James and Esther King Biomedical Research Program

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Researcher and Organization	Project Title	General Audience Abstract
Steven Ames, Mayo Clinic	Integrated Intervention for Cigarette Smoking and Binge Drinking in Young Adults	Despite the very high prevalence of smoking in young adults, little effort has been made to address the needs of this group. Substantial evidence links high-risk styles of alcohol use, such as binge drinking, to smoking cessation treatment failure. Related to this, evidence supports the hypothesis that a decrease or cessation of binge drinking should be associated with a decrease or cessation of tobacco use. Thus, an innovative approach with young adult smokers who are binge drinkers is to design an intervention focused on eliminating both of these behaviors. This study will explore the intriguing possibility that an intervention that integrates these two treatment elements might be more effective for smoking cessation than treating cigarette smoking independent of binge drinking. An important additional benefit of this treatment is its potential to reduce binge drinking, which is a high-risk form of alcohol consumption. To our knowledge, we are the only group that is working to explore this innovative line of research.
Adam Wanner, University of Miami	The Airway Microbiome in COPD	The air passages are exposed to a variety of environmental factors including microorganisms. In healthy persons, the host defenses of the lung clear the microorganisms thereby preventing them from causing an inflammatory response that can damage the airways. It is known that cigarette smoke impairs the host defense system allowing colonization of the airways with microorganisms, and this could be one mechanism whereby cigarette smoke causes COPD. In this study, we are testing the hypothesis that the types of microorganisms harbored by the lung are different in smokers and non-smokers, and also different in smokers with and smokers without COPD. We will do this by obtaining secretions from the lung by bronchoscopy (an instrument that allows the operator to view the airways), determining the spectrum of microorganisms in them using a modern gene-based technology, and relating the microbial findings to the presence or absence of clinical COPD in smokers. Should such a relationship emerge in the study, therapeutic agents directed at selected microorganisms could be developed as a novel treatment for COPD.

Jason Lang, Nemours Childrens Clinic	Environmental Genetics (En-Gen): Effect of Environmental Tobacco Smoke and Genetic Variation on Leukotriene Production in Asthmatic Children	Environmental Tobacco Smoke (ETS) imposes an enormous disease burden, particularly among vulnerable groups such as children and those with respiratory conditions such as asthma. Asthmatic children exposed to ETS are at risk for accelerated loss of lung function and worsening disease severity, and may be at increased risk for death, though the precise mechanisms remain unclear. Leukotrienes are molecules made in the body that can cause inflammation. Since active smoking increases leukotriene production, leukotriene-driven inflammation may be a critical component of ETS-induced asthma. This grant is an ancillary study to a multi-center pediatric asthma study conducted by the American Lung Association Asthma Clinical Research Centers (ALA-ACRC). We will determine associations among ETS exposure, leukotriene production, and asthma severity. We expect that ETS exposure will increase both production of leukotrienes and asthma severity. We will also determine associations between leukotriene gene alterations, leukotriene production, and asthma symptoms. Variations in certain genes may alter ETS-induced leukotriene production and resulting asthma severity. This study may better delineate how ETS worsens asthma in children. Common gene variations may place certain individuals at severe risk of ETS-lung injury. Genetic analysis paired with data on ETS exposure and asthma severity may greatly contribute to the goal of improved and personalized asthma care.
Jon Alexander, University of Florida	Investigation of the Role of Corticotropin-Releasing Factor in the Basolateral Amygdala during Nicotine Withdrawal and Stress Induced Reinstatement	The overall idea guiding the proposed studies is that the negative feelings or emotions experienced as a result of quitting smoking and the tendency start smoking again during stressful times are in part, a result of altered functioning of the signaling protein known as corticotropin-releasing factor within the basolateral amygdala, a brain area that plays a critical role in regulating emotional states. Experiments will be conducted in the basolateral amygdala (BLA) to determine if blockade of corticotropin-releasing factor 1 (CRF1) receptors and stimulation or blockade of corticotropin-releasing factor 2 (CRF2) receptors reverses the negative feelings or emotions experienced as a result of quitting smoking and the tendency start smoking again during stressful times. Specific Aim 1 will examine the role of CRF1 and CRF2 receptors within the BLA in the negative feelings or emotions experienced as a result of quitting smoking. Specific Aim 2 will examine the role of CRF1 and CRF2 receptors within the BLA in the tendency start smoking again during stressful times. The

		experimental approach is based on the idea that long-term smoking induces changes in CRF signaling within the BLA and that these changes are associated with the negative feelings or emotions experienced as a result of quitting smoking and the tendency start smoking again during stressful times. Understanding of the role of CRF within the BLA may help in the development of improved treatments for tobacco addiction.
Jessica N. Chang, Bay Pines VA Healthcare System	Antioxidant Transcription Factor Regulation and Alzheimer's Disease	In response to cigarette smoking the human body produces detoxifying antioxidants to combat carcinogens. While these antioxidants are beneficial, sometimes they are not sufficient to prevent disease. Recent studies show that in addition to cancer, smoking may also increase a person's risk for Alzheimer's disease (AD). Alzheimer's is a progressive disease characterized by cognitive and memory loss, affecting almost 500,000 people in Florida and 25 million people world-wide. The reasons for the increased risk are not yet understood. One commonality between smoking and Alzheimer's disease is the same antioxidant, is affected. In moderate smokers this factor is increased as a protective mechanism, but this protection is lost and a decrease in antioxidants is observed in heavy smokers. This same antioxidant reduced in Alzheimer's patients, suggesting that the detoxifying enzymes may be the link between smoking and AD. This grant aims to further investigate which detoxifying enzymes are altered in AD, and whether increasing a specific factor will improve the prognosis of AD in a mouse model.
Medhi Wangpaichitr, Miami VA Healthcare System	Targeting ROS and Tumor Metabolism to Selectively Kill Cisplatin Resistant Lung Cancer	Lung cancer is one of the leading causes of death in the United States and the most common cancer found in South Florida. While early stage lung cancer can be treated by surgical resection, chemotherapy remains the mainstay for treatment for locally advanced and metastatic disease. Cisplatin or its analog carboplatin is one of the main drugs which have been utilized for the treatment of both small cell lung cancer (SCLC) and non small cell lung cancer (NSCLC). Development of platinum resistance is inevitable and hinders the likelihood of achieving remission and hence leads to poor survival. We have found that all cisplatin resistant lung cancer cells express higher baseline levels of oxidative stress. Moreover, these resistant cells appear to change their metabolic pathway in order to adapt to survive under high oxidative stress condition. By identifying and targeting this pathway, cisplatin resistant cells can be selectively killed. The knowledge gained from this proposed work will help to improve treatment outcome and survival in these patients. Furthermore, by being able

		to identify which patient will respond to treatment, this will help reduce costs and hospitalizations of cancer patients. Thus, our proposed work will not only represent a new avenue for overcoming cisplatin resistance but also assist in future selection of patients who will benefit from this treatment.
Yuhui Wen, University of Miami	Mechanisms of Hypoxia-Induced Dendrite Degeneration	Low oxygen level (hypoxia) caused by tobacco smoking is a risk factor for stroke. Once the brain has been traumatized by hypoxia, a flurry of molecular signals encourages healthy neurons to kill themselves. The degeneration (beading and fragmentation) of dendrites, which play an important role in the integration of information flow from one neuron to another, is tightly associated with loss of key brain function. Previous studies have shown that Drosophila melanogaster (a species of fruit fly) exhibits remarkable resistance to lack of oxygen. In preliminary studies, we found that wild-type flies under anoxia (extreme condition of hypoxia, 0.1 percent O2) did not exhibit any beading or fragmentation of dendrites. However, flies heterozygous nmnat (total protein level is reduced), showed severe dendrite beading and fragmentation under the same anoxic conditions. These results suggest that normal levels of NMNAT offer protection for dendrites under anoxia. For the next three years, we will further dissect the underlying mechanism and role of chaperone NMNAT in hypoxia-induced dendrite degeneration. We will start a genetic screen to find new dendrite protective factors. These studies will shed light on hypoxia-induced brain injury and are important to develop effective stroke treatment.
Xiangyang Xie, Sanford-Burnham Medical Research Institute	Functional Analysis of Novel Akt Substrate ASC2D in Glucose Transport System and its Role in Insulin Resistance	Smoking -- long known to increase the risk of cardiovascular disease -- is also associated with an increased risk of developing type 2 diabetes. It has been found that smokers are more than twice as likely to develop the condition of insulin resistance, and type 2 diabetes than non-smokers. Impaired sugar glucose transport into the cells for metabolism is the hallmark of type 2 diabetes. Insulin regulates glucose metabolism primarily through the activation of glucose transport system that is impaired under insulin resistant state in humans. Here, we discovered an unknown protein molecule ASC2D, which is essential for insulin-stimulated glucose transport in the fat cells. In this proposal, we plan to further investigate the molecular mechanisms whereby ASC2D regulates glucose transport and the movement of glucose transporter GLUT4 inside the cells. In addition, we will also investigate how ASC2D is regulated in normal control mice and mice treated with nicotine and high fat

		diet. This project will lead to uncovering a novel signal pathway involved in the regulation of glucose metabolism, and provide new insight for potential therapeutic targets for smoking- and high fat diet-induced diabetes.
Ciceron Yanez, University of Central Florida	Synthesis and Evaluation of Small Molecule Photoactive Bcl-2 and Bcl-XL Inhibitors for Pro-apoptotic Photodynamic Lung Cancer Therapy	In the U.S., 1 in 5 deaths are tobacco related. There are at least 15 kinds of cancers associated with tobacco, including oral cavity, pharynx, larynx, lung, esophagus, pancreas, uterine cervix, and kidney cancer. By far, lung cancer accounts for the highest rate of mortality of tobacco related deaths. Resistance of certain cancers to current treatments, even to emerging treatments such as tyrosine kinase inhibitor (TKI) drugs, has been a recent problem in cancer therapy. This proposal aims to aid in solving drug resistance in tobacco-related cancer treatment. We propose to make and use small-molecules that will react with light once they reach the malignant tissue. The affected tissue should undergo apoptosis, a selective and organized type of cell death, more efficiently in the presence these new drugs because once the affected tissue is exposed to light, the signaling pathways that avoid apoptosis within the cell will be blocked. Traditional photodynamic therapy reagents (like photofrin) absorb light linearly, so they absorb throughout the light path, translating to poor penetration. The molecules that we propose are designed to absorb two-photons very efficiently, restricting absorption of light to the point where it is being focused, enabling better tissue penetration and increased 3D control of the photochemical reaction that blocks the anti-apoptotic routes in the malignant tissue. This will make treatment much more selective, minimizing collateral damage.
Hong Zheng, Moffitt Cancer Center & Research Institute	Regulation of SirT1 Activity by Extra-Cellular pH in Lung Cancer	Tobacco is one of the strongest cancer-causing agents. Tobacco use is associated with a number of different cancers, including lung cancer, chronic lung diseases, and other diseases. According to recent research, tobacco smoke maintains an acidic systemic environment that may facilitate the development of lung cancer. Previous studies have shown that lung cancer cells overexpress SirT1, but its role or mechanism of pathogenesis is unknown. SirT1 is an important regulator of energy metabolism and stress resistance while DBC1 (a gene) has emerged as a novel regulator of SirT1 and a potential signaling mediator in the SirT1 pathway. This study addresses the regulation of SirT1 activity by extra-cellular pH in lung cancer. We plan to characterize the structure of acidosis- (abnormally high acidity) induced DBC1

		fragment and to identify how it is generated, testing the significance of DBC1S production on SirT1 activity and tumor cell response to chemotherapy. As a result, we will be able to determine how SirT1 contributes to tumor progression and treatment resistance, and whether SirT1 inhibitor can be used in the treatment of lung cancer.
Cesar Borlongan, University of South Florida	Blood Brain Repair in Cell Therapy for Stroke	Smoking can cause lung and other cancers, coronary heart disease, chronic respiratory disease, and other diseases, including stroke. This proposal advances the motto "you break it, we repair it". Blood-brain barrier (BBB) breakdown negatively influences central nervous system (CNS) regenerative processes after brain injury. Intravenous administration of a heterogenous cell population containing stem or progenitor cells shows benefit in animal models of stroke. We recently ascribe this functional recovery in transplanted stroke animals to the presence of endothelial progenitor cells in the grafted cell population. Whereas cell-based technologies are largely designed to break the BBB for delivery of therapeutics into the brain, we are taking a novel approach of repairing the BBB damage in stroke. The treatment of ischemic stroke is limited to tissue plasminogen activator (tPA) which only benefits less than 3 percent of stroke patients due to the drug's narrow 3-hour therapeutic window and its detrimental side effects related to BBB damage. That 1) stroke is accompanied by BBB damage, 2) tPA adversely contributes to this BBB damage, and 3) cell therapy can afford BBB repair, form the basis of our overarching hypothesis. Our aim is to show that a treatment regimen directed at BBB repair will restore CNS homeostasis and enhance neuronal regeneration in stroke. Our long-term goal is to advance clinical application cell therapy for stroke.
Jin Cheng, Moffitt Cancer Center & Research Institute	Targeting AKT Pathway in Lung Cancer	Lung cancer is the leading cause of cancer-related death in the U.S.; 85-90 percent of such cases are associated with tobacco use. Current lung cancer non-surgical treatment is based on chemotherapy and radiation, and improvement in survival and quality of life has been observed. However, the disease is eventually refractory to these treatments. Therefore, there is a need to develop new therapies. Hyperactivation of Akt, an enzyme causing tumor development, is detected in more than 50 percent of lung cancer cases and is closely associated with chemo- and radio-resistance as well as EGFR and mTOR (key proteins involved in cancer cell growth) inhibitor resistance. Tobacco activates Akt, which is believed to mediate tobacco-induced lung cancer. We have identified two AKT inhibitors. API-2 is currently in

		clinical trial, and API-1 is a new small molecule inhibitor of Akt. These two inhibitors significantly decrease tumor growth and induce cancer cell death. Therefore, the goal of this project is to determine whether AKT inhibitors can be used as potential therapeutic and chemoprevention agents to inhibit AKT-dependant lung cancer cell growth, and as chemo- and radio-sensitizers to overcome the resistance of chemo-radiotherapy, EGFR inhibitors, and ineffectiveness of mTOR inhibitors. These investigations will provide important information on Akt inhibitor use for combinational clinical trials and chemoprevention of lung cancer.
Valentina Echeverria Moran, Bay Pines VA Healthcare System	Investigating Cotinine to Improve Memory and Prevent Tobacco Abuse in Subjects with Cognitive Impairment due to Psychiatric Disorders	Attention and memory are affected in conditions such as schizophrenia, Alzheimer's disease (AD), posttraumatic stress disorder (PTSD) and depression. In the U.S.A. more than 25 million individuals are affected by these conditions. In these affected individuals, cognitive deficits greatly diminish their ability to work and socialize, and the currently available medications do not target this problem. These mental health conditions are associated with higher tobacco consumption and a deficiency in the nicotinic acetylcholine receptors. It is believed that tobacco use is related to the self-administration of nicotine to reduce symptoms such as anxiety and feelings of depression, and to stimulate the nicotinic receptors to improve memory and attention. Cotinine, the main metabolite of nicotine, stimulates these receptors, improves memory in AD mice, reduces anxiety and presents a good safety profile in humans. At the preclinical level, the present work proposes to investigate the use cotinine on memory impairment induced by PTSD and schizophrenia using three mouse models of stress/PTSD and a drug-induced model of schizophrenia. This research will permit to predict the utility of cotinine in reducing cognitive impairment and anxiety present in individuals with these mental conditions. This is intended to be a first step toward the investigation of cotinine in preventing or reducing smoking behavior in individuals with psychiatric conditions.
Alan Fields, Mayo Clinic	Combined PKCiota and mTOR Inhibition for Treatment of Advanced Non-Small Cell Lung Cancer	Non-small cell lung cancer (NSCLC) is the most common form of lung cancer and the leading cause of cancer death in the United States. Ninety percent of NSCLC cases are linked to tobacco smoking, the major cause of this disease. Despite the best available treatments, the five-year survival rate for NSCLC patients is only 15 percent . The dismal outlook for NSCLC patients has prompted a search for more effective strategies to treat this deadly smoking-related disease. We recently identified a new

		lung cancer-causing gene termed PKC iota. PKC iota is activated in the majority of NSCLC tumors. We have also discovered a small molecule PKC iota inhibitor, aurothiomalate (ATM), which shows potent activity against NSCLC tumors in pre-clinical studies. ATM has synergistic activity when combined with a second drug that inhibits a second cancer-related gene, mTOR. In this project, we will conduct a Phase I/II clinical study to determine the safety and efficacy of ATM when combined with an mTOR inhibitor, temsirolimus, in patients with NSCLC. We will also develop tumor biomarkers of PKC iota and mTOR activity as predictors of response to therapy, and identify biomarkers of PKC iota and mTOR activity in circulating lymphocytes. These studies aim to develop a novel therapy to better treat NSCLC as well as develop tests to monitor and predict how patients respond to this therapy.
Alan Fields, Mayo Clinic	Atypical PKC Signaling in Lung Cancer Stem Cells	Lung cancer is the number one cause of cancer death in the US with a five year survival rate of only 15%. Cigarette smoking, the major cause of lung cancer, is responsible for 90% of lung cancer cases. Our long term goal is to better understand what drives lung tumor formation and progression, and translate this knowledge into better treatment strategies. Human lung tumors contain cells termed lung cancer stem cells that are necessary for lung tumor maintenance and progression. Given their critical role in lung cancer, lung cancer stem cells must be eliminated to effectively treat lung cancer. However, these cells are highly resistant to current cancer drugs. We have identified a gene, PRKCI, that is necessary for lung tumor formation. We hypothesize that PRKCI controls the ability of lung cancer stem cells to form lung tumors and that PRKCI is an attractive therapeutic target. In this proposal we will determine how PRKCI causes lung cancer stem cells to form tumors. Completion of these studies will enhance our understanding of lung tumor formation and progression, and identify novel treatment strategies that target deadly lung cancer stem cells. These studies are a critical step in the development of new, more effective therapeutic approaches for the treatment of smoking-related lung cancer.
Margaret Byrne, University of Miami	Identifying and Addressing Cancer Outcome Disparities in Breast and Lung Cancer	Many minority and poor communities exhibit increased cancer risk and higher rates of cancer death. Higher smoking rates are partly to blame, however other factors are also responsible. Our long-term goal is to identify and eradicate disparities and provide optimal treatment for all cancer patients. The goal of this study is to determine whether and where disparities exist for two of the most

		common and lethal tobacco-associated cancers, lung and breast, then to communicate that knowledge to Florida communities. An interdisciplinary team of cancer surgeons, oncologists, statisticians, epidemiologists and community educators will work together to accomplish these goals. First, a massive and very detailed database will be made by linking the Florida Cancer Data System, which collects diagnosis and treatment data for every Florida cancer patient, to the Agency for Health Care Administration database, which collects extensive inpatient and outpatient data for individual patients. The resulting dataset will cover the treatment, health and outcomes status of ~500,000 cancer patients. Subsequent analysis of this dataset will be powerful enough to detect disparities in diagnosis, treatment and outcomes for patient groups representing even a small fraction of the population. Next, we will identify causes of the disparities. Finally, we will present our findings in culturally appropriate ways to patients, health-care providers and at-risk communities in order to improve cancer care.
Alfred Lewin, University of Florida	Developing Gene Therapy for Age Related Macular Degeneration	Age-Related macular Degeneration (AMD) is a blinding disease affecting as many 1 in 3 people over 70. Smoking is a major risk factor for developing this disease. This is a project to develop a mouse model of early AMD and to test two novel approaches to gene therapy in that model. Our goals are to better understand the cause of AMD and to develop a treatment for early stages of the disease. We have created a genetically modified mouse strain in which a protective enzyme is missing from the one layer the retina, the part of the eye that absorbs light. In our previous experiments, we observed that reducing levels of the enzyme caused serious structural damage similar to that occurring in early (“dry”) AMD. In this project, we will determine the time course of these retinal changes and see if they progress to the “wet” form of AMD, in which leaky blood vessels sprout into the retina causing loss of central vision. We will also attempt to gene transfer methods to arrest the progression of AMD-like changes. First, we will use a non-harmful virus to deliver genes that prevent the generation of reactive oxygen molecules by an enzyme called NOX. In patients, blocking NOX could slow the progression of AMD. Our second approach will be to restrain the release of a molecular signal that stimulates inflammation in the retina. We believe that blocking inflammation will prevent the damage to the retina seen in AMD.
Glenn Micalizio,	A Future for	A collaborative program is described to enable a new

Scripps Research Institute	Natural Product-Inspired Hsp90 Inhibitors in the Search for Clinically Relevant Chemotherapeutic Agents	future for the discovery of therapeutically relevant anticancer compounds. Hsp90 is a validated therapeutic target that has been recognized as the most novel and broadly applicable anticancer target being explored today. That said, no Hsp90 inhibitors have made it to the market. While the search for clinically relevant Hsp90 inhibitors is a focus of numerous pharmaceutical and academic laboratories, the program described here defines an innovative and exceptionally powerful pathway to the discovery of natural product-inspired Hsp90 inhibitors. Addressing the core limitation to exploration, we aim to accomplish discovery in a manner that has never before been possible. The expertise of organic chemists will provide a means to design and prepare natural product- inspired Hsp90 inhibitors that will avoid the well-understood limitations of derivatives in clinical trials through the application of state-of-the-art chemical methods. Thorough biochemical analysis will guide the search for a collection of the most potent and selective Hsp90 inhibitors ever described. The significance of the proposed project is in the ability to open a pathway to chemotherapeutic discovery that is not currently available. As such, the efforts described define an innovative entry to the discovery of new therapeutically relevant anticancer agents.
Maria Jose Miguez, Florida International University	Cytokines an Underlying Cause of Health Disparities in Tobacco Related Diseases	The biological mechanisms whereby tobacco use may influence the development of tobacco related diseases (TRD), particularly among certain racial ethnic groups, are incompletely defined. Nonetheless, our preliminary data indicated that there are some differences in the production of cytokines, and thus raise the question of whether differences on other key cytokines may exist, and if they contribute to the observed differences in TRD. 99. Therefore, one of the goals of this application is to consider the profile of cytokine production among different groups (people living with or without HIV, Black and whites who are or not smokers), so that any disparities in the TRD burden are appropriately attributed. Considering the burden of tobacco related diseases in our society, early detection of those at risk will be of paramount importance. So, to be pro-active, our second goal is to identify the potential usefulness of cytokines in recognizing specific segments of the population bearing a greater risk of developing TRD.
Ana Palacio, University of Miami	Improving Adherence to Cholesterol Lowering	Coronary heart disease is one of the leading causes of morbidity and mortality in the U.S. There is significant evidence that taking a cholesterol lowering medication known as a “statin” for more than 1-2 years significantly

	Medications among Minority Populations in Florida: A Randomized Trial	reduces the risk of having serious cardiovascular outcomes even among subjects that do not have known heart disease. Yet adherence to statin therapy is suboptimal particularly among minority populations. Studies have consistently found that at one year only half of the subjects that started a statin continue to take it. Many other health disparities in cardiovascular care and outcomes have also been recently reported. We propose to compare the effectiveness of a phone based behavior modification program which uses motivational interviewing (MINT) to usual care, using a randomized design. MINT has shown good results at improving health behaviors and smoking quitting rates.We will recruit 1200 subjects from a large health benefits carrier who received a new statin prescription and who are either African American or Hispanic, will randomize them to either group. We will administer a telephonic survey assessing barriers to adherence, self reported adherence and a smoking history questionnaire at baseline and at the end of the study at 24 months to evaluate the impact of the intervention. Also, we will use claims data to objectively determine if the intervention increases by at least 15% the proportion of subjects who refill their statin adequately at 2 years.
Roger Papke, University of Florida	Therapies to Improve Smoking Cessation in Neuropsychiatric and Depressed Patients	Smoking is more common in people with depression and other neuropsychiatric conditions than in the general population. It is believed that for these individuals, smoking is driven by a desire to self-medicate through the stimulation of the alpha7 type of nicotine receptor that is decreased under stressful conditions, and in neuropsychiatric populations. Varenicline (Chantix) a drug recently developed to help people quit smoking is a weak stimulator of the alpha7 beta2* receptors associated with addiction and dependence. It is believed to partially replace and suppress the rewarding effects of nicotine. However, our data show that varenicline also further decreases the function of alpha7 receptors. Reports of suicide and worsened depression in patients taking varenicline has led the FDA to issue a black box warning on the drug. We have shown that GTS-21, an approved drug for human studies, currently in clinical trials for schizophrenia, is a selective partial activator of alpha7 receptors. We have data indicating that GTS-21 should reverse the negative effects of varenicline on the alpha7 receptors of the brain and we propose that if GTS-21 is given as an adjunct therapy to varenicline, it would lessen depression and improve successful smoking cessation in a patient population at high risk for depression or other

		neuropsychiatric disorders. We will test this idea with humans at high risk for mental illness and conduct pre-clinical studies to validate and advance this approach.
Alexander Parker, Mayo Clinic	The Molecular Epidemiology of Renal Cell Carcinoma	Renal cell carcinoma (RCC) is by far the most common form of kidney cancer. Of interest, the number of people diagnosed each year with RCC, as well as those who eventually die from this cancer, has been steadily increasing in the United States (and Florida) for more than three decades. Despite this, the underlying causes of RCC remain poorly understood. In our application, we propose to conduct a large case control study of RCC in order to improve our understanding of the causes of RCC. This project will be the result of a combined effort by investigators at two Florida academic medical institutions (Mayo Clinic and Moffitt Cancer Center) and will involve recruitment of 1,400 individuals with RCC and 1,400 controls with no history of cancer. Using data and tissue samples collected from these individuals, we will build on the current knowledge that smokers, obese individuals and those with a history of urinary tract infections are at increased risk of developing RCC. That is, we will test specific hypotheses regarding exactly how these factors work at the cellular level in the kidney to cause RCC. By doing so, we have the potential to enhance our understanding of how these common exposures increase a person's risk of RCC. As such, our findings could translate into new intervention strategies (i.e. better risk stratification, early detection, and/or chemoprevention) that would ultimately reduce the burden this cancer places on individuals and society at large.
Hamisu Salihu, University of South Florida	Preventing Fetal Body and Brain Size Reduction in Low-Income Smoking Mothers: A Randomized Clinical Trial	Since smoking cessation programs during pregnancy have been only partially successful, especially in low-income subpopulations, it is important to develop interventions that include a strategy to reduce the undesirable impact of smoking during pregnancy. Current low-strength folic acid prescribed to pregnant women is insufficient to compensate for depleted blood folate levels among smokers. This proposal seeks to assess the value of higher-strength folic acid (in comparison to standard of care) combined with smoking cessation program in reducing the negative effects of tobacco smoke on the fetal body and brain. Three follow-up visits are planned during which participating pregnant women will be administered questionnaires and will undergo ultrasound examinations. Maternal blood will also be collected for the testing of folic acid levels and other related substances. All study participants will be followed until delivery at which time umbilical cord blood will be

		collected for assessment of brain growth and development. At birth the infant's body and brain growth limits will also be measured. The two groups will then be compared to determine the effectiveness of higher-strength folic acid supplementation in improving fetal body and brain growth among smokers. This study will provide important information for subsequent follow-up of these infants to determine whether the intervention improves future intellectual, behavioral and physical development.
Charles Rosser, M. D. Anderson Cancer Center	A Multidisciplinary Approach to Improve Patient Outcome in Bladder Cancer - A Tobacco-Related Disease	Bladder cancer (BCa) is among the five most common malignancies worldwide. The major risk factor associated with the development of BCa is tobacco smoke. BCa cases for 2010 are estimated at 60,000, with estimated deaths at 12,700. Despite these figures, BCa remains a cancer that is poorly understood. There are currently several pressing issues regarding (1) how and why BCa develops, (2) what factors in the urine can be used to diagnosis or monitor BCa and (3) what novel therapies could be used alone or in combination to improve patient outcomes. In this application, we have assembled a collaborative team from a variety of medical and research fields in order to harness the incredible resources in Florida for the express purpose of addressing these pressing issues with regard to BCa. Our intention is to conduct five projects: one focused on a new therapeutic agent for BCa, one focused on new targets for drug development, one focused on determining new markers for early BCa detection, one focused on how smoking increases the risk of developing BCa and one focused on the effects of cigarette smoke extract on tumor growth. At the participating institutions, M.D. Anderson Cancer Center Orlando, H. Lee Moffitt Cancer Center, University of Miami School of Medicine, Mayo Clinic Florida and University of Central Florida, we have the resources, study personnel, and patient populations to complete the necessary tasks in a timely and successful manner.
Dominick Angiolillo, University of Florida	Effects of Cigarette Smoking on Clopidogrel Induced Antiplatelet Effects in Patients with Coronary Artery Disease	Cardiovascular disease affects over 80 million people in the U.S. and is the most important cause of mortality. Smoking is a strong risk factor for cardiovascular disease as it has a number of adverse effects, including increasing platelet activation, which in turn increases blood clots. The P2Y12 receptor is a key platelet receptor that influences blood clots as shown by studies in high-risk patients with coronary artery disease (CAD). Thus, there is a clinical benefit associated with antiplatelet agents that block this receptor as it plays a pivotal role in patients with CAD. Importantly, smoking affects the response to

		inhibitors of P2Y12 receptor. This project will use comprehensive and innovative functional assessments to better elucidate how smoking affects P2Y12 response. The project will test the central hypothesis that cigarette smoking enhances chemical alterations of clopidogrel (medication to prevent blood clots) and that the inhibition of platelet P2Y12 effects are greater in smokers compared to non-smokers. The studies are clinically significant since they advance our knowledge of how smoking influences P2Y12, a key therapeutic target for the treatment of CAD patients. These investigations are part of our long-term goal of defining the best antiplatelet treatment strategy in high-risk patients with CAD.
Christopher Armishaw, Torrey Pines Institute for Molecular Studies	Alpha-Conotoxins as Subtype-Specific Nicotinic Acetylcholine Receptor Antagonists for Studying Tobacco Addiction	Tobacco addiction and nicotine dependence is a major health issue that can lead to many associated illnesses. While there are several therapeutic options available to treat tobacco addiction, a more thorough understanding of the mechanisms of nicotine dependence in the brain is required to develop more effective and safer smoking cessation treatments with fewer side effects. Nicotine acts on the nervous system by activating nicotinic acetylcholine receptors, which leads to a release of dopamine that causes the pleasurable effects of smoking. There are many different types of nicotinic acetylcholine receptors, each of which plays a different role in nicotine addiction. As such, blocking certain types of these receptors may help reduce cravings in smokers. However, a major problem for researchers lies in identifying the specific role for each type of nicotinic acetylcholine receptor in the nervous system. Toxins that originate from venomous marine cone snails may hold the key for discovering new research tools that can lead to a better understanding of tobacco addiction. One class of cone snail toxins, the a-conotoxins can distinguish between different types of nicotinic acetylcholine receptors. Our goal is to use a-conotoxins to develop potent compounds that block specific types of nicotinic acetylcholine receptors that are involved in tobacco addiction. An important outcome will be the development of new research tools and drug leads for treating nicotine dependence.
Juan Del Valle, Moffitt Cancer Center & Research Institute	Natural Product-Inspired Approaches Targeting Mcl-1	Despite great strides in our understanding of cancer, the development of targeted therapies remains a slow and arduous process. Many cells that proliferate in an uncontrolled manner avoid death due to an overabundance of protective, or pro-survival, proteins. Once proteins such as these are identified, chemists are

		charged with the task of developing molecules that can modulate or block their function. Unfortunately, many protein interactions are difficult to target with small molecules and chemical efforts represent a significant bottleneck. In order to accelerate this process, researchers have increasingly looked to nature for new leads. Natural products offer a wealth of structural diversity and often exhibit potent anticancer activity. The development of efficient synthetic strategies to access structurally complex analogues is critical to the discovery of new anticancer agents. The goals of this project are to synthesize natural product-inspired compound libraries and to evaluate their ability to modulate Mcl-1, a protein that contributes to uncontrolled cell proliferation and drug resistance. The long-term objective is to discover structurally novel Mcl-1 inhibitors that will aid in our understanding of cancer signaling and may ultimately be used as anticancer therapeutics.
Priya Gopalan, University of Florida	A Phase II Clinical Trial of the CDK 4/6 inhibitor, PD 0332991 in Previously-Treated, Advanced NSCLC Patients with wildtype RB and Inactivated CDKN2a	Lung cancer is the leading cause of cancer death worldwide. Smoking is the predominant risk factor. Despite the use of newer drugs, survival rates are poor. To improve survival while minimizing side effects, treatments will need to be tailored to a patient's genetic profile. In this project, we aim to treat patients with a specific abnormality in their Retinoblastoma (RB) gene pathway. Abnormalities in the RB pathway are found in all patients with non-small cell lung cancer (NSCLC), the most prevalent type of lung cancer. We propose a clinical trial and laboratory experiments to study the efficacy of PD 0332991, a drug that blocks a specific protein in the RB pathway that is abnormally turned on, in combination with decitabine, an FDA-approved drug that targets this pathway by a separate mechanism. By including only patients with a specific tumor genetic profile, we anticipate a good response. Importantly, using genetic markers to define the patients that are likely to respond to therapy will also allow us to identify other genetic markers for potential resistance to therapy, and allow for optimal planning for subsequent studies. Benefits of our study include: 1) the evaluation of a novel drug combination in patients with specific tumor characteristics; and 2) laboratory studies evaluating other drugs to prevent the potential development of resistance. Our project will thus significantly advance the personalized treatment of lung cancer.
Jhanelle Gray, Moffitt Cancer Center & Research Institute	Combination Immunotherapy for Lung Cancer	Despite chemotherapy, patients with metastatic adenocarcinoma of the lung have poor prognosis; therefore, novel modalities such as immunotherapy are

		being developed for this disease. A new lung cancer vaccine, GM.CD40L, has been developed at the Moffitt Cancer Center. This vaccine contains GM-CSF and expresses CD40L. These proteins work together to activate immune cells to migrate to the regional lymph nodes where an amplified immune response can occur, leading to tumor cell killing. In early-stage human clinical trial testing, the GM.CD40L vaccine was found to be a safe method to deliver anti-tumor cell immune responses and was found to diminish disease burden in a variety of solid tumors[1]. CCL21 is a protein that may amplify the T cell responses to this vaccine. Based on this information, a phase II randomized study is proposed to evaluate two vaccine formulations (allogeneic tumor cells plus GM.CD40L bystander cells plus or minus CCL21L) in patients with adenocarcinoma who have failed first-line therapy. The specific aims of the study are to evaluate: (1) clinical efficacy, and (2) the development of specific anti-tumor immune responses. In the future, the information obtained from this project will serve as the basis for a more expansive project where vaccine therapy is compared with standard therapies. The long-term goals are to develop a safe, feasible, and effective therapy that will improve the outcomes of individuals with adenocarcinoma of the lung.
Jingjiao Guan, Florida State University	Array-Based Fiber FISH for Genetic Analysis of Lung Cancer	Lung cancer is the leading cause of all cancer deaths and tobacco smoking is responsible for the prevalence of this disease. Although tremendous efforts have been devoted to the treatment of lung cancer, conventional therapies have been ineffective to increase the survival rate over the past decade. However, an emerging treatment strategy based on the detection and characterization of genetic mutations underlying this disease has shown great potential to significantly improve the situation. Fiber fluorescence in situ hybridization (fiber FISH) is a powerful assay for confirming, identifying, and quantifying cancer-relevant mutations, but the conventional fiber FISH suffers from various drawbacks. I propose here to develop a novel array-based fiber FISH that can allow more reliable identification and more accurate quantification of mutations in lung cancer. Successful development and application of this technique will deepen our understanding of lung cancer genetics and lay a solid foundation for developing more effective therapies against this devastating disease.
Donghwa Kim, Moffitt Cancer Center & Research Institute	Determine Clinic Pathological Significance of	Lung cancer is the leading cause of cancer-related death in the world. The risk of developing lung cancer is directly related to smoking because patients eventually resist

	Alteration of NGB and Regulation by AKT2 in Lung Cancer	chemotherapeutic drugs and radiotherapy. Therefore, there is a need to understand the molecular mechanism of this resistance. Activation of the AKT(one of the oncogenic protein families that plays an important role in cell signaling for tumor development) pathway by tobacco components increases lung epithelial cell proliferation and survival, and inhibits apoptosis (programmed cell death) in response to DNA damage. Similarly to AKT, mTOR regulates cellular processes critical to tumorigenesis such as cell growth, proliferation, and metabolism, and many cancers are characterized by aberrant activation of mTOR, including lung cancer. Recently, we identified a tumor suppressor protein, NGB, which is associated with AKT and mTOR and is frequently altered in various human tumors include lung cancer. Overexpression of NGB significantly reduced lung cancer cell growth, proliferation, and metastasis. Therefore, compelling evidence may be implicating NGB as a bona fide tumor suppressor. However, this hypothesis has not yet been tested in vivo. We plan to address this question using a novel mouse model with NGB loss of function. This study will provide important insights to the research community to guide potential strategies to inhibit cancer progression.
Douglas Kojetin, Scripps Research Institute	Dynamic Regulation of Allosteric Communication Networks in PPARgamma Pharmacology	Our long-term goal is to understand how nuclear receptor (NR) transcription factors (proteins that bind DNA at specific sites to regulate copying) function and contribute to disease development and progression for purposes of preventative and therapeutic control. Tobacco smoke contributes to numerous health issues; in heart disease patients, tobacco smoke significantly affects the expression of a particular NR protein, PPARg. As an obligate heterodimer (protein partner) with the NR protein RXRa, PPARg regulates the expression of genes involved in cellular metabolism, growth, differentiation, and inflammation. Recent emphasis has been placed on a new class of selective PPARg drugs that preserve beneficial properties and decrease side effects of current PPARg drugs. Understanding how existing PPARg drugs work will aid the development of next-generation PPARg drugs. To accomplish this, we will test the hypothesis that pharmacologically distinct PPARg ligands (drugs) modulate (1) the surface used for interacting with other proteins and the ability to preferentially recognize one protein over another of RXRa/PPARg; (2) the function of RXRa/PPARg via unique allosteric (change in shape and activity) changes in dynamics. We predict these studies will provide new insight into how ligands control PPARg

		function and may provide the basis for the development of novel therapeutics for disease treatment.
Sergei Kusmartsev, University of Florida	Tumor-Infiltrated Myeloid Cells and Prostaglandin Catabolism in Human Bladder Cancer	Bladder cancer (BC) is common urologic cancer. Smoking causes about 50% of all BC. Existing therapies require a strong immune response. However, expressions of immunosuppressive factors by cancers lead to the formation of an immunosuppressive environment that protects cancer from immune system surveillance. This proposal takes advantage of the finding that a significant portion of tumor is represented by tumor-infiltrated inflammatory CD11b cells. These cells are recruited by tumor from bone marrow and play major supportive role in bladder cancer progression via local inhibition of immune response. Most evident immunosuppressive factor in BC is prostaglandin E2 (PGE2). PGE2 is synthesized by cycolooxygenase-2 (COX-2), and biologically degraded by 15-PGDH. Our preliminary results show that CD11b cells isolated from tumor tissue produce large amounts of PGE2 but show reduced ability to inactivate it because of low expression of 15-PGDH. The major goal of proposed research is to establish whether correction of PGE2 imbalance in BC microenvironment can reverse immunosuppressive function of tumor-infiltrated CD11b cells and lead to the inhibition of tumor grow. To achieve this goal we will explore the mechanisms underlying the association between imbalance of PGE2 and the immunosuppressive behavior of CD11b cells within the BC. Obtained results may directly lead to development of new therapeutic interventions for treatment of BC.
Xiao Li, University of South Florida	A Rapid and Sensitive Optical Spectroscopic Method for Simultaneous Determination of Cotinine, Trans-3'-hydroxycotinine and Thiocyanate In Vitro	This proposed research intends to develop a rapid, robust and simple method based on optical spectroscopy to determine the concentration of three tobacco-related biomarkers simultaneously: cotinine, trans-3'-hydroxycotinine and thiocyanate. Compared with conventional methods, the technique offers several advantages: little sample preparation, simple, fast, wide concentration range, and direct detection in blood, urine or plasma. Preliminary data showed that the method is sensitive enough for the detection of nicotine and thiocyanate in both non-smokers and smokers, but not for trans-3'-hydroxycotinine in non-smokers yet. Therefore, the aims are: 1. improve its sensitivity through the use of nanoparticles with special structures; 2. optimize it in terms of stability, reproducibility, and selectivity; 3. validate the method by investigating the effect of cotinine, trans-3'-hydroxycotinine and thiocyanate on the survival of neuronal cells in vitro and

		explore their relationship with the decreased incidence of Parkinson's disease in smokers. The long-term goal of the research is to develop a multipurpose and clinically applicable tool which is useful in both molecular diagnosis and scientific research. Information obtained from such methods will shine light on the interplay between tobacco-related chemicals and many diseases including various cancers and lead to possible therapeutic approach for such diseases.
Jaroslava Miksovska, Florida International University	Conformatinal Dynamics in Vertebrate Hexacoordiante Hemoglobins	Two hexacoordinate heme proteins have recently been discovered in humans and other vertebrates. (Hexacoordinate heme means that there are six ligands in this pocket-shaped protein that hold a heme ion or iron atom in place). Neuroglobin (Ngb) has been found predominantly in brain tissue where it plays an important role in the protection of neuronal tissue under conditions of hypoxia (deprived of adequate oxygen) and ischemic (lack of blood supply) stress. Cytoglobin (Cygb) is found in connective tissue of body organs including lung, heart and brain. The physiological role of this protein has not been fully established; however, evidence points to its role in protecting cells against oxidative stress. Cygb was also associated with several types of cancer including sporadic non-small cell lung cancer, and head and neck cancer. Our long-term goal is to determine the physiological function of Ngb and Cygb and to understand the structure-function relationship in the family of hexacoordinate globins. Our goal in this study is to provide detailed information about ligand-induced changes in the structure of vertebrate hexacoordinated heme proteinsand thus provide important insight into the mechanism of ligand interactions with Ngb and Cygb, which will lead to a clearer understanding of the role of these proteins in brain injuries and cancer and ultimately to novel therapeutic targets for the treatment of stroke.
Jose Pinto, Florida State University	Understanding the Molecular Mechanisms of Troponin Mutations in Cardiac Muscle Dysfunction	Long-term tobacco use induces a ventricular hypertrophic response (increase in size) that compensates for damage to the myocardium (heart muscle) and can eventually result in heart failure. Hypertrophic cardiomyopathy (HCM) and Restrictive cardiomyopathy (RCM) are cardiovascular diseases that cause severe cardiac disability and heart failure. These diseases possess a genetic component that is an inherent risk factor for familial heart disease and are greatly affected by tobacco use. An urgent need exists for the development of therapeutic approaches that can tailor the myofilaments' (single functional unit that is responsible for the muscle contraction) contractile response. HCM is a common

		cardiac disorder and main cause of sudden death in the young. RCM is not well understood; however, it results in abnormal diastolic function and impaired ventricular filling. Mutations in Troponin, the protein that binds Ca2+ and regulates cardiac muscle contraction, have been linked to HCM and RCM, and both mutations induce sensitization of the myofilament to calcium. Calcium sensitization may contribute to the development of many overlapping features of both diseases. However, the clinical aspects of each disease are distinct. This project will explore the molecular mechanisms that underlie HCM and RCM mutations and will delineate specific in vitro and in situ observable differences in characteristics that arise from these inherited mutations.
Liyong Wang, University of Miami	Understanding the Mechanisms of Smoking on Complex Diseases from NOS2A-Smoking Interaction	Cigarette smoking (CS) is a strong environmental factor for many complex diseases, including Parkinson's disease (PD), and age-related macular degeneration (AMD). Prevention and treatment of diseases related to CS requires knowledge on the molecular mechanisms underlying the effect of CS, which remains elusive. The genetic revolutions of the past decade have provided substantial insights into the etiology of complex diseases. Statistical evidence for interaction between CS and genes has been reported in many cases. For example, genetic variants in the inducible nitric oxide synthase (NOS2A) gene have been associated with increased disease risk and significantly interacted with CS in PD and AMD. Understanding the mechanisms for these statistical interactions has great potential to make novel findings on CS's biological effects as the genetic approach tackles the question from an unconventional angle. Previous genetic studies on NOS2A, however, are likely to miss the causal variants that are yet to be identified. As the first step to illustrate the CS-NOS2A interaction, this study will map all CS-interacting variants in NOS2A using the cutting-edge genomic technologies and evaluate them in well-powered datasets using PD and AMD as primary disease models. NOS2A have been associated with other CS-related diseases, such as cancer and stroke. Therefore, knowledge gained from this study will be a valuable resource for studies focusing on other CS-related diseases.
Karen Young, University of Miami	Importance of c-kit in Neonatal Lung Development and Disease	Cigarette smoking is the most preventable cause of prematurity, low birth weight and infant death. According to the Center for Disease Control, approximately 10% of mothers in the USA smoke during pregnancy and smoking attributable neonatal expenditures are more than $150 million. One of the leading reasons for this economic

		burden is prematurity and the lung disease which result from this early birth. This lung disease, so called chronic lung disease of prematurity or bronchopulmonary dysplasia (BPD) occurs in 30-50% of preterm infants with birth weights
Mandip Sachdeva, Florida A&M University	Dual Channel Spray Dried Self-Emulsified Oral Formulation for Treatment of Lung Cancer	Lung cancer accounts for 185,000 deaths per year in N. America which is more than any other type of cancer. Tobacco smoking is by far the most important risk factor for lung cancer and contributes to 85% of the total lung cancer cases. Successful treatment of the lung cancer is not possible due to insufficient amount of the therapeutic drug reaching the actual tumor site and associated unacceptable toxicity. Scientists are attempting to improve the lung cancer treatment by novel drugs with lesser toxicity issues. Our laboratory has been working with a novel PPAR-gamma; agonist (1,1-bis (3I-indolyl)-1-p-biphenyl) methane)[DIM-P] which has shown to have potent anticancer activity either alone or in combination with other anticancer agents with no toxicity. However, our initial animal studies demonstrated poor oral absorption of DIM-P, which may limit its development as a single agent. To overcome this limitation, we have developed self-emulsified spray dried formulation of DIM-P (SESD) which has shown to have an enhanced bioavailability by about 30percent in animals and is expected that this will translate to significant higher pharmacodynamic activity. The ultimate objective of this proposal is to prepare and commercialize a capsule formulation of SESD by generating the preclinical data in orthotopic and metastatic lung tumor models. The results from these studies will generate significant interest from various pharmaceutical companies and will lead to its further development
Wei Li, University of Miami	Global Mapping of Autoantigen Biomarkers for AMD	Age-related macular degeneration (AMD) affects more than 1.75 million individuals in the United States and is a leading cause of irreversible blindness in industrialized countries. Smoking increases the risk of AMD. Early diagnosis is important because treatment will delay or reduce the severity of the disease. AMD is currently diagnosed by its clinical symptoms with retinal imaging technologies. The disease course and prognosis are diverse and unpredictable. Protein biomarkers will facilitate early diagnosis and preventive therapy. The critical barrier to early diagnosis of AMD is that there are few biomarkers available with reliability. The long-term goal of this project is to systematically identify all AMD-relevant self-antigens as biomarkers to globally determine their blood autoantibody activity for AMD diagnosis with

		minimal invasion. The objective is to globally identify AMD-relevant self-antigens by a newly developed technology. The hypothesis is that the new technology globally identifies all AMD-relevant self-antigens. The hypothesis will be investigated by globally identifying AMD-relevant self-antigens with the new technology, independently validating identified self-antigens and analyzing their reliability as biomarkers for AMD diagnosis. The outcomes of this project will develop a new technology for global identification of all self-antigens as biomarker for AMD early diagnosis. The new technology is broadly applicable to many other diseases for diagnosis.
Chuanhui Dong, University of Miami	Gene-Smoking Interactions and Atherosclerosis	Despite the decline in mortality rate, cardiovascular disease (CVD) and stroke remain the leading causes of death in the United States, accounting for 34 percent of all deaths in 2006. In most cases, these clinical vascular diseases result from atherosclerosis. Cigarette smoking is one of the most important modifiable risk factors for CVD and stroke and impacts all phases of atherosclerosis. However, the degree of the cigarette smoking-induced damage also varies from individual to individual. This inter-subject variability in response to the effects of smoking may be largely due to the between-individual difference in genetic susceptibility, in addition to the variability in the presence of other environmental risk factors. The overall objective of the proposed study is to identify the genetic factors that modify the effects of smoking on the risks for precursors of atherosclerosis and clinical vascular diseases. Specifically, we will first conduct a genome-wide association scan for new genetic variants that influence the effects of smoking on the precursor conditions of atherosclerosis, then replicate the observed smoking-gene interactions in an independent sample, finally, apply the identified genetic moderators to risk stratification for stroke, myocardial infarct, and vascular death. The information obtained from this project will contribute to our understanding of race-ethnic disparities in cardiovascular disorders and the development of more effective prevention strategies.
Jamie Fernandez, University of South Florida	Neuroendocrine Disruption and Nicotine Preference in a Rat Model of Postpartum Depression	Postpartum depression (PPD) is a devastating disease occurring in 10-15 percent of women. Women who suffer from PPD appear to be sensitive to postpartum hormonal changes. Further, of women who quit smoking during pregnancy, those who develop PPD are at an increased risk of smoking relapse. Animal models exhibiting both PPD and nicotine preference have not been studied and thus, the mechanistic relationship between the two is

		unknown. Studies investigating susceptibility to drug abuse have indicated that animals who respond to a novel open field with high activity (HR) exhibit alterations in response to stress and a greater preference for nicotine compared with animals who respond with lower activity (LR). Thus, parallels between HR rats and rats exhibiting PPD suggest that the former may be used as a 'model' to study maternal behavior, susceptibility to depression, and increased nicotine preference. These studies will use differences between HR and LR rats to investigate PPD and nicotine dependence. It is predicted that postpartum, HR rats will exhibit both depression and a greater tendency to neglect their young as compared to LR rats. It is also anticipated that HR rats exhibiting signs of PPD will have higher nicotine preference, and that estrogen treatment will reduce this preference for nicotine. These studies will provide evidence on the etiology of PPD and a rationale for the use of estrogen replacement to prevent nicotine relapse during the postpartum period.
Monica Hooper, University of Miami	Serotonergic Function and Impulsive Responding in Treatment-Seeking Smokers	Smoking cessation is the most important behavior change a person can make. Smokers often have problems regulating their emotions, which lower their chances of quitting. Low serotonergic function (SF) relates to less control over emotion. Also, childhood maltreatment may create a web of implicit negative associations in memory. No previous research has examined whether low SF combines with early adversity to reduce the odds of quitting smoking. This study will examine relationships between the serotonin transporter gene, early adversity, and smoking cessation. Treatment-seeking adult smokers (N = 80) will receive 8 sessions of cognitive behavioral therapy for cessation, plus nicotine patch therapy. Participants will complete measures of smoking history, early childhood adversity, and genotyping for the 5HTTLPR polymorphism, an SF marker; they will also complete a cognitive task assessing control over emotional material. We expect that (a) adverse early experience will combine with the S genotype (marking low SF) to show impulsive responses to emotion; (b) adverse experience will combine with the S genotype showing deficits in control over emotional material; and (c) poor cognitive control over emotions will reduce smoking cessation, made worse by emotional challenges while quitting. This study will be the first to link genetic s and cognitive patterns to smoking cessation. Findings will be relevant to tailoring interventions based on genotype, and reducing smoking and related diseases.
Jinliang Li,	CIP4 Scaffold	Heart failure is a syndrome of major public heath

University of Miami	Protein Regulation of Cardiac Myocyte Hypertrophy and Survival	significance accountable for nearly 300,000 deaths each year. One of the leading causes of heart failure is coronary heart disease, and tobacco exacerbates coronary heart disease and atherosclerosis. Tobacco smoke contains many harmful chemicals, including high levels of carbon monoxide and nicotine. There is also evidence that chronic exposure to tobacco byproducts results in heart dysfunction and ultimately heart failure. Despite modern therapy, survival is very low after a heart failure diagnosis. A better understanding of the cellular and biochemical mechanisms that control how the heart responds to stress may yield better therapeutic regimens with decreased mortality. My laboratory is focused on how the muscle of the heart grows in response to increased demands for heart pumping. This process is also named by cardiac hypertrophy. We recently found that a new scaffold protein, CIP4, binds to the hypertrophic enzyme Calcineurin and affects myocytes hypertrophy and survival. In this grant, I will explore how CIP4 regulates those signals through specific protein-protein interaction in myocytes. By using a new genetically-modified mouse, I will test whether CIP4 may be a target for therapeutic intervention, preventing the onset of heart failure.
Dmitriy Minond, Torrey Pines Institute for Molecular Studies	Inhibitors of ADAM Proteases for Lung Cancer Therapy and Research	The connection between tobacco use and lung cancer is well established. Non-small cell lung cancer (NSCLC) is the leading cause of death from cancer in both men and women in the United States. A family of enzymes (proteases) called ADAM are known to be involved in cancer cell proliferation. We are proposing to discover selective inhibitors of specific members of the ADAM protease family that can be used in lung cancer research and therapy. Most of the ADAM inhibitors developed to date bind the active site zinc, resulting in off-target toxicity. We hypothesize that non-zinc-binding inhibitors that bind a secondary binding site (exosite) will selectively inhibit specific ADAM proteases and will slow or stop the progression of lung cancer. There are currently no publicly available selective inhibitors of ADAM proteases. The overall aim of this project is to develop substrates that will interact with exosites of ADAM proteases thus facilitating the discovery of selective exosite inhibitors of specific members of the ADAM protease family via screening of potential drug candidates. The successful completion of the aims of this grant will have the following impact on long-term goals of the James and Esther King Biomedical Research Program: (1) it will contribute towards a discovery of treatment for lung

		cancer, and (2) it will expand the knowledge of biomedical and translational researchers working in the field of lung cancer.
Charles Saunders, Florida State University	Improving Surveillance Measures of Tobacco Use in Florida's Adolescent Population	The goal of this study is to determine what effect peer, familial and media influences have on adolescent smoking in order to improve the surveillance and evaluation component of Florida's youth anti-tobacco initiatives. Social influences are among the most important and consistent factors associated with adolescent smoking. However, the manner in which social influences manifest, and more importantly, how they interact in a population of adolescent smokers is not well understood. Therefore, this study will have three specific aims. Aim 1: Obtain detailed information over a three year period on the social influence of media, family and peers from a representative population of Florida adolescents. Aim 2: Determine the effect of social influences and the manner in which they interact on Florida's adolescent smokers and non-smokers using well-defined statistical techniques and measures from the social interaction/social network literature. Aim 3: Using the results from Aim 2 coupled with the current surveillance and evaluation of tobacco control program methodologies developed by the Centers for Disease Control and Prevention and World Health Organization, improve the current surveillance measures of adolescent smoking in Florida, particularly for subgroups based on gender, age and ethnicity.
Gary Wang, University of Florida	Molecular Identification of Subgingival Bacteria Associated with Progression of Periodontitis in Smokers	Periodontal disease (or gum disease) is a common problem facing smokers. It is a chronic bacterial infection that affects the gums and bone supporting the teeth. The infection begins when bacteria in the dental plaque spread and grow under the gum line. As infection progresses, gums become inflamed and detached from the teeth. If left untreated, the infection can destroy the bones, leading to tooth loss. It is well known that smoking causes more rapid progression and more severe disease, and that response rates to therapy are lower in smokers. Although bacteria associated with periodontal disease are known to be more common in smokers, there are no tests that can screen these high-risk patients for periodontal disease or treatment response. Our long-term goal is to be able to screen and predict disease before symptoms develop. We hypothesize that smoking enhances the survival of periodontitis-associated bacteria in periodontal pockets. To test this hypothesis, we will use the latest genomic technology to prospectively compare oral bacteria in current smokers to non-smokers, thereby determining the effect of smoking on these bacteria. We

		will then further investigate this relationship as it relates to periodontal disease and treatment outcome. Once it is known which bacteria predict disease and treatment outcome, screening tests can be developed and clinicians will be able to diagnose and treat on the basis of bacterial compositions before the onset of symptoms.
Dileep Yavagal, University of Miami	Intra-arterial Mesenchymal [Animal] Stem Cell Delivery in a Canine Model of Acute Ischemic Stroke	Millions of smokers are disabled or die as a result of stroke in the U.S. To date, the only FDA approved treatment for 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. Recently, multiple types of stem cells have been shown to be beneficial for the treatment of stroke in experimental studies in mice and rats. Mesenchymal stem cells (MSCs) derived from the adult non-hematopoietic compartment of the bone marrow hold the greatest promise for use in human clinical trials in stroke due to several desirable clinical attributes, including the absence of ethical concerns and the cells' immunoprivileged status. Studies of MSCs delivered via catheter into the carotid artery after stroke have proven beneficial in the rodent models. However, there are no studies of MSCs in large animal models of stroke. This is crucial prior to launching human clinical trials, as biologically the large animal brain is much closer to the human brain. Thus, in the current application our main goal is to test safety and effectiveness of intra-carotid delivery of four escalating doses of MSCs given 36- 48 hours after induction of ischemic stroke in a large animal model.
Robert Gillies, Moffitt Cancer Center & Research Institute	Radiomics of Lung Cancer Screening	This year, 160,000 Americans will die of lung cancer, and worldwide, annual deaths will exceed 10 million by the year 2030. Significant improvements can come from earlier detection. Recently, the National Lung Cancer Screening Trial, NLST, with over 53,000 participants, showed that participants screened annually with computed tomographic (CT) had a significant survival benefit. CT scans detect "suspicious nodules", which have to then be biopsied to determine if they are cancerous. 25% of all patients had biopsies and, of these, over 90% were not cancer, meaning that the biopsy could have been avoided. Furthermore, of the cancers detected by annual screening, 80% were aggressive and advanced when first discovered. This suggests that there is a subpopulation of patients who were at higher risk and perhaps should be screened at more frequent intervals. In

		this study, we will re-analyze the 75,136 CT scans in the NLST database at higher resolution with a specialized computer program developed for this purpose. These data will be used to determine if computer measurement of small, nearly invisible changes ("Radiomics") will be able to identify those patients at higher risk, and thus who need to be scanned more frequently; and those who are lower risk, who may not need biopsies and who may be scanned less frequently. It is important to note that, if this method proves successful, it will improve the diagnostic power of CT scans with no increase in costs.
Antonis Zervos, University of Central Florida	A New Signaling Pathway in Myocardial Ischemic Injury	The leading cause of death in the United States is heart disease and there is a clear correlation between cigarette smoking and the development and progression of cardiovascular disease. The American Heart Association estimates that 52% of heart disease deaths are directly related to cigarette smoking. Numerous studies have shown cigarette smoking can increase the incidence of myocardial infarction (MI), coronary artery disease (CAD) and atherosclerosis. Even passive smoking, environmental or second hand, increases the incidence of cardiovascular disease. Our proposed studies are focused on a recently isolated protein, the Abro1/KIAA0157 that is predominantly expressed in the heart and very little is known about its normal function. However, we have shown that the level of this protein is significantly upregulated in the heart of patients with CAD, as well as in the hearts of mice following experimentally induced MI injury. Our preliminary data suggest that Abro1 is a scaffold protein that recruits other polypeptides to form a K63-linked deubiquitinating (DUB) complex called BRISC (BRCC36-containing isopeptidase complex). Neither the role of Abro1 protein, nor the BRISC complex's function, has been characterized in the normal heart or in heart disease. We expect our studies to unravel a new pathway involved in CAD, as well as MI injury, and provide new targets for preventive and therapeutic interventions in the management and treatment of heart disease.
Masanobu Komatsu, Sanford-Burnham Medical Research Institute	Development of novel lung-targeted biologics for idiopathic pulmonary fibrosis	Idiopathic pulmonary fibrosis (IPF) is a chronic, progressive, irreversible, and lethal lung disease with the median survival of 3 years after diagnosis. IPF occurs in middle-aged and elderly adults (median age of diagnosis 66 years). Today, IPF has a mortality rate that exceeds that of many cancers. IPF affects more than 200,000 Americans, and takes an estimated 40,000 lives each year. The most important and common environmental risk factor of this devastating disease is cigarette smoking. Despite years of research, there is no effective drug

		treatment for IPF. At present, lung transplantation is the only therapy that prolongs survival in advanced IPF. The current primary challenge in IPF is, therefore, to find effective therapeutic approaches that will reverse or stop the progression of the disease. The objective of this research project is to test a novel therapeutic agent, CAR-decorin, in a preclinical model of IPF. CAR-decorin is a unique anti-fibrotic compound we recently developed. It has an ability to 'seek for' the fibrotic lesion in the lung when administered via intravenous injection. Because of its 'target-seeking' activity, it is expected that CAR-decorin accumulates efficiently in the site of fibrosis formation, producing an excellent therapeutic effect. In this project, we will demonstrate the target specificity (Aim 1), efficacy (Aim 2), and mechanism of action of the CAR-decorin therapy (Aim 3).
Dmitry Ivanov, University of Miami	The Role of Danger Signals in Retinal Ischemia	Endogenous 'damage signals' (so-called damage-associated molecular patterns or DAMPs) are generated following tissue injury, such as that induced by ischemia-reperfusion (IR). In turn, DAMPs act through pattern recognition receptors initiating an additional immune response that initiates positive feedback loops increasing tissue damage. Tobacco smoking (TS), a known important risk factor for IR injury, has pro-inflammatory effects and mediates oxidative stress leading to tissue damage. Thus, TS can even amplify DAMPs effect after IR. Our long-term goal is to study if the presence of these DAMPs after IR injury in the central nervous system (CNS) poses an additional risk to cells that have survived the initial ischemic insult. HMGB1 is the prototypic DAMP molecule. Our preliminary data show that treatment with neutralizing anti-HMGB1 antibody significantly improves outcome following IR injury. We demonstrated that HMGB1 receptors mediate damage and inflammation triggered by IR injury. Here we hypothesize that HMGB1-mediated inflammation contributes to IR injury. The hypothesis will be tested in a series of experiments outlined in the specific aims: 1) to determine whether HMGB1 coordinates the immune response of the retina to IR; 2) to determine whether HMGB1 mediates injury of retinal neurons after IR via oxidative stress. The results of this study will stimulate the discovery of innovative strategies to help patients suffering from IR injury.
Hans Peter Larsson, University of Miami	Voltage Sensor Roles in the Physiology and Pathophysiology of a Heart K+	Tobacco smoking is the leading preventable cause of death in United States. Tobacco smoke causes cardiovascular diseases that include heart arrhythmias, stroke, and myocardial infarction. The mechanism of how tobacco smoke causes cardiovascular diseases is

	Channel	beginning to be understood, but many risk factors are still not well understood. Nicotine is the main ingredient in tobacco smoke that cause cardiovascular diseases. Nicotine increases the heart rate, elevates blood pressure, and increases the likelihood of cardiac arrhythmias. People with inherited mutations in proteins called potassium channels have an increased risk of cardiac arrhythmias, which can cause sudden cardiac death. Over 300 different mutations have been found in patients with cardiac arrhythmias. Potassium channels regulate the length of the cardiac contraction and mutations of these channels cause a prolongation of the cardiac contraction, a syndrome that is called Long QT Syndrome. Long QT syndrome is a risk factor for cardiac arrhythmias and cardiac sudden death. The mechanism for how mutations in potassium channels cause cardiac arrhythmia is not well understood. We are investigating the mechanisms by which mutations in potassium channels cause cardiac arrhythmias. We anticipate that an understanding of the mechanisms causing cardiac arrhythmias will lead to better treatments of cardiac arrhythmias and the prevention of cardiac sudden deaths.
Stephen Coombes, University of Florida	Cortical and Subcortical Brain Function in Chronic Stroke	Tobacco use doubles the risk for stroke and leads to strokes occurring a decade earlier than in non-tobacco users. Every year in the US, approximately 795,000 people suffer a new or recurrent stroke. At 6-months post stroke 26% of stroke survivors need help with daily activates such as grooming, dressing, and cooking. These activities are compromised because stroke often disrupts the areas of the brain that control hand function. Understanding brain function during visually guided movements after stoke is important because assessment and rehabilitation approaches involve visually guided movements. The current proposal will first characterize how stroke influences brain activity during a visually guided grip task, and second, will manipulate visual feedback during the task to increase activity in specific regions of the affected motor system. Increased activity in the affected motor system has been associated with better recovery. Our long term goal is to use the findings from this proposal to develop novel treatments that engage affected motor circuits to optimize recovery after stroke. The current proposal is consistent with the goals of the James and Esther King Biomedical Research program because our findings will: 1) improve the health of Floridians with stroke by developing novel approaches to optimize rehabilitation, 2) expand the foundation of biomedical knowledge related to brain function in stroke,

		and 3) attract additional funding from outside the state.
Jia Fang, Moffitt Cancer Center & Research Institute	Functions of MPP8 in Tumor Suppressor Gene Silencing and Lung Cancer Progression	Lung cancer is the leading cause of cancer-related death world-wide. Approximately two thirds of patients are diagnosed at an advanced stage, and of the remaining patients who undergo curative surgery, 30-50% have a recurrence with metastatic disease. The current therapeutic obstacles call for an improved understanding of the molecular mechanisms driving lung cancer progression. It is now evident that activation of epithelial-mesenchymal transition (EMT) can promote tumor cell progression through the basement membrane and invasion into the surrounding microenvironment, such as the lymph and blood vascular systems, contributing to intra or extravasation. EMT is tightly regulated by a diverse array of signal pathways including various epigenetic mechanisms. Recently, we have demonstrated that a novel methy-H3K9 binding protein MPP8 has important functions in promoting lung cancer cell motility and invasion. Functional characterizations suggest that MPP8 coordinates a complex epigenetic network to repress tumor suppressor genes during EMT and tumor progression. We thus propose extended studies to further assess the roles of MPP8 in lung cancer malignant progression and elucidate molecular mechanisms. Because of the reversible properties of the epigenetic modifications, completion of this project will not only significantly impact on our understanding of tumor progression, but also facilitate the future development of epigenetic strategies to improve lung cancer treatment.
Brian Lally, University of Miami	Genomic Prediction Models of Lung Cancer Survival and Treatment Response	Lung cancer is the leading cause of cancer-related deaths in the United States. Tobacco contributes to 90% of lung cancer. The health burden of tobacco will only be exacerbated while underserved minority populations have higher smoking rates. Our and others' data clearly demonstrated that African Americans have the worst survival. The overall survival and quality of life are associated with tumor stage and poorly defined intrinsic factors. We hypothesize that smoking may contribute to racial/ethnic disparities in lung cancer treatment response and mortality because African Americans: (1) are more susceptible to tobacco-related lung cancer risk and (2) have more advanced lung cancer tumors. To achieve the long-term overarching goals in reducing disparities in lung cancer mortality and quality of life, our primary goal is to evaluate racial/ethnic disparities in treatment response and quality of life in three racial/ethnic groups of lung cancer patients, African Americans, Hispanic Whites, and non-Hispanic Whites.

		We will test the hypothesis that worse lung cancer outcomes are associated with smoking and somatic mutations (changes in DNA that cause cancer), that have greater impact in underserved populations. With a large tri-racial/ethnic population, promising preliminary data, high-throughput technology, and skilled multidisciplinary collaboration, we are in an exceptional position to conduct the proposed research.
Lirong Peng, Moffitt Cancer Center & Research Institute	Regulations and functions of Tip60 and hMOF	Cells are exposed to DNA damage during normal development processes. These damages are dangerous to cells if they are not repaired or eliminated. Mammalian cells have an intricate machinery of DNA damage response (DDR) to repair and eliminate damaged DNA. Aberration of DDR will impair the integrity of the genome and lead to tumorigenesis. Understanding the underlying mechanisms of DDR is critical for prevention, diagnosis, and treatment of cancer. Histone acetylase (Tip60 and hMOF) and deacetylase (SIRT1) critically regulate DDR. However, how Tip60/hMOF synergistically acts with SIRT1 in DDR is unclear. Though Tip60/hMOF and SIRT1 are aberrantly expressed in multiple types of cancer, their exact roles in the development of lung cancer remain unclear. I found that SIRT1 dynamically deacetylates Tip60/hMOF and inhibits its enzymatic activity and apoptotic function. Overexpression of Tip60/hMOF or depletion of SIRT1 results in higher drug resistance in lung cancer cells. Therefore, I propose to examine the regulation and functions of Tip60/hMOF in lung cancer with the following aims. 1) Examine roles of SIRT1-mediated deacetylation of MOF/Tip60 in DDR. 2) Dissect the mechanisms and machineries by which SIRT1 dynamically interacts with Tip60/hMOF during DDR. 3) Explore the roles of Tip60/hMOF and SIRT1 in the tumorigenesis of lung cancer. This study will give new insights to the development of new therapeutic treatments for lung cancer.
Lina Shehadeh, University of Miami	Modulation of miR-30e in Nicotine-Enhanced Atherogenic and Osteogenic Pathways	Cigarette smoking is as a major risk factor for atherosclerotic vascular disease, including coronary artery disease and stroke. When risk factors are present, atherosclerosis is initiated early as evidenced by fatty streaks on the vessel wall that in turn promote a cascade of responses in the underlying endothelium and smooth muscle (SM) layers. Vascular smooth muscle cells (VSMCs) are especially implicated in the pathogenesis of atherosclerosis and therefore, identification of pathways that mediate the smoking-related dysfunction of these cells and their responses to cholesterolemia and fatty streaks may unravel new therapeutic targets. Nicotine,

		the addictive compound in cigarette smoke, exacerbates neointimal and atherosclerotic plaque formation. Using a bioinformatics approach, we determined that the host gene NFYC of microRNA-30e is repressed by smoking in human lungs and blood, and validated nicotinic regulation of miR-30e targets in VSMCs in culture. The long term goal is to develop an RNA molecule (miR-30e) into a therapeutic agent for smoking-related vascular disease. The specific aims of the proposal are to 1) determine the action of miR-30e on nicotine-enhanced cholesterol biosynthesis, vascular plaque formation, and SM differentiation, 2) determine the mechanism by which miR-30e regulates SM differentiation, and 3) determine the mechanism by which miR-30e regulates osteogenic differentiation of mesenchymal stem cells with nicotine treatment.
Jose Silva, University of Miami	Involvement of hypothalamic non-protein coding RNAs in the metabolic response to prenatal nicotine exposure in offspring	Although cigarette smoking during pregnancy is associated with adverse fetal, obstetrical, and developmental outcomes, 15–20% of all women smoke throughout the duration of pregnancy. Recent human epidemiological studies have shown a strong relationship between maternal smoking and subsequent obesity and type 2 diabetes in the offspring. Of the 4000 chemicals in cigarette smoke, animal studies suggest that prenatal nicotine exposure (PNE) to nicotine alone may result in postnatal metabolic alterations associated with obesity and type 2 diabetes. These studies further suggest that PNE has long-lasting effects on hypothalamic body weight regulation. Animal studies indicate that PNE alters the functioning of hypothalamic neurons regulating food intake and body weight, in particular that of Pro-opiomelanocortin (POMC) producing neurons. We hypothesize that PNE predisposes to late-onset metabolic disturbances that are associated with gene expression changes in POMC neurons. We will resort to a mouse model mimicking human maternal nicotine abuse from adolescence throughout pregnancy to determine whether PNE alters body weight regulation and gene expression in hypothalamic POMC neurons of offspring. These studies will also focus on expression changes of long non-protein coding ribonucleic acids (long ncRNAs) capable of regulating gene expression. We hope to identify new egulatory mechanisms involved in the metabolic response to prenatal nicotine exposure in offspring.
Hoshang Unwalla, University of Miami	Restoring the Mucociliary clearance enhancing	Cigarette smoking leads to chronic bronchitis (CB) and COPD and is characterized by depressed mucociliary clearance (MCC). Understanding the pathophysiological mechanisms that lead to impaired MCC in chronic

	properties of inhaled beta-2-agonist bronchodilators in chronic bronchitis.	bronchitis is important to public health, especially if we can identify new treatment regimens for a disease with few therapeutic options. COPD is treated with & beta-2-adrenergic receptor agonists available in an aerosolized form for inhalation. Apart from smooth muscle relaxation, beta-2-agonists like albuterol and salmeterol can also improve the MCC by increasing the ciliary beat frequency, activating the cystic fibrosis transmembrane conductance regulator and increasing epithelial permeability. However this is effect is depressed in chronic bronchitis associated with COPD and smokers. Our data indicates that cigarette smoke can interfere with the MCC enhancing property of albuterol/salmeterol by activating TGF-beta1. Coincidently, TGF-beta1 is also implicated in COPD. Thus understanding the mechanism by which cigarette smoke exposure inhibits TGF-beta1 signaling may allow us to develop therapeutics to bypass this inhibition. Simultaneously, this proposal will develop an aptamer-siRNA chimera targeting the TGF-beta signaling pathway to restore the ability of beta-2-agonists to enhance mucociliary clearance in these airway diseases. Anti-TGF-b therapeutics would also find application in lung cancer and reversing airway remodeling in COPD and asthma.
Gaofeng Wang, University of Miami	Determine smoking susceptibility loci in age-related macular degeneration.	Age-related macular degeneration (AMD) is the number one cause of legal blindness in the USA. Tobacco smoke is the strongest and most consistent risk factor for AMD. People who have smoked at least 100 cigarettes have approximately triple the risk of developing AMD compared to individuals who have never smoked. How tobacco smoke causes AMD remains largely unclear. The long-term objective is to uncover the biological role of smoking in AMD and to develop innovative therapeutic strategies to prevent and treat smoking-related damages in the retina. Specifically, this study aims to identify genes being damaged in the retina when exposed to cigarette smoke. To approach this goal, mice are chosen as a convenient model due to the ability to carefully provide a measured exposure to tobacco smoke. A similar project in humans is more difficult due to variable genomic backgrounds and inability to control tobacco exposures experimentally. Next generation DNA sequencing technology will be used to screen for DNA damages (methylation) and changes in gene expression. Initial findings of candidate smoking susceptibility loci will be further tested in a second dataset for verification. The knowledge derived from this study will unveil the effects of smoking on the genome and will be crucial for future studies to develop and test treatment or even prevent

		the onset of AMD in humans.
Sion Williams, University of Miami	High resolution mapping and quantitation of somatic mitochondrial DNA variants in heart failure.	Mitochondria are cellular organelles that play an essential role in energy production. As they are involved in controlling metabolism of tissues like heart and are capable of producing oxidative damage, they are thought to play a role in role in the development and progression of heart failure. Mitochondria carry their own small genome called mitochondrial DNA (mtDNA) and around 20 years ago it was discovered that mtDNA accumulates mutations both as we age and as a result of oxidative damage due to environmental stresses like tobacco use. Because many different mtDNA mutations occur at very low frequency in a tissue, research on mtDNA mutations and has been very difficult. By applying new DNA sequencing technology we have been able to overcome these limitations and have found that a larger and more complex spectrum of mtDNA mutations is present in aging brain than was previously known. We will apply the same technology to normal and failing heart tissue to build a more accurate picture of what happens to mtDNA and mitochondrial gene expression in heart failure. As it is thought that cells accumulate mtDNA mutations randomly, we will also examine what patterns of mtDNA mutations occur in individual cardiomyocytes (heart muscle cells) that have defective mitochondria. This research will help answer long standing questions on the contribution of mtDNA mutations to the development and progression of heart failure.
Naohiro Terada, University of Florida	T3SS-MEDIATED CARDIOMYOCYTE ENGINEERING	Cigarette smoking is a major cause of coronary heart disease, which leads to heart attack. Cellular reprogramming technologies have great impact in regenerative medicine and hold tremendous therapeutic potential to replace damaged heart tissues caused by ischemia. We have recently developed minimally cytotoxic bacterial strains that can efficiently deliver reprogramming peptides into mammalian cells. The current proposal will establish an easy, affordable and safe direct-reprogramming method to engineer transplantable human cardiomyocytes, thus significantly advancing cellular transplantation therapy toward ischemic heart diseases.
Omaida Velazquez, University of Miami	Role of the Notch Signaling in Atherosclerosis & Stem Cell-Mediated	Atherosclerosis and its associated complications, including heart attack and stroke, remain the primary cause of death in humans. Smoking is one of notorious risk factors triggering artery injure that allows plaque to build up along the lining of arterial walls. We recently submitted a RO1 application to the National Heart, Lung

	Arterial Repair	and Blood Institute (NHLBI) and proposed to test and develop a novel stem cell-based therapy via delivery of repair-competent bone marrow cells (RC-BMC) to the atherosclerotic lesions to reverse the pathological process. The overall goal of this proposal is to investigate the involvement of Notch signaling in these processes, and to test whether Notch activity in RC-BMC can be modified to enhance healing of atherosclerotic lesions. Meanwhile, we are developing a clinically-relevant method for atherosclerotic lesion-specific repair cell delivery using nanoparticle targeting of RC-BMC to injured endothelial cells. Outcomes from the proposed work will complement our current efforts to identify novel and critical molecular and cellular targets and to develop new translational therapeutics for the treatment of atherosclerosis. Our work will also pave the way for developing a versatile platform of targeted cell delivery method for genetically specified cell-based therapeutic approach to treat not only atherosclerosis, but also many other diseases.
Jie Wu, Moffitt Cancer Center & Research Institute	Protein Tyrosine Kinase-Phosphatase Interaction in Tumorigenesis	Lung cancer is a major cause of death in America associated with tobacco consumption. The unmet medical need in lung cancer therapy calls for better understandings of how lung cancer cells regulate their growth and survival in order to identify cancer cell-specific vulnerability and the strategies to attack it. The Myc oncogene is often overexpressed in lung cancer. While Myc promotes tumor growth, Myc overexpression also raises vulnerability because these cells become addictive to cellular mechanisms required to support the abnormal demand of the Myc function. In this study, we aim to understand how another lung cancer oncogene called Shp2 protein tyrosine phosphatase protects lung cancer cells from Myc overexpression-induced death. By understanding the mechanism, we can uncover novel targets to attack the Myc oncogene-associated vulnerability in lung cancer cells and use the knowledge to develop more effective treatment for lung cancer.
Danuta Szczesna-Cordary, University of Miami	Functional and Structural Consequences of FHC-linked RLC Mutations	Familial Hypertrophic Cardiomyopathy (FHC/HCM) is a genetic cardiac disorder originating from mutations in major sarcomeric proteins, including the ventricular regulatory light chain (RLC) of myosin. It is manifested by enlarged heart mass and can lead to sudden cardiac death (SCD), especially in people under the age of thirty. Unfortunately, there is no cure for FHC and it is now becoming evident that any effective therapy must target the underlying, mutation-specific, disease mechanisms. Many HCM patients have no symptoms and live an

		uneventful life; however, there are specific RLC mutations, which can result in heart failure and SCD. This grant will explore the mechanisms underlying different FHC phenotypes, from benign to malignant and determine the molecular and environmental risk factors in disease progression. Tobacco use is an obvious candidate for environmental triggers of cardiomyopathy. Continuous cigarette smoking is anticipated to result in a limited blood and oxygen supply adversely affecting the ability of the heart to efficiently pump blood. Nicotine, the main tobacco ingredient is recognized to affect cardiovascular function via sympathetic neural stimulation, but its detrimental consequences and involvement in cardiac disease and/or SCD are not known. Understanding the mechanisms by which RLC mutations lead to HCM will provide insight into tobacco/nicotine-induced development of a cardiomyopathic phenotype and progression to heart failure.
Alexander Agoulnik, Florida International University	Vascular effects of relaxin receptor agonists	Tobacco smoking is the leading preventable cause of premature death in the United States. Smoking is an established risk factor for acute heart failure; however the treatments and clinical outcomes for patients with this disease have not changed substantially over the past few decades. Relaxin is a natural peptide normally produced in reproductive organs; it has vasodilatory and anti-fibrotic effects in heart, lung, and other organs. Recently the relaxin hormone showed favorable results in a clinical trial in patients with acute heart failure. It reduced shortness of breath, improved all-cause and cardiovascular mortality and the progression of heart failure. However, using the recombinant relaxin peptide has significant drawbacks because it is rapidly metabolized, has high production costs, must be administered intravenously, and has a risk of inducing an adverse immunological reaction. Most of these restrictions could be eliminated by the use of bioactive small molecule compounds with relaxin-like properties. The proposed project is based on our discovery of such compounds. The data indicate that the compounds have high biological activity, low toxicity, and superior stability in experiments performed in isolated cells. The current proposal is designed to confirm the biological activity of small molecules in live animals. The use of such compounds as alternatives to the relaxin hormone will have numerous benefits in treating of this deadly disease.
Teng Ma, Florida State University	Development of a Bioreactor	The goal of the project is to develop a spinner flask bioreactor-based strategy for scalable expansion of three-

	Strategy for Scalable Expansion of Human Mesenchymal Stem Cell Aggregates for Heart Diseases	dimensional (3D) human mesenchymal stem cell (hMSC) aggregates. The project is motivated by the need for clinical production of hMSC in a scalable and regulatory-compliant cell production system. Transplantation of adult hMSC has demonstrated significant clinical benefit in treating ischemic heart disease in patents and there are an increasing number of clinical trials that require large quantities of hMSC. However, the cell production technology capable of cell expansion while maintaining their viability and therapeutic remains a major technology barrier in the translation of hMSC therapy to cardiac diseases. Conventional laboratory methods are not scalable for clinical application and associated with culture-induced changes in cell properties, comprising their survival and therapeutic potency. Our laboratory has developed a unique approach to obtained 3D hMSC aggregates with improved viability and function. The proposal is built on these preliminary studies and explores a novel spinner flask bioreactor-based strategy to expand the 3D aggregates in a scalable bioreactor system. The results of the proposed studies will provide the basis for a novel and commercializable spinner flask bioreactor system for large scale production of 3D hMSC aggregates that meet the clinical requirements.
Miroslav Gantar, Florida International University	Enhancing the Activity of Anticancer Drugs by a Natural Product - Phycocyanin	Anticancer drugs are notorious for their harsh side effects. The objective of this project is to improve the efficacy of the currently available anticancer drugs and to allow their usage at much smaller dosage. We have previously reported on anticancer activity of C-phycocyanin, a natural, non-toxic product that was isolated from the novel cyanobacterium Limnothrix sp. 37-2-1. In our in vitro experiments, we found that when only 10% of a typical dose of the anticancer drug was combined with C-phycocyanin, the cancer cells were killed at higher rate than when this drug was used alone at full dose. Based on these findings, we suggest that C-phycocyanin can potentially improve the efficacy of the currently available anticancer drugs and therefore diminish its harsh side effects in the patient. In order to prove our hypothesis, we need additional experimental data using animal studies. For that purpose, mice will be injected with the lung cancer tumor cells and then the mice will be treated with (i) the anticancer drug alone, (ii) C-phycocyanin alone and (iii) combination of the drug and C-phycocyanin. We expect that the most significant regression of tumor will occur with the combined treatment.
Hendrik Luesch,	Development of	Natural products show outstanding potential for the

University of Florida	a scalable synthetic method for lyngbyastatin 7 as a new treatment of pulmonary diseases	development of new drugs. We have discovered marine natural products that have extremely potent activity against a protein called elastase that is overactive in chronic obstructive pulmonary disease (COPD) and other respiratory disorders. Our data suggest that these compounds, especially the compound termed lyngbyastatin 7, have superior properties compared to an approved drug to manage elastase overactivity. We aim to generate large quantities of this potent elastase inhibitor, which is necessary for further drug development. Our goal is to demonstrate the feasibility of large-scale chemical synthesis to generate enough lyngbyastatin 7 for extensive preclinical testing.
Robert Henning, Haley VA Hospital	Umbilical Cord Cells in Hydrogels Mend Smokers Broken Hearts/Umbilical Cord Progenitor Cells in the Treatment of Acute Myocardial Infarction	Each year 1 million Americans experience acute heart attacks (AMI) and 500,000 die from AMIs. Tobacco use is an important cause of AMIs. Smokers are 4 times more likely to develop AMIs and 3 times more likely to die from AMIs than non- smokers. 50% of MI deaths occur in people aged 35 to 69, with an average loss of 22 years of life. New treatments for smokers with AMIs are needed! We are investigating stem cells from human umbilical cord blood to limit AMI size. Since 4 million births occur each year in the US, there is a tremendous resource of umbilical cord blood stem cells (hUCBC) after newborn delivery potentially available for heart repair. Our initial results in rats with AMIs indicate that hUCBC secrete biologically active factors that limit myocardial inflammation and the size of AMIs, contribute to new blood vessels, and preserve normal heart function. However