paralleled by alterations in the expression of genes linked to obesity and cardiovascular disease in the hypothalamus, a region of the brain associated with physiological homeostasis or balance. The primary goal of this research proposal is to evaluate for the first time the impact of prenatal nicotine exposure on the gut-microbiome during two different time points: during pregnancy and later during adulthood in the offspring. We hypothesize that PNE induces changes in the maternal gut-microbiome and changes in the placental barrier, which exposes the fetus to elevated levels of microbial metabolites (short chain fatty acid, SCFAs), hormones (leptin), and inflammatory cytokines. Moreover, disruption of the prenatal environment promotes epigenetic changes in gene expression in the offspring brain and a sustained change in the offspring gut-microbiome. These alterations linked to the gut-microbiome-brain axis promotes obesity and related diseases in adulthood. Using a pre-clinical rodent model, changes in gene expression in the fetus and adult offspring will be monitored and the effects of the maternal gut-microbiome alone versus nicotine on the overall adverse effects associated with prenatal nicotine exposure will be determined. We also propose to identify whether associated changes in the gut-microbiome of offspring are linked to the predisposition to develop high blood pressure when offspring are re-exposed to SCFAs or leptin. Finally, we will evaluate whether a simple fecal microbiome transfer can reverse or attenuate adverse health outcomes associated with prenatal nicotine. The focus of this project is to provide new insights into potential novel and inexpensive therapies for individuals unwillingly exposed to nicotine in utero, as well as the 23% of Florida's population that are current tobacco smokers that also more likely to be obese, develop diabetes, and cardiovascular disease.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Ashok Saluja, PhD	University of Miami	Evaluating Mechanisms of Stromal Modulation by Novel Anti-Cancer Drug Minnelide	Tobacco smoking is one of the major risk factors for pancreatic cancer, a disease with very poor survival rates. The poor prognosis in this disease is attributed to the presence of a dense fibro-inflammatory stroma consisting of the extracellular matrix, stromal cells and the infiltrating immune population. This creates a complex tumor microenvironment that is conducive to an aggressive disease. However, several therapies targeting just the stromal component have often resulted in increased metastasis and poor outcomes. This suggests that targeting only stroma is not sufficient and that there is a need for an "ideal" therapy" that will not only target the stromal cells but will also target tumor cells and actively prevent the tumor-stroma-immune crosstalk. Minnelide, a water-soluble pro-drug of triptolide, developed by our group, has recently completed Phase I clinical trials and is currently awaiting Phase 2 trial. Our preclinical studies show that at a dose of 0.4mg/kg, Minnelide is an effective cytotoxic compound that targets stromal cells and multiple pathways in tumor cells while having almost no effect on normal healthy cells. Our recently completed Phase I trial shows that that maximum tolerated dose for Minnelide is 0.67mg/m2. This safe dose translates to 0.2 mg/kg in mice. Unfortunately, at this low dose Minnelide does not affect tumor epithelial cells and does not impact tumor growth. Intriguingly, our preliminary data suggest that at this dose, Minnelide depletes the stromal ECM, thus relieving the interstitial pressure on the blood vessels and leading to better drug delivery. Our preliminary data also show that at this low dose, Minnelide decreases proliferation, increases vitamin A accumulation and decreases synthesis of ECM in cancer-associated fibroblasts (CAFs), without any significant change in their viability, suggesting reversion to a quiescent state. Based on this observation, we hypothesize that at a lower dose, Minnelide inactivates the CAFs, pushes them to quiescence and modulates the tumor-stroma-immune crosstalk. Furthermore, we hypothesize that combination of this stromal modulator effects of Minnelide with cytotoxic effects of chemotherapy, e.g. gemcitabine should lead to strong anti-tumor effects. This hypothesis will be tested by 3 aims: Specific Aim 1: Evaluation of the effect of combination of Minnelide and gemcitabine in Animal models: We will evaluate anti-stromal low dose of Minnelide in combination with cytotoxic doses of Gemcitabine in multiple stroma-rich models of pancreatic cancer. Specific Aim 2: Elucidation of the Mechanism by Which Minnelide Induces Stromal Reprogramming. We will confirm our findings that Minnelide inactivates CAFs and pushes them towards quiescence. Specific Aim 3: Determine the effect of stromal modulation by Minnelide on oncogenic pathways in tumor cells and pro-tumorigenic immune pathways. We will evaluate how treatment of CAFs with Minnelide leads to modulation of Stroma-Tumor and Stroma-immune interaction for therapeutic gain. Specifically, we will study the impact of Minnelide treatment of CAFs on signaling pathways in tumor cells. The impact of Minnelide treatment of CAFs on its ability to modulate Th1/Th2 balance, Treg levels and CD8+ activity will also be evaluated.)

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Nipun Merchant, PhD	University of Miami	Reprogramming the Tumor Microenvironment in Pancreas Cancer to Enhance Immunotherapy	Pancreatic cancer (PDAC) remains a major therapeutic challenge because of its chemoresistance. Three major contributors to therapeutic resistance that have been difficult to overcome in PDAC are mutations in the KRAS oncogene, the presence of dense fibrosis in the tumor that acts as a barrier to drug delivery and prevents infiltration of immune cells that can attack tumor cells, and a tumor microenvironment (TME) that renders the tumor ineffective to immunotherapy. Our efforts at targeting proteins associated with mutant RAS, have shown that MEK inhibition (MEKi) results in reciprocal activation of STAT3 signaling, which confers therapeutic resistance and continued PDAC cell growth. Combined inhibition of JAK/STAT3 (STAT3i) and MEKi overcomes this therapeutic resistance following RAS inhibition by preventing STAT3 reactivation. We have now identified a novel mechanism showing that combined MEKi and STAT3i also inhibits tumor fibrosis and enhances infiltration CD8+ cytotoxic T lymphocytes (CTL) that can kill tumor cells while suppressing regulatory T cells (Tregs) and myeloid derived suppressor cells (MDSCs) in the TME that prevent the immune system from attacking the tumor. Combined MEKi and STAT3i also results in reduced tumor burden and improved survival in genetically engineered mouse models (GEMs) of PDAC. This change in the TME, however, is accompanied by sustained expression of proteins that render tumor cells ineffective to immunotherapy such as PD-L1/PD-1 and CTLA-4. Our preliminary results further show that combined MEKi and STAT3i with an antibody that targets PD-1 can harness the effects of these immune checkpoint inhibitors for an enhanced anti-tumor response. Therapeutic strategies that reprogram the tumor stroma to enhance the effects of T cells that attack the tumor and suppress the cells that make the tumor ineffective to immunotherapy are of paramount importance as they have the potential to revolutionize treatment for pancreatic cancer and improve clinical outcomes. Our central hypothesis is that MEKi and STAT3i will reprogram cellular components of the PDAC TME to stimulate infiltration of CD8+ CTLs and overcome the immunosuppressive milieu of PDAC to enhance the effects of checkpoint inhibition (CPI) for a durable and sustained anti-tumor response. This will be proven by the following specific aims: Aim 1: Determine if checkpoint inhibition with MEKi and STAT3i will improve survival in GEMs of PDAC? Safety and efficacy of MEKi/STAT3i and anti-PD1 and/or anti-CTLA-4 antibodies treatment response will be determined in two different GEMs of PDAC; Aim 2: Determine if changes in the tumor and immune microenvironment induced by MEKi/STAT3i and checkpoint inhibition result in a durable and sustained anti-tumor immune response in GEMs of PDAC. In this aim, we will also attempt to detect if the cellular changes in the TME, pre- and post-treatment can predict response; Aim 3: Determine the specific effects of inhibiting MEK and/or STAT3 in stromal cells or tumor cells on the immune cells in PDAC. This work will not only evaluate a novel treatment strategy for PDAC, but may uncover potential biomarkers of response to checkpoint inhibitors that may lead to further clinical investigation of immune checkpoint blockade with targeted therapies in other cancers such as colon cancer and lung cancer.

Fiscal Year 2017-2018 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Tomar Ghansah, PhD	University of South Florida	SHIP-1: A Potential New Molecular Target for the Treatment of Pancreatic Cancer.	Tobacco usage is still the single largest cause of preventable cancer deaths in the United States (U.S.). Individual tobacco smokers are twice to three times more likely to develop pancreatic cancer (PC). PC is ranked among the top five causes of cancer death in the U.S. and in the State of Florida. It has been predicted that PC will become the second leading cause of death in U.S. by 2020. PC is one of the deadliest cancers that has less than a five-year survival rate in most patients. One of the main reasons why there is a poor prognosis for PC is due to fact that there are no effective screening procedures for early detection. In addition, current treatments (immunotherapy and chemotherapy) are ineffective for PC patients due to the chronic inflammatory microenvironment. PC tumor-derived factors (TDF) cause the expansion of immunosuppressive regulatory Myeloid Derived Suppressor Cells (MDSC) and regulatory T cells (Treg), which suppress anti-tumor immune responses. Thus, the investigation of new molecular target(s) are warranted to develop effective treatment for PC. For instance, Src Homology 2-containing Inositol Phosphate-1 (SHIP-1) is a vital protein that regulates MDSC and Treg development and function which impacts tumor immunity. We have shown that PC dampens SHIP-1expression which corresponds with the loss of MDSC and Treg homeostasis and an increase in tumor burden in mice. Therefore, we propose that targeting SHIP-1 would reduce tumor-associated MDSC and Treg expansion, enhance host anti-tumor immunity and reduce tumor burden. We also recently discovered that Apigenin (API), a Casein Kinase 2 (CK2) inhibitor, acts as a better SHIP-1 enhancer than the current standard treatment, Gemcitabine (GM), in pancreatic tumor-bearing mice. We hypothesize that PC dampens SHIP-1 dependent signaling, causing increased MDSC and Treg activity, creating an inflammatory tumor microenvironment resistant to treatment. Rescuing SHIP-1 (e.g. using API as one of the tools) will reverse this trend, facilitating treatment of PC. The following Specific Aims will test this hypothesis: Aim 1. Determine the distinct role of SHIP-1 in immunosuppressive and effector immune cells in PC. Aim 2. Identify SHIP-1 dependent signal transduction events controlling MDSC and Treg subsets in PC. Aim 3. Test whether the restoration of SHIP-1 expression increases anti-tumor immune responses and improves current PC treatments. We will use orthotopic pancreatic cancer models (clinically relevant to humans with PC) and state-of–the-art technology to address the aims of this study. The expected outcome of this study may lead to the successful identification of a novel adjuvant therapeutic strategy that can improve PC patient's quality of life and survival. This proposal is in alignment with James Esther King (JEK) initiative to support research scientists in the State of Florida to develop new and better treatment options to reduce the death rates for PC patients.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Gloria Salazar, PhD	Florida State University	Nutritional Interventions to Alleviate Cardiovascular Disease Mediated by Tobacco Use	Smoking and aging are two major risk factors in cancer and cardiovascular disease development (CVD). Although recent reports showed that smoking stimulates senescence in the lung, it is unknown whether smoking also accelerates senescence of the cardiovascular system. In this proposal, we proposed the novel hypothesis that cigarette smoke and nicotine accelerate vascular senescence promoting the development of atherosclerosis. We hypothesize that aging and smoking activate a common molecular mechanism that depends in part on the NADPH oxidase Nox1 and activation of the senescence associated secretory phenotype (SASP), a process by which senescent cells modify the microenvironment inducing inflammation, oxidative stress and tissue dysfunction. We demonstrated that polyphenols isolated from blackberries reduce oxidative stress and senescence induced by angiotensin II (Ang II), a strong stimulator of senescence and CVD, by inhibiting Nox1 in vascular smooth muscle cells (VSMCs). Further, overexpression of Nox1 and nicotine induce senescence. Our novel preliminary data show that blackberry supplementation reduced senescence and atherosclerosis in ApoE knock out mice in vivo and that nicotine alone is enough to increase atherosclerosis in the ApoE knock out mice. Nox1 produces superoxide and has a dual role in CVD and cancer. In the cardiovascular system, Nox1 activation by Ang II promotes atherosclerosis and hypertension, while in the lung Nox1 promotes metastasis of lung cancer cells. In this proposal, we will test the hypothesis that inhibition of Nox1 by blackberry polyphenols reduces the SASP, thus diminishingsenescence and atherosclerosis caused by tobacco smoke. We will test this hypothesis through the following aims: 1) determine the contribution of Nox1 to the development of the SASP and senescence of VSMCs induced by cigarette smoke and nicotine; 2) define the molecular mechanism by which blackberry polyphenols regulate the Nox1/SASP pathway to reduce senescence of VSMCs; and 3) determine the role of blackberry polyphenols in vascular senescence and atherosclerosis induced by cigarette smoke and nicotine *in vivo*.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Shelley Tworoger, PhD	H. Lee Moffitt Cancer Center and Research Institute	Early life exposures and risk of developing ovarian cancer	Ovarian cancer is the fifth leading cause of cancer death in the U.S., and sixth leading cause in Florida. Since most cases are diagnosed at an advanced stage, identifying novel risk factors is crucial to reduce incidence and mortality. Several lines of evidence suggest that early life exposures may be relevant to ovarian cancer risk. Most ovarian cancer risk factors (e.g., parity, oral contraceptive use) occur during childbearing years, suggesting a susceptibility window earlier in life. Also, in our own data, a larger body size at age 10 was associated with reduced ovarian cancer risk while body mass index during adulthood was associated with higher risk. These data suggest that exposures occurring during a critical period in early life may uniquely influence risk. Our objective is to evaluate several early life exposures (cigarette smoking, social adversity and abuse, and physical activity) and risk of ovarian cancer in later life. Better understanding of the role of early life factors in ovarian cancer risk may help inform development of targeted prevention strategies. First, current cigarette smoking is associated with a 2-fold increased risk of mucinous ovarian cancer tumors. Yet, few studies have examined the potential impact of early life exposure to cigarettes on risk. We will generate novel data on ovarian cancer risk in relation to age at smoking initiation, having a parent that smoked inside the home during childhood, and having a mother that smoked during pregnancy. In addition, in our own data we observed a 2-fold increased risk of ovarian cancer among women with 67 PTSD symptoms vs. those that did not experience trauma. Early life stress is a common underlying cause of PTSD and other distress disorders in adulthood, and has a potential role in altering ovarian development at the time of exposure. Thus, we propose to examine ovarian cancer risk in relation to early-life social adversity and abuse. Finally, our initial data suggest higher levels of adult premenopausal physical activity modestly increase ovarian cancer risk while postmenopausal physical activity has no impact on risk. To extend these findings, we propose to examine associations with physical activity during middle school, high school, and ages 1822. These data will help fill in gaps in knowledge about how physical activity may impact risk of ovarian cancer differently over the life course, and whether physical activity profiles could be useful in risk prediction. To conduct these epidemiologic studies we propose to leverage data from the prospective, ongoing Nurses' Health Study cohorts which enrolled >200,000 women across the U.S. Since enrollment, participants have completed biennial questionnaires about their medical history, health behaviors, and early life exposures. Ovarian cancer cases are identified by self report and death registry, and confirmed by medical record review or cancer registry linkage. For each proposed analysis, we will analyze risk overall and by histotype. Secondarily, among cases with archived tumor tissue, we will measure tumor immune cell profiles, allowing assessment of associations with tumors that exhibit an immunosuppressive signature. Overall, we propose to use a unique life course approach to improve identification of women at high-risk of ovarian cancer. Our findings, together with other known health effects of these exposures in early life, will give further support for implementing novel interventions in this time period.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Miguel Villalona Calero, MD	Miami Cancer Institute, Baptist Health South Florida	Assessment of Efficacy of Immunotherapy in combination with PARP inhibition in advanced cervical cancer patients functionally competent or deficient for the Fanconi Anemia repair pathway	Cervical cancer is the third most common gynecological cancer in the US, and women who smoke and are HPV positive have up to 3 times the risk of developing cervical tumors compared to nonsmokers. The incidence remains elevated in the Hispanic population, who also present with more advanced disease and worse mortality. Notable disparities in this ethnic subgroup correlate with poor access to healthcare and lower socioeconomic status. Hispanics make up the largest ethnic minority in Florida, therefore, the disease is an important public healthcare concern in the state today. A newly approved option in second line treatment for advanced cervical cancer is the immune checkpoint agent pembrolizumab, an IgG4 monoclonal antibody which blocks binding of PD1 to PDL1 and PDL2, helping restore T-cell immune response. This was based on a very low overall response rate (ORR) of only 13.3%. Therefore, better strategies are needed to increase the efficacy of immune checkpoint blockade and a proposed concept is by increasing tumor mutational burden (TMB) and neoepitopes expressed on cancer cells. This could be achieved through combining immunotherapy agents with compounds causing DNA damage or inhibition of DNA repair, such as poly(adenosine diphosphate [ADP]–ribose) polymerase (PARP) inhibitors, which lead to accumulation of DNA single-strand and consequently double-strand breaks in patients with BRCA mutated tumors who are innately deficient in homologous repair (HR). BRCA genes collaborate with several others in the Fanconi Anemia (FA) HR pathway, so we developed an immunofluorescence based method, FancD2/DAPI/Ki67 (FA Triple Stain Immunofluorescence FATSI), which permits the observation of FancD2 foci formation (or lack thereof) in the nucleus of proliferating cells in paraffin embedded tumor tissues. We screened over 600 patients in a clinical trial and found a functional deficiency in 29% of solid tumors. We also showed it is safe to administer the PARP inhibitor veliparib combined with the DNA damaging agent mitomycin C to patients with FA deficient tumors. We are completing a trial of pembrolizumab in solid tumors functionally competent or deficient for the FA repair pathway with encouraging results in 2 cervical cancer patients with Hispanic ethnicity. Therefore, we hypothesize that cervical cancer patients will have improved responses to pembrolizumab when given in combination with olaparib, a potent PARP inhibitor, and the FATSI assay could serve as an indicator of tumor response to immune checkpoint inhibition in FA deficient tumors. In order to support this hypothesis, we will undertake the following research aims: 1) Assess the efficacy of the combination of pembrolizumab and olaparib in patients with advanced cervical cancer. 2) Investigate whether functional deficiencies in the FA pathway of cervical cancers will correlate with improved response to the combination. To accomplish Aim 1, we will perform a phase II efficacy study of the combination in advanced cervical cancer patients after failure of first-line standard therapy at the Miami Cancer Institute and other Florida satellites. The primary objective is immune-ORR to the combination and the study will accrue a total of 44 patients in a Simon two-stage design for goal efficacy of >35%. To accomplish Aim 2 we will perform the FATSI assay, next generation sequencing and TMB of tissue and blood specimens of patients at baseline and correlate with response to therapy.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Alicja Copik, PhD	University of Central Florida	Adoptive PM21-NK cells with PD-L1 blockade for treatment of lung cancer	Non small cell lung carcinoma makes up 85% of all lung cancer cases and is the leading cause of cancer--related death. Although immunotherapy with checkpoint inhibitors has been a breakthrough for patients with advanced stage lung cancer, the response rate is still low and many patients eventually relapse. This project aims to develop clinically translatable immunotherapeutic strategies for lung cancer treatment to increase the response rate to the approved checkpoint inhibitor therapies and lower relapse rate. To achieve the proposed goals, the project will leverage the unique capabilities of ex vivo expanded natural killer (NK) cells reprogrammed to be highly activated through exposure to membrane particles (PM21) or exosomes(EX21) derived from IL21 expressing feeder cells (K562mbIL2141 bbl, mb21FCs). These PM21-particle stimulated NK cells produce IFNgamrna in response to encounters with tumor cells to induce PDL1 expression. Induced PDL1 can be then targeted by humanized antiPDL1 and further enhance tumor killing by NK cells via antibody dependent cell cytotoxicity (ADCC). Killing via ADCC is more resistant to immunosuppression and represents the most powerful mode of NK cells cytotoxicity. NK cells are also known to recruit other immune cells, such as dendritic cells, as well as cytotoxic and helper T cells to further direct complete elimination of cancer. We hypothesize that this approach has the potential to tum "cold tumors", "hot" to greatly improve treatment outcomes. Our method using nanoparticles (PM21) and exosomes (EX21) derived from mb21FCs further introduces new therapeutic dimensions by 1) a feeder cell-free expansion and stimulation system that can produce high NK cell numbers; 2) persistence of response through repeat injections of activated NK cells; and 3) reprogramming of NK cells ex vivo or in vivo, without genetic modification of the immune cells. Specific Aims will test parameters to inhibit the immunosuppressive environment and enhance NK cell antitumor activity. The treatment of recurrent or relapsed cancer is a difficult challenge for traditional cancer therapies, but recent results from Phase 1 trials in acute myeloid leukemia (AML) using activated NK cells show a 30% decrease in relapse over historical data. Collectively, new alterations to NK cell-based therapeutics are expected to advance treatment responses in order to improve outcomes of patient with advanced stage lung cancer that did not respond to or relapsed on antiPD1/PDL1 therapy.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Ramzi Salloum, PhD	University of Florida	Clinically-Efficient Strategies to Address Tobacco Smoke Exposure in Pediatric Practice	Tobacco use and tobacco smoke exposure (TSE) remain the leading preventable causes of mortality and morbidity for families in Florida and nationwide. Tobacco cessation in parents adds an average of 7 years to their life, eliminates most of their children's TSE, decreases the odds that children become tobacco users, and improves the financial status of disadvantaged families. The pediatric setting presents unique and important opportunities to address parents' tobacco use to reduce TSE in children. The effectiveness of tobacco control strategies in clinical settings is well--established, yet, compliance in pediatric practice remains low. Consequently, it remains unclear how to best support the uptake and sustainability of delivering evidence-based tobacco control interventions to parents in pediatric practice. This gap must be filled to inform the development of integrated and sustainable support that will effectively reduce TSE in children and families. Our overarching goal is to design clinical support strategies to enhance the delivery of tobacco control interventions in pediatric practice that can be scaled for wider implementation. Training for providers and clinic staff promotes best practices for tobacco control in clinical care, but implementation remains insufficient due to barriers to clinical efficiency, including competing time constraints during an office visit. However, the diffusion of electronic health records (EHRs) into clinical practice increases opportunities to engage clinics in intervention approaches that are potentially more sustainable by capitalizing on existing clinical processes. We propose a two-pronged approach to enhance implementation: 1) training providers and office staff on current best practices; and 2) deploying a brief EHR-based intervention in conjunction with provider-engaged adaptations to fit the intervention into practice workflow. Specifically, we will assess the feasibility and efficacy of a scalable, automated EHR tool for tobacco screening and counseling, along with provider training on best practices, among diverse clinics in the OneFlorida Clinical Research Consortium. Using an NCI-designated Research Tested Intervention Program (RTIP), we developed and piloted an innovative HER-based process that *confidentially* screens parents pre or in-visit for the use of tobacco and nicotine products. Tobacco users watch a brief evidence-based tobacco control video tailored to the tobacco product(s) they use, as assessed through the screening process. The PCP receives the screening results in the EHR to enable counseling in addition to EHR-based referral to cessation services. We will pursue the following aims: (1)Determine the feasibility of a parental tobacco control intervention combining EHR and provider training features to implement in a group randomized trial of 6 clinics. The primary outcome for feasibility is patient reach (i.e., receipt of the intervention) and the secondary outcome will be abstinence from tobacco use at 6 months; (2) Identify the predictors of patient reach among parents who are tobacco users. Potential predictors include patient sociodemographic factors and other health risk behaviors; and (3) Measure variability in implementation outcomes and identify whether implementation is moderated by practice and provider factors including provider self-efficacy, practice capacity for change and adaptive reserve, and clinic-level patient social characteristics.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Maria Zajac-Kaye, PhD	University of Florida	Testing novel drug combination for pancreatic cancer	According to the American Cancer Society, exposure to tobacco products is one of the most important risk factors for pancreatic cancer. For example, smokers have a 2fold excess risk for pancreatic cancer compared to never smokers. Approximately 25% of pancreatic cancers are thought to be caused by cigarette smoking and there is evidence implicating cigar and pipe smoking as well as the use of smokeless tobacco products. There are no effective systemic treatments for advanced pancreatic cancer which is now projected to be the second leading cause of cancer-related deaths in the US by 2030. Surgery provides the only curative therapy for PDAC but less than 20% of patients are suitable candidates due to challenges to detect cancer when it is surgically removable. While modest improvements in survival have resulted from the use of complex and toxic chemotherapy regimens such as FOLFIRINOX in patients with advanced disease, survival remains largely unchanged. This realization led to the Recalcitrant Cancer Research Act H.R.733 passed by Congress which focused on pancreatic ductal adenocarcinoma (PDAC) and emphasized the broad public interest in testing new treatment approaches. We propose to investigate novel therapeutics that better exploit the molecular basis of pancreatic cancer. The overall goal of this research proposal is to use a newly established animal model to test novel drug compounds in treatment of pancreatic cancer. We established an animal model for pancreatic cancer by generating genetically engineered mice that conditionally express mutant KRAS and human Thymidylate Synthase (TS) in the pancreas. TS, an essential enzyme for DNA synthesis and repair is aberrantly overexpressed in a range of human cancers including PDAC. My laboratory demonstrated that overexpression of TS in the pancreas promoted aggressive PDAC development and markedly reduced survival of *KRAS* mutant mice. TS overexpression is also linked with resistance to gemcitabine and 5FU which are the primary chemotherapy treatments for PDAC. Thus, the goal of this proposal is to develop new treatments for pancreatic cancer using unique TS allosteric inhibitors identified in our laboratory. Our newly developed derivatives do not induce feedback activation of TS levels and are not associated with drug resistance when tested in PDAC tumor cell lines. In addition, our preclinical data show that TS inhibitors synergistically enhance RAS/PI3K/AKT/mTOR inhibition in vitro. In this project we plan to test new TS inhibitors alone or in combination with mTOR inhibitors using this novel in vivo hTS/*Kras* PDAC model. In addition, we propose to determine the antitumor activity of the same drug combination in human samples using patient derived xenografts (PDX) from smoking and nonsmoking associated freshly collected PDAC biopsies. This research will lay the groundwork for NCI R01 application within 3 years and a personalized investigator-initiated clinical trial in 5 years. Our ultimate goal is that the innovative targeting of TS and KRAS pathways will provide a new effective strategy for patients with advanced PDAC.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Vikas Dudeja, MD	University of Miami	Mechanism of Smoking Induced Promotion of Pancreatic Cancer	Despite decades of research, the outcomes of pancreatic cancer are largely unchanged. While the pathogenesis of pancreatic cancer is far from clear, smoking is one of the major risk factors for pancreatic cancer. However, the mechanism by which smoking increases the risk of pancreatic cancer, or for that matter, any cancer is still being unraveled. Recent years have seen an increase in our understanding of the role of gut microbiome in health as well as in the pathogenesis of cancer. Intriguingly, our preliminary data suggest that administration of NNK (commonly used as surrogate for smoking in studies evaluating pathogenesis of smoking induced cancers), one of the key tobacco-specific nitrosamines, in mice leads to increased pancreatic cancer growth and a remarkable change in the gut microbiome. Excitingly, our preliminary data also suggest that depletion of gut microbiome with a broad spectrum, poorly absorbable antibiotics cocktail counteracts the growth promoting effects of NNK. This suggests that pancreatic cancer growth-promoting effect of NNK is mediated by modulation of gut microbiome. Also, NNK administration leads to decreased infiltration of activated cytotoxic T cells suggesting that NNK reduces anticancer immune response. Intriguingly, depletion of gut microbiome prevents NNK induced suppression of anticancer immune response. Based on this we hypothesize that "Smoking inhibits anticancer immune response by modulating gut microbiome". We will evaluate this novel hypothesis in the current grant proposal. In aim 1 of the current grant proposal we will confirm our preliminary finding that smoking promotes pancreatic cancer progression through modulation of gut microbiome. For this, we will use genetically engineered mouse model as well as orthotopic models of pancreatic cancer. The effect of smoking (simulated by use of NNK or smoking chambers) on the tumor growth and progression will be measured with and without gut microbiome depletion (with use of broad spectrum, poorly absorbable, antibiotics cocktail). While NNK is one of the most commonly used compound to recapitulate the effects of smoking, use of smoking chambers more closely recapitulates the effect of smoking as tobacco smoke has more than 7,000 chemicals. Finally, we will use fecal transplantation strategy to further confirm the role of gut microbiome modulation in the effect of tobacco smoking on tumor growth and progression. In aim 2 of the current grant proposal we will characterize the gut microbiome changes induced by smoking in models of pancreatic cancer. We will use various strategies to elucidate which bacterial community mediates tumor-promoting effect of smoking. Finally, in aim 3, we will evaluate the immunological changes induced by smoking and will establish the role of smoking induced gut microbiome modulation in these immune changes. The proposed studies will not only elucidate novel mechanisms by which tobacco smoking promotes tumor growth, these studies will also lead to discovery of novel strategies which can counteract harmful carcinogenic effects of tobacco smoking. For instance, the effect of tobacco smoke induced microbiome changes can be counteracted with novel strategies (antibiotics vs probiotics). Furthermore, these studies will elucidate how cigarette smoking affects immune response against tumor cells as well as the role of smoking on gut microbiome in modulating immune functions. Thus, these studies are novel and significant.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Kunjan Dave, PhD	University of Miami	Nicotine exposure and intracerebral hemorrhage.	Smoking is one of the main risk factors for spontaneous intracerebral hemorrhage (sICH): the deadliest subtype of stroke. However, the effect of smoking on outcomes following sICH is not known. Despite being the cause of significant morbidity and mortality, sICH remains the least treatable stroke subtype. Continued cerebral bleeding leading to hematoma expansion is highest in the first 3 hours after symptom onset and may continue in large number of patients between 3 and 24 hours after the onset. Hematoma volume in sICH patients correlates with the 30day mortality rate. Currently there is no proven therapy to prevent hematoma expansion in sICH patients, and thus clinicians are not able to offer more than supportive care. The prevention of continued bleeding in sICH has been a promising therapeutic target. Dr. Jy (Co-investigator) and his group have studied red blood cell microparticles (RMP) as hemostatic agents for over a decade. Strong preliminary results from Dr. Jy's laboratory has demonstrated that RMP support and enhance hemostasis at sites of bleeding, RMP are effective in correcting hemostatic defects in both platelet and coagulation disorders, and remain effective in the presence of antiplatelet drugs, and RMP are equally effective in treating microvascular and macrovascular bleeding. The goal of this project is to determine the effect of chronic nicotine exposure on outcomes following sICH and if RMP treatment improves post-sICH outcomes (histological, behavioral, and inflammation, among others) in chronic nicotine-treated rats via limiting hematoma volume. We will use preclinical models of autologous blood and collagenase induced sICH. We hypothesize that chronic nicotine exposure will worsen outcomes following sICH and RMP will be able to limit hematoma expansion in a clinically relevant animal model of sICH. We will test this hypothesis in this proposal.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Sulagna Banerjee, PhD	University of Miami	Role of Microenvironment in enrichment of aggressive CD133 population in Pancreatic Cancer	Tobacco smoking is considered to be one of the major risk factors for pancreatic cancer, a disease with very poor survival rates. The poor prognosis in this disease is attributed to the presence of a dense fibroinflammatory stroma consisting of the extracellular matrix, stromal cells and the infiltrated immune population. This creates a complex tumor microenvironment that is conducive to an aggressive disease that is resistant to therapy, extremely metastatic and prone to recurrence. Studies from our group as well as others have shown that increased expression of membrane protein CD133 contributes to aggressive biology in PDAC. These cells are treatment refractory, extremely metastatic and contribute to tumor recurrence. Recent studies show that cancer cells undergo dynamic interconversion between aggressive and nonaggressive states as a result of their interaction with the other cells in the microenvironment. This plasticity between an aggressive and non-aggressive phenotype adds to the challenges for developing a viable therapy against pancreatic cancer that can prevent recurrence and overcome therapeutic resistance. Understanding the molecular mechanism of this dynamic interconversion thus holds the key for developing successful therapy against pancreatic cancer. Our previously published study shows that the CD133+ aggressive cells exhibit an altered metabolic profile from the nonaggressive population, which offers them a distinct survival advantage. It is well known that pancreatic tumors have a robust fibroinflammatory stroma that is extremely reactive. Our preliminary data showed in the presence of the stroma, there is a distinct enrichment of CD133+ cells. Our results show that this enrichment is due to the signaling mediated by the secreted cytokine IL6 from the stromal cells. Secreted IL6 also contributes to an altered metabolic phenotype in the CD133+ cells that is responsible for their survival advantage and aggressive phenotype. Based on these observations we hypothesize that the stromal component of the microenvironment promotes aggressive biology and metabolic reprogramming in a population of tumor cells resulting in a resistant phenotype. This metabolically rewired cells also lead to an immune suppressive microenvironment, thereby resulting in a tumor that is unresponsive to most therapy. We thus propose that targeting the stromal secretion will inhibit this plasticity and overcome therapeutic resistance pancreatic cancer. This hypothesis will be validated by (A) Evaluating the role of stromal component in inducing metabolic reprogramming in pancreatic cancer; (B) Elucidating the mechanism by which metabolic reprograming leads to a survival advantage in CD133+ population in PDAC; and (C) Whether the stromal secretion can be targeted to overcome plasticity and therapeutic resistance in pancreatic cancer.

Fiscal Year 2018-2019 James and Esther King Biomedical Research Grants

Principal Investigator	Principal Investigator's Organization	Project Title	General Audience Abstract
Rex Philpot, PhD	University of South Florida	The effects of chemotherapy for breast cancer on the central nervous system	Smoking is linked to a higher risk of breast cancer in younger, premenopausal women, with some studies indicating as much as a 40% increase in risk. A majority of younger women diagnosed with breast cancer live for several decades following diagnosis and treatment, therefore there is a considerable need for research focusing on the long-term quality of life of breast cancer survivors. Chemotherapy-related cognitive deficits (CRCDs) are a common outcome of cancer treatment, occurring in up to 75% of patients. For some, cognition improves after treatment is complete, but impairment in one or more cognitive domains persists indefinitely for more than 50% of those who experience a cognitive deficit. These deficits can persist for greater than 20 years following treatment, interfering with daily functioning, the ability to return to work and reducing quality of life. Although many cancer patients experience CRCDs and it is clear that these deficits are associated with changes in brain structure and function, the mechanisms underlying the occurrence of these deficits are not understood and there are no treatments approved for this condition. Findings suggest that the manifestation of CRCDs involve reductions in estrogen and/or tumor and chemotherapy-associated increases in proinflammatory cytokines, but it is unclear how these consequences translate into long lasting cognitive deficits. The proposed studies will model CRCDs using cyclophosphamide (CYP) and doxorubicin (DOX), agents used to treat breast cancer, to induce deficits in the working, spatial and/or procedural memory of mice with breast cancer (MMTVPyVT mice). This model will be used to investigate the hypothesis that chemotherapy-induced reductions in circulating estrogen renders cholinergic neurons uniquely vulnerable to injury and death in tumor-bearing individuals that receive chemotherapy, and that selective muscarinic acetylcholine receptor (mAChR) agonists can be used to prevent, or to treat, CRCDs that persist following chemotherapy. Aim 1 will use tumor-free and tumor-bearing mice to test the hypothesis that CYP+DOX administration reduces cholinergic cell number in the central nervous system (CNS) and assess whether reductions in estrogen or increases in proinflammatory cytokines are related to the loss of cholinergic neurons or the manifestation of CRCDs. Aim 2 will test the hypothesis that selective mAChR agonists can treat CYP+DOX-induced cognitive deficits in MMTVPyVT mice and, if these agonists are effective, determine whether these drugs adversely affect tumor growth or the efficacy of the chemotherapeutic agents. Aim 3 will mechanistically test the hypothesis that CYP+DOX-induced reductions in high affinity choline uptake in neurons of the central nervous system, decreased ACh synthesis and/or impaired regulation of proinflammatory cytokines is/are a consequence of chemotherapy-induced reductions in estrogen. Results of these studies will determine whether CRCDs are a consequence of an estrogen-mediated impairment of cholinergic function and whether this impairment is associated with the presence of tumors and/or increases in proinflammatory cytokines. In addition, these studies will determine whether selective mAChR agonists can treat CRCDs and evaluate the impact of mAChR agonists on tumor growth and the effectiveness of chemotherapy. These findings will help identify a mechanism for the occurrence of CRCDs and establish mAChR agonists as a viable treatment.