



THE HARVARD CLINICAL
AND TRANSLATIONAL
SCIENCE CENTER

Biomedical Collaborative Pilot Grants Cycle 1

The Harvard Catalyst Pilot Grants Program sponsored three cycles of Biomedical Collaborative Pilot Grants, which were directed at improving human health. These grants aimed to support clinical or translational research pilot studies that would lead to sustainable, innovative and collaborative projects with a high potential to impact human health.

The Cycle 1 funding opportunity was open to any faculty member holding a Harvard University faculty appointment. A collaborative team was required and inter-disciplinary and inter-institutional collaborations were encouraged. In this cycle, 62 grants were awarded of up to \$50,000 per funded proposal to support a one-year pilot research project.

Funding decisions for the first cycle of Biomedical Collaborative Pilot Grants were announced in January 2009.

For more information about these awards, see the following [news article](#).

Finding of a Bacterial Version of Human Anticoagulant Protein VKOR in *M. tuberculosis* Allows Approaches Both to Antibiotics Against Tuberculosis and Novel Inhibitors of Blood Clotting

Principal Investigator: Jonathan Beckwith, PhD, Harvard Medical School

Co-Investigator(s): Eric Rubin, MD, PhD, Harvard School of Public Health
Bruce Furie, MD, Beth Israel Deaconess Medical Center
Tom Rapoport, PhD, Harvard Medical School
Dana Boyd, PhD, Harvard Medical School
Markus Eser, PhD, Harvard Medical School
Rachel Dutton, Harvard Medical School
Jun-Rong Wei, PhD, Harvard School of Public Health
Weikai Li, PhD, Harvard Medical School
Sol Schulman, Harvard Medical School
Barbara Furie, PhD, Beth Israel Deaconess Medical Center
Mila Bronstein, PhD, Beth Israel Deaconess Medical Center
John Danziger, MD, Beth Israel Deaconess Medical Center

We approach two significant health problems: 1) increased antibiotic resistance of tuberculosis and 2) the need for constant monitoring of patients taking the anticoagulant Warfarin. The problems are connected by our discovery that bacterium *Mycobacterium tuberculosis* expresses a protein which is a homologue of vitamin K epoxide reductase (VKOR), a human protein which is the target of the anticoagulant warfarin. Mycobacterial VKOR helps protein folding as part of the pathway leading to disulfide bond formation in exported proteins. A sensitive assay for inhibitors of VKOR comes from our finding that Mycobacterial VKOR also can assist the bacterium *Escherichia coli* in making disulfide bonds. The anticoagulant warfarin inhibits the Mycobacterial VKOR activity in *E. coli*. We will test a large library of chemicals for inhibition of Mycobacterial VKOR in *E. coli*. Chemicals showing inhibition will be assayed for inhibition of *M. tuberculosis* growth and the human VKOR activity. Human VKOR will also be tested for its ability to promote disulfide bond formation in *E. coli*. A positive result would allow to distinguish inhibitors that act on mycobacterial VKOR and not human VKOR or vice versa. The role of VKOR in *Mycobacteria* will be elaborated and structure of a bacterial VKOR determined. These studies should assist in designing strategies for obtaining VKOR inhibitors for treating tuberculosis or inhibiting coagulation. The Aims of this project are 1) to characterize the role and essentiality of VKOR in *Mycobacteria*; 2) to carry out high through-put screening for small molecule inhibitors of Mtb VKOR; 3) to assess human VKOR activity in *E. coli*, potentially allowing direct screening for new classes of anticoagulants.

Visual Distortion in Amblyopia

Principal Investigator: Peter Bex, PhD, Schepens Eye Research Institute

Co-Investigator(s): David G. Hunter, MD, Children's Hospital Boston

Amblyopia is the leading cause of visual impairment in children and affects approximately 3-5% of the population worldwide. The disorder causes a syndrome of monocular visual deficits, including reduced acuity, a loss of contrast sensitivity and visual distortions that persist after optical correction and in the absence of observable ocular pathology. The primary causes of amblyopia are anisometropia, a refractive imbalance between the eyes, and strabismus, a misalignment of the ocular axes that can cause the eyes to turn in (esotropia) or out (exotropia). Existing treatments for amblyopia temporarily reduce vision in the fellow eye (by patch occlusion or blurring with eye drops) in order to force the child to use their amblyopic eye. Recent results from animal models of amblyopia show that correlated binocular vision is essential for successful recovery from experimentally-induced amblyopia. The uncorrected monocular visual distortions in amblyopia decorrelate binocular vision, suggesting that they may play a critical role in the development of amblyopia and the outcomes of treatment that are currently overlooked. This proposal introduces novel techniques to measure, monitor and correct visual distortions in children with amblyopia. Visual distortions will be examined throughout the course of standard clinical treatment, without modification of therapy, in order to study their prevalence, their role in the prognosis of amblyopia treatment and the recovery of visual function. The technology developed in this proposal will be applied in a computer game-based therapy that will be evaluated as a potential binocular approach for patients who do not respond to existing treatments.

A Pilot Trial of Cytosine Deaminase-Expressing Mesenchymal Stromal Cells in Glioblastoma Therapy

Principal Investigator: Peter Black, MD, PhD, Brigham and Women's Hospital

Co-Investigator(s): Xandra Breakefield, PhD, Massachusetts General Hospital
Rona Carroll, PhD, Brigham and Women's Hospital
Lata Menon, PhD, Brigham and Women's Hospital
Andrew Norden, MD, Brigham and Women's Hospital
A. Gregory Sorenson, MD, Massachusetts General Hospital
Patrick Wen, MD, Brigham and Women's Hospital
James King, MD, Brigham and Women's Hospital

Recurrent glioblastoma is an aggressive and rapidly fatal brain tumor for which no effective treatment exists. Many potential therapies fail due to the infiltrative manner in which GBM cells grow into the normal brain parenchyma. Recent data from our laboratory indicate that bone marrow-derived mesenchymal stromal cells (MSCs) have the ability to migrate toward infiltrating tumor cells in mouse models. Therefore, they represent potential cellular delivery vehicles for therapeutic agents. We have genetically modified MSCs to produce the enzyme cytosine deaminase (CD), which converts the non-toxic pro-drug 5-fluorocytosine (5-FC) to the cytotoxic agent 5-fluorouracil (5-FU). Local administration of MSC-CD cells to human xenograft mouse glioma models results in a significant reduction in glioma growth. We have also labeled genetically modified MSCs with Feridex, a paramagnetic ferumoxide that is imaged using conventional magnetic resonance imaging (MRI) scans. We propose a pilot clinical trial in which patients with recurrent GBM undergo tumor resection followed by injection of autologous, genetically engineered, CD-expressing, Feridex-labeled MSCs into the resection cavity. Post-operatively, time will be allowed for MSC migration to the vicinity of infiltrating tumor cells, and then patients will be treated with 5-FC. This will be converted to 5-FU by the MSCs and should kill nearby tumor cells. 5-FU has been shown to produce a striking bystander effect. Patients will have brain MRI scans at regular intervals post-operatively to observe the migration of the Feridex-labeled MSCs, to assess the impact of 5-FC treatment, and to evaluate tumor response.

Assessment of Biodistribution and Tissue Concentration of Lapatinib in Brain and Brain-metastases of Patients with HER2 Positive Early-stage and Metastatic Breast Cancer by Non-invasive Fluorine-19 Magnetic Resonance Spectroscopy *In Vivo*

Principal Investigator: Nicolas Bolo, PhD, McLean Hospital

Co-Investigator(s): Steven J. Isakoff, MD, PhD, Massachusetts General Hospital

Background: Approximately 40,000 women in the United States will develop HER2 positive breast cancer each year, nearly one third of metastatic patients will develop CNS metastases over the course of their disease, and brain metastases are increasingly the first site of relapse after adjuvant therapy. Lapatinib is a small-molecule FDA approved drug that targets HER2. Limited evidence that lapatinib may cross the bloodbrain barrier suggests that it might therefore have a role in preventing central nervous system (CNS) progression. Although recent studies show promising clinical benefits, to date clinical evidence demonstrating the levels of lapatinib in the CNS of patients with or without brain metastases is lacking. Fluorine-19 magnetic resonance spectroscopy (19F MRS) is a method allowing detection and biodistribution of fluorinated drugs and our preliminary studies predict that lapatinib will be detectable in CNS *in vivo*. Aims: The primary aim of this pilot study is to assess the biodistribution and brain accumulation of lapatinib levels in patients with HER2 positive early-stage and metastatic breast cancer by non-invasive 19F MRS *in vivo*. The secondary aim of the study is to assess the 19F MRS and 19F MRSI signal characteristics of lapatinib in brain tissue *in vivo* in order to optimize the experimental methods and sensitivity of detection for future studies of lapatinib and other fluorinated anti-cancer drugs in heart and liver. Objective: The results of this initial pilot will provide us sufficient preliminary data to seek additional funding support from grant agencies and industry for larger scale and broader scope studies investigating lapatinib biodistribution in brain, liver and heart, cardiotoxicity and elimination kinetics.

The Effect of Air Pollution on Morbidity and Mortality in End-Stage Renal Disease

Principal Investigator: M. Alan Brookhart, PhD, Brigham and Women's Hospital

Co-Investigator(s): Francine Laden, ScD, Harvard School of Public Health
Jaime Hart, ScD, Brigham and Women's Hospital
Daniel H. Solomon, MD, MPH, Brigham and Women's Hospital
Wolfgang Winkelmayr, MD, ScD, Brigham and Women's Hospital

Chronic and acute exposure to ambient particulate matter is associated with increased risk of mortality and other adverse health outcomes in the general population. We hypothesize that patients with end-stage renal disease (ESRD) may be particularly vulnerable to these adverse effects owing to their 25% annual mortality rate and high burden of comorbid conditions such as hypertension, diabetes, COPD, ischemic heart disease, and anemia. If ESRD patients prove to be particularly susceptible to the adverse effects of air pollution, simple and inexpensive interventions could decrease mortality risk and improve quality of life in patients with ESRD. We propose to use Harvard Catalyst funds to bring together a group of clinicians, epidemiologists, and environmental health scientists to conduct an assessment of the risks that air pollution poses to the 500,000 people with ESRD in the US. The project will leverage data resources and research expertise unique to each member of the group. With one year of funding, we propose the following three aims: 1) creation of a longitudinal database containing clinical and health service utilization data on incident ESRD patients from the United States Renal Data System merged with geospatial time-series data on ambient particulate matter; 2) development of preliminary data on the association between various measures of ambient particulate matter exposure and all-cause mortality, incident cardiovascular hospitalizations, and response to treatment with recombinant human erythropoietin and 3) development of an R01 application for NIDDK based on preliminary results.

Computer-Aided Volumetry of Pneumothoraces in MDCT Images for Emergency Care

Principal Investigator: Wenli Cai, PhD, Massachusetts General Hospital

Co-Investigator(s): Marc A. DeMoya, MD, Massachusetts General Hospital
Robert A. Novelline, MD, Massachusetts General Hospital
Xiaozhou Ma, MD, Massachusetts General Hospital
M. Umar Butt, MD, Massachusetts General Hospital
Ashwin Asrahi, MD, Massachusetts General Hospital

Trauma kills approximately 150,000 people each year, in which chest trauma accounts for one quarter of trauma deaths. Pneumothorax, a potentially life-threatening condition, is present in 30-40% of patients suffering chest trauma. Traditionally, it has been diagnosed with chest radiography (CXR) with a false-negative rate approaching 50%. With the rapid development of multi-detector computed tomography (MDCT), such as 64-slice MDCT, CT scanning of trauma patients becomes not only feasible but clinical routine for emergency care. However, the pneumothorax management, the decision of chest tube drainage (CTD), still depends on individual physician's observation and experience. Given the increasing number of diagnosed pneumothoraces using MDCT images and the high complication rate of CTD, clinical investigators search a size/volume-based guideline for better pneumothorax management, which requires the efficient and accurate measurement of pneumothoraces that is currently not routine in emergency rooms because of the absence of available software. The purpose of this project is to develop a computer-aided volumetry (CAV) tool that will provide a time-efficient scheme for detection and measurement of pneumothoraces for trauma patients imaged with MDCT. A size/volume-based guideline of pneumothorax management will be formulated to provide a reliable prediction of the need of CTD treatment, and thus improves the surgeons' performance in treatment of chest trauma patients.

Validation and Development of the Serum Exosome Assay for Human Glial Tumors

Principal Investigator: Bob Carter, MD, PhD, Massachusetts General Hospital

Co-Investigator(s): Fred Hochberg, MD, Massachusetts General Hospital
Aygun Sahin, PhD, Massachusetts General Hospital
Xandra Breakefield, PhD, Massachusetts General Hospital
Johan Skog, PhD, Massachusetts General Hospital

Gliomas are one of the most rapidly lethal human malignancies with an average survival of 15 months after diagnosis. Recent data suggests that genetic subtyping of gliomas may correlate with survival and treatment response. Most genetic testing in glioma is based on direct tissue sampling, which limits the frequency with which sampling can be done, the ability to detect new genetic alterations at recurrence, and the frequency with which sampling can be performed during a patient's course. We recently observed that a novel serum assay can be used to detect tumor specific genetic alterations in brain tumor patients and propose to further develop this into a clinically useful assessment of tumor genetic status. In this proposal, we seek to develop a simple blood test that can be used to diagnose, genetically characterize, and measure therapeutic response in patients with malignant glioma tumors. This proposal is based on the recent observation that human brain tumors shed "exosomes", which are membrane microvesicles that can encapsulate tumor RNA species. Our objective is to translate this observation into a clinically useful assessment of tumor gene expression status via a serum test. Our initial three aims are to 1) optimize the initial specimen handling and processing of serum exosomes to maximize RNA yield and quality; 2) expand our initial observation that serum exosomes can be used to detect EGFRvIII mutations in human brain tumors to a larger cohort of patients with varying histologic grades of tumor and correlate EGFRvIII exosome status to initial tumor grade, volume, MRI characteristics, treatment response, tumor recurrence, and survival; and 3) to explore the utility of serum exosomes for the detection of additional novel genetic or expression alterations in gliomas.

Modeling Hepatitis C Virus Infection in Primary Human Hepatocytes Using Microscale Human Liver Tissue

Principal Investigator: Raymond Chung, MD, Massachusetts General Hospital

Co-Investigator(s): Sangeeta Bhatia, MD, PhD, Massachusetts Institute of Technology
Daniel G. Anderson, PhD, Massachusetts Institute of Technology
Andrew Tai, MD, PhD, Massachusetts General Hospital
Shuhei Yoshida, MD, PhD, Massachusetts General Hospital

HCV is the leading cause of liver cancer. Only half of patients respond to current interferon-based therapy. A major block to the development of effective strategies to fight HCV has been the inability to authentically model infection in human hepatocytes. Current models, in transformed cell lines, are unable to accurately identify the basis of interferon (IFN) nonresponsiveness because of cell culture adaptations in these transformed cells. In this project, we propose to exploit new breakthroughs in culture of primary human hepatocytes (PHH) and gene delivery to take early steps toward defining the basis for nonresponsiveness of HCV to interferon. In Aim 1, we will for the first time develop methods to efficiently transfer PHH from clinical needle, wedge biopsy, or explant liver specimens to a promising micropatterned coculture system that supports robust, differentiated human hepatocytes for up to 6 weeks. In Aim 2, we will optimize the ability (previously demonstrated for a clonal infectious HCV strain) to sustain the HCV infection in chronically infected PHH from HCV+ source patients. Finally, in Aim 3, we will develop methods of efficiently introducing RNAi and DNA constructs corresponding to putative host regulators of HCV into infected PHH and assess their effects on HCV replication. To deliver these genes we will use highly effective lipidoid-siRNA and DNA complexes. In so doing we ultimately hope to manipulate PHHs from nonresponsive patients such as African-Americans and convert them to interferon-responsive cells. This proposal brings together three research groups across two institutions with unique strengths in tissue engineering, gene and RNAi delivery, and hepatitis C virus biology.

Determining the Temporal and Spatial Distribution of Tuberculosis in Lima, Peru: Uncovering Clues for Improving Interventions Against Multidrug Resistant Disease

Principal Investigator: Ted Cohen, MD, DrPH, Brigham and Women's Hospital

Co-Investigator(s): Christopher Paciorek, PhD, Harvard School of Public Health
Hsein-Ho Lin, MD, Harvard School of Public Health
Sonya Shin, MD, Brigham and Women's Hospital
Joaquin Blaya, Harvard Medical School

Background: The emergence of highly drug resistant forms of *M. tuberculosis* threatens the effectiveness of standard tuberculosis control interventions. Despite a declining incidence of tuberculosis in Peru over the past decade, the prevalence of multidrug resistance among new cases continues to rise, especially among those in impoverished and densely populated areas of Lima. New methods are urgently required to improve the speed at which individuals with drug resistant disease are identified and placed on appropriate second-line therapy, both to achieve better outcomes for those directly affected and to prevent secondary spread within communities. **Objective:** Our objective is to determine if geospatial and basic demographic information from new patients can be used to predict the probability of drug resistance among incident tuberculosis cases in two districts of Lima, Peru. **Aims:** Our aims are to i) collect geospatial, demographic, and laboratory information on all notified tuberculosis cases in Lima Este and Lima Norte between January 2005 and December 2007; ii) test the hypothesis that the spatial-temporal distribution of drug resistant tuberculosis is different from that of drug sensitive tuberculosis; iii) identify high risk locations for drug resistant tuberculosis; and iv) test the hypothesis that the use of geographic information can help to predict drug resistance among newly diagnosed tuberculosis patients.

Induced Pluripotent Stem Cell Lines from Patients with Primary Immunodeficiency

Principal Investigator: George Daley, MD, PhD, Children's Hospital Boston

Co-Investigator(s): Luigi Notarangelo, MD, Children's Hospital Boston

Primary immunodeficiencies (PIDs) comprise over 130 different diseases characterized by defects in the host's ability to fight infection. Since the first description of PID more than 50 years ago, the study of these diseases has illuminated a variety of genes and pathways and contributed to a better understanding of the immune system. Human PIDs manifest an extraordinary heterogeneity of mutations with significant phenotypic variability, and there is a poor understanding of the relationship between the specific mutations in target genes and how they disturb lymphoid development and immune function. Recently, novel methods have been developed to produce pluripotent stem cell lines by somatic cell reprogramming, and it is now possible to establish disease-specific stem cell lines from individual patients. In this project, we propose to generate a repository of such induced Pluripotent Stem Cells ("iPS" cells) from PID patients for use in basic research on the pathophysiology of immune deficiency. In future work, we propose to explore the differentiation of iPS cells from PID patients into lymphoid populations *in vitro* to examine how different disease alleles influence the formation of specific lymphoid populations. We will also investigate methods to repair gene defects in the pluripotent cells, and protocols to direct the differentiation of iPS cells into hematopoietic stem cells for transplantation therapies. This humanized cell biological model is needed to advance our understanding of disease pathology and to explore novel strategies for gene repair and cell replacement therapy.

Angiopietin-1 as a New Cardioprotective Drug for Attenuating Ischemia/Reperfusion Injury

Principal Investigator: Susan Dallabrida, PhD, Children's Hospital Boston

Co-Investigator(s): Maria A. Rupnick, MD, PhD, Children's Hospital Boston
Ronglih Liao, PhD, Brigham and Women's Hospital
Renee Dickie, PhD, Harvard School of Public Health
Shou Ching Shih, PhD, Beth Israel Deaconess Medical Center
Giannoula Klement, MD, Dana-Farber Cancer Institute
Robert Gallegos, MD, PhD, Brigham and Women's Hospital
David Zurakowski, PhD, Children's Hospital Boston
Nesreen Ismail, Children's Hospital Boston
Elke Pravda, Children's Hospital Boston
Soeyn Nguy, Brigham and Women's Hospital

This proposal is from a team of 8 investigators, many of whom are junior faculty, from diverse disciplines and 6 different institutions (CHB, BWH, HSPH, BIDMC, DFCI, HMS). We are collaborating on a new initiative with great promise for clinical translation as a novel therapeutic to attenuate ischemia/reperfusion injury. Background: Cardiac ischemia causes the most heart disease-related deaths in the industrialized world. Blocked coronary arteries deprive downstream heart tissue of oxygen and nutrients, resulting in ischemia and injury. If flow is not restored, a myocardial infarction occurs. The standard-of-care is to remove the obstruction as quickly as possible to limit injury and avoid an infarction. However, the oxygen-poor tissue is damaged and fragile. Reperfusion can cause further damage and cell death, resulting in ischemia/reperfusion (IR) injury. There are no established drugs to prevent or treat IR injury. Objective: To address this vital human health issue, we will use a bench-to-bedside strategy. We will conduct pre-clinical studies of a novel target, angiotensin-1 (ang1) in a mouse IR injury model & analyze human heart tissue from patients with ischemia and IR injuries. Our pilot data support the notion that ang1 and an ang1-derived peptide will reduce IR injury. Aims: (1) Define the effect of ang1 and related peptide, QHREDGS on IR injury using a mouse model; (2) Analyze the angiotensin system, reperfusion injury survival kinase signals, reactive oxygen species, and cell death in human heart samples with IR injury; (3) Use genomic and proteomic tools to define key regulators of IR injury in mouse and human hearts.

The Effect of Galvanic Vestibular Stimulation and Non-spatial Attention Training on Functional Outcomes in Neglect Patients

Principal Investigator: Joseph DeGutis, PhD, Veterans Affairs Boston Healthcare System

Co-Investigator(s): Paolo Bonato, PhD, Spaulding Rehabilitation Hospital
David Wilkinson, PhD, University of Kent
William Milberg, PhD, Veterans Affairs Boston Healthcare System
Regina McGlinchey, PhD, Veterans Affairs Boston Healthcare System

Background: The neglect syndrome is a debilitating constellation of spatial and non-spatial attention deficits that is common after a right hemisphere stroke. Highly predictive of a poor stroke outcome, the presence of the neglect syndrome in the acute phase of recovery results in increased dependence, lengthened hospital stay, and decreased effectiveness of physical and occupational therapy. Though many attempts have been made to treat neglect, improvements have either been short-lived or addressed only a subset of patients' symptoms. Additionally, the 'real-world' outcomes of neglect treatments have been poorly characterized. Objective: Our laboratory has pioneered two novel and innovative therapies for neglect: galvanic vestibular stimulation (GVS) and non-spatial attention training (NSAT). These therapies have demonstrated compelling improvements in neglect symptoms on standard laboratory tests, effectively demonstrating "proof of principle." We now need to determine if the observed improvements translate to real-world functional gain. Aims: To do this, we propose to collaborate with individuals at Spaulding Rehabilitation Hospital who can both provide access to a large pool of neglect patients and perform sophisticated assessments of functional improvement. Specifically, we seek to establish an infrastructure at the VA and Spaulding that will support the development of novel neglect therapies and determine whether GVS and NSAT elicit 'real-world' functional gain. The outcomes of this proposal will allow us to further develop these therapies and make them more accessible to patients.

Generation of Functional Human Myocardial Tissue from Embryonic Stem Cells and Induced Pluripotent Stem Cells for the Development of Cellular Models of Human Disease and Drug Discovery and Design

Principal Investigator: Ibrahim Domian, MD, PhD, Massachusetts General Hospital

Co-Investigator(s): Kevin Kit Parker, PhD, Harvard School of Engineering and Applied Sciences
Leslie Caron, PhD, Massachusetts General Hospital
Adam Feinberg, PhD, Harvard School of Engineering and Applied Sciences

Advanced heart failure represents a major unmet clinical need, arising from the loss of viable and/or fully functional cardiac muscle cells. At this point in time, drug-therapy remains the cornerstone of treatment for heart failure. Despite optimum drug therapy, heart failure represents a leading cause of mortality and morbidity in America. A major challenge in drug development is the identification of cellular assays that accurately recapitulate normal and diseased human myocardial physiology *in vitro*. Current approaches for drug discovery are based on animal model systems and consist of either *in vivo* toxicology or *in vitro* cellular assays on rodent (rat and mouse) neonatal cardiac myocytes. Although these approaches have been useful in the past, important differences exist between rodent and human cardiomyocytes as well as between normal and diseased human cardiomyocytes. Generating functional myocardial tissue from a renewable patient-specific source would be a major advance in the field: It would allow for the development of disease specific cellular assays for drug development and discovery and would lay the foundation for cardiac regenerative medicine. Human embryonic stem (ES) cells and, more significantly, human induced pluripotent stem (iPS) cells represent a potentially renewable patient-specific source of ventricular progenitor cells and mature ventricular myocytes. Accordingly, our specific aims are to (1) isolate disease-specific committed cardiac ventricular progenitors from human ES and iPS cells, (2) generate functional myocardial tissue from committed ventricular progenitor cells, and (3) utilize ES and iPS-derived myocardial tissue for *in vitro* drug screening.

Pre-Clinical Development of Histone Deacetylase Inhibitors for the Treatment of Sickle Cell Disease

Principal Investigator: Benjamin L. Ebert, MD, PhD, Brigham and Women's Hospital

Co-Investigator(s): Maureen M. Okam, MD, MPH, Brigham and Women's Hospital
Keith G. Mansfield, DVM, Harvard Medical School
James Bradner, MD, Dana Farber Cancer Institute
Carlo Brugnara, MD, Children's Hospital Boston
John Higgins, MD, Brigham and Women's Hospital
David G. Nathan, MD, Dana Farber Cancer Institute

Sickle cell disease (SCD) is a severe hereditary blood disease that affects approximately 70,000 individuals in the United States and over 200,000 births per year in Africa. Tremendous insights into the molecular pathogenesis of SCD have been slow to translate to effective therapies. Biochemical, epidemiological, and clinical studies have conclusively demonstrated that increased levels of fetal hemoglobin inhibit polymerization of sickle hemoglobin and significantly ameliorate the disease. The only FDA-approved drug in the US for SCD is hydroxyurea, to which only 25% of adult patients derive significant benefit. We and others have accumulated extensive evidence *in vitro* that vorinostat, a histone deacetylase (HDAC) inhibitor, induces the expression of fetal hemoglobin in primary erythroid cells and is therefore an excellent candidate for further pre-clinical development in sickle cell disease. In this proposal, we will perform a dose escalation pharmacodynamic study of vorinostat in a phlebotomized cynomolgus macaque primate model, the model used to establish the efficacy of hydroxyurea. We will also examine whether vorinostat induces fetal hemoglobin in non-phlebotomized animals, greatly facilitating future studies. Leveraging collaboration across Harvard Institutes, we will examine the effects of vorinostat on globin mRNA levels, fetal hemoglobin protein levels, F cell frequency, globin locus acetylation, and red blood rheology. These studies will establish an experimental paradigm and network of collaborators for future pre-clinical studies that will lead directly to clinical trials in sickle cell disease.

Intraoperative Functional Brain Mapping

Principal Investigator: Emad Eskandar, MD, Massachusetts General Hospital
Anna Yaroslavsky, PhD, Massachusetts General Hospital

Co-Investigator(s): Sameer Sheth, MD, PhD, Massachusetts General Hospital

A central tenet in neurosurgery is avoidance of new postoperative neurological deficit. Because prognosis for patients with brain tumors is often directly related to extent of resection, avoidance of damage to critical structures must be balanced against the desire for complete resection. This goal is especially challenging when operating in or near “eloquent cortex”, or regions subserving known specific functions, such as sensation, motor control, or language. Given individual neuroanatomical variations, eloquent regions must be delineated at the time of surgery, within the individual patient. We propose creating high-resolution intraoperative functional maps of the cortical surface in patients undergoing open brain surgery in or near eloquent cortex. To do so, we will employ optical imaging of intrinsic signals (OIS), a technique used successfully for high-resolution functional brain mapping in animals but rarely, and with limited clinical success, in humans. Polarized light imaging (PLI) techniques developed in the field of dermatology for identifying skin cancers will be applied to improve the accuracy of optical imaging. We will determine how well avoidance of regions we identify as “active” predicts prevention of postoperative deficit. We hope that by improving our ability to safely navigate these treacherous regions, we can make a significant positive impact in patients with otherwise potentially untreatable neurological disorders.

Understanding the Mechanisms of Venous Arterialization to Improve Care of Hemodialysis Patients

Principal Investigator: David Friedman, MD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Seth Karp, MD, Beth Israel Deaconess Medical Center

More than 350,000 Americans with kidney failure depend on hemodialysis for survival. An arteriovenous fistula (AVF), surgically created using a patient's artery and vein, allows for high blood flow to a dialysis machine for filtration. Normally, veins exposed to arterial flow develop arterial properties, a process called venous arterialization. However, in up to 50% of dialysis patients, particularly women, African Americans, and those with diabetes, the venous limb of the AVF fails to arterialize. Using mouse models and human tissue studies in parallel, we will investigate the molecular mechanisms underlying this process. Understanding these mechanisms in mice and humans will suggest pharmacologic experiments in our system that may quickly lead to human studies.

Development of Novel Antibiotics that Inhibit a Large Family of Bacterial Virulence Proteins

Principal Investigator: Marcia B. Goldberg, MD, Massachusetts General Hospital

Co-Investigator(s): Gregory D. Cuny, PhD, Brigham and Women's Hospital
Daniel J. Slade, PhD, Massachusetts General Hospital

Abstract withheld at the request of the investigator.

HarvardShare

Principal Investigator: Stephen Goldsmith, JD, The John F. Kennedy School of Government at Harvard University

Co-Investigator(s): Zachary Tumin, MA, Harvard Kennedy School of Government
Jason Pryde, MA, Harvard Kennedy School of Government
Erica George, Harvard Law School

HarvardShare is a Harvard-branded, open-source translational research network that will be a venue for interactive collaboration among Harvard faculty, policy makers and practitioners. Focusing on the test case of addressing the health impacts of adverse weather from climate change, this network will accelerate the pace of discovery and innovation to this fast-moving policy challenge by leveraging social media applications available both inside and outside of the Harvard community.

Metabolite Profiling and Neurotransmitter Analysis for Biomarker Discovery in Children with Colonic Dysmotility

Principal Investigator: Allan Goldstein, MD, Massachusetts General Hospital/The Broad Institute

Co-Investigator(s): Leonel A. Rodriguez, MD, Children's Hospital Boston
Robert Gerszten, MD, Massachusetts General Hospital/The Broad Institute
Samuel Nurko, MD, Children's Hospital Boston
Ana Souza, MD, Children's Hospital Boston
Kristin Hart, Children's Hospital Boston
Eugene Rhee, MD, Massachusetts General Hospital/The Broad Institute

Abnormal colonic motility is a major cause of severe, chronic constipation refractory to medical therapy, occurring in children and adults with slow transit constipation, irritable bowel syndrome, and other disorders of colorectal function. Despite its severity and prevalence, however, its causes are largely unknown and the available drug therapy is limited. We propose an innovative, multi-institutional, and cross-disciplinary study aimed at identifying novel serum biomarkers associated with colonic contractility in children in order to enhance our understanding of the neurochemical basis of colonic motility and the pathophysiology of dysmotility and intractable constipation. In Aim 1, metabolite profiling, a technology that allows quantification of hundreds of serum metabolites in a single experiment, and quantitative assessment of serum neuropeptides will be used to characterize the neurohumoral and metabolic responses to stimulation of colonic contractions during colon manometry studies in control subjects and in children with intractable constipation. Aim 2 will examine neurotransmitter expression by immunohistochemistry in colorectal tissue obtained from these subjects. In Aim 3, validated quality of life and intestinal function questionnaires will be used to assess clinical outcome and response to treatment, and to correlate these with results obtained in Aims 1 and 2. This study will enhance our understanding of the mechanisms underlying normal colonic contractility and the pathophysiology of dysmotility, identify patterns for disease categorization, reveal predictors of response to therapy, and enable discovery of new areas for basic investigation and new targets for pharmacotherapy.

Non-invasive Real-time Metabolic Imaging Using Hyperpolarized Pyruvate to Assess the Warburg Effect in Tumors

Principal Investigator: Aaron Grant, PhD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Pankaj Seth, PhD, Beth Israel Deaconess Medical Center
Vikas Sukhatme, MD, PhD, Beth Israel Deaconess Medical Center
Elena Vinogradov, PhD, Beth Israel Deaconess Medical Center
David Alsop, PhD, Beth Israel Deaconess Medical Center
Robert Lenkinski, PhD, Beth Israel Deaconess Medical Center
Han Xie, BA, Beth Israel Deaconess Medical Center

There is considerable excitement in exploring new metabolic targets for cancer therapy. The Warburg effect - enhancement of fermentative glycolysis (pyruvate to lactate conversion) - may provide cancer cells with a survival advantage. A major question is whether cancer cells are selectively killed if this effect is reversed. We and others have shown that an RNAi mediated knockdown of LDH-A (lactate dehydrogenase-A) results in slower tumor growth. Objective: Here we propose to utilize hyperpolarized magnetic resonance (MR) contrast agents to evaluate non-invasively whether the flux of pyruvate to lactate is altered in a xenograft tumor model by LDH-A knockdown and by the drug dichloro-acetate (DCA), which enhances pyruvate's entry into the TCA cycle, and to assess if this "pharmacodynamic" measurement correlates with tumor growth. Aims: Aim 1: Does LDH-A blockade result in decreased lactate formation *in vivo* in a xenograft model of non small lung cancer as assessed by hyperpolarized pyruvate? Aim 2: Does administration of DCA result in decreased lactate formation *in vivo* as assessed by hyperpolarized pyruvate? These studies will set the stage for rapid evaluation of clinical trials using metabolic inhibitors such as DCA. In summary, this project is a collaboration among radiologists, basic scientists and cancer researchers which involves a novel technology - that is scalable to humans - to explore a promising and relatively untapped area of cancer research (tumor metabolism).

Metabolite Profiling of Urine and Blood in Patients with Proteinuric Kidney Disease

Principal Investigator: Anna Greka, MD, PhD, Massachusetts General Hospital

Co-Investigator(s): Martin Pollak, MD, Brigham and Women's Hospital
Robert Gerszten, MD, Massachusetts General Hospital/The Broad Institute
Eugene Rhee, MD, Massachusetts General Hospital/The Broad Institute

Diabetes is a growing global public health problem, due to the ongoing obesity epidemic. A significant portion of diabetic patients go on to develop kidney disease which is manifest as progressively severe proteinuria – the inappropriate spilling of protein into the urine – often leading to kidney failure. While the mechanisms of acquired proteinuric kidney disease remain elusive, the most important insights have been derived from analyses of hereditary forms of the disease, such as Familial Focal Segmental Glomerulosclerosis (FSGS). Interestingly, recently developed, specialized biochemical and computational techniques have enabled the study of the human “metabolome,” the collection of chemicals produced and processed by the body in health and disease states. This is accomplished by taking advantage of tandem mass spectrometry technology to reliably identify changes in metabolites in human biological samples. This technique has been successfully used to detect changes in metabolic profiles in patients undergoing exercise stress testing or experiencing a myocardial infarction. Little is known about the metabolome in the setting of diabetes and kidney disease, and, in fact, the content of the urine metabolome remains largely uncharted. We propose to harness the tools recently established in the field of metabolite profiling (or “metabolomics”), to study blood and urine derived from patients with diabetic kidney disease and FSGS, as compared to healthy controls. This pilot study will allow us to chart the urine metabolome, and to discover novel risk markers for diabetic and proteinuric kidney disease.

A Multiplexed Hybrid Microarray/MALDI Platform for High Throughput, High Sensitivity and High Resolution Clinical Protein Biomarker Discovery and Quantitative Validation by Mass Spectrometry

Principal Investigator: Xuesong Gu, PhD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Towia Lieberman, PhD, Beth Israel Deaconess Medical Center
Simon Dillon, PhD, Beth Israel Deaconess Medical Center
Martin Sanda, MD, Beth Israel Deaconess Medical Center
Krystyn J. Van Vliet, PhD, Massachusetts Institute of Technology
George Whitesides, PhD, Faculty of Arts and Sciences, Harvard University
Sunyoung Lee, PhD, Massachusetts Institute of Technology

Background: Rapid unambiguous identification and quantitation of diagnostic and prognostic protein biomarkers from bodily fluids would provide critical information to the clinician as they evaluate and monitor their patients for categorization and staging of the disease state. Mass spectrometry (MS) enables rapid and precise identification and quantitation of molecules with biological relevance. However, the power of this technology has yet to generate significant impact in the clinic. MS reports the exact molecular weight of a molecule as well as identifying structure or sequence. This sensitive technique has the potential to definitively characterize a wide-range of molecules relevant to the monitoring of patient health. Objective: MS has been developed as a sensitive and specific tool for the research lab and we predict that with well-designed, purposespecific engineering this technology is ready to positively impact the clinical lab. We propose to develop a novel hybrid mass spectrometry-based microarray platform that combines mass spectrometry with protein microarray and ELISA that will be able to identify and validate new biomarkers for disease as well as routinely assay for them in patient samples in a high throughput, high sensitivity and high specificity fashion. Specific Aims: We will develop and test a laser desorption/ionization (LDI) surface with chemistries for attaching proteins and oligonucleotide barcodes. We will validate the sensitivity/specificity of the surface with attached antibodies to recognize their binding peptide by MALDI tandem MS. We will validate the performance of our technology to test real biological samples (prostate cancer) for known disease biomarkers (PSA).

The Impact of Immigration and Customs Enforcement Efforts on Immigrant Health

Principal Investigator: Karen Hacker, MD, MPH, Cambridge Health Alliance

Co-Investigator(s): Laurel Leslie, MD, Tufts University
Robert Marra, Cambridge Health Alliance
Alex Pirie, Immigrant Service Providers Group/Health
Antonio Amaya, La Comunidad Inc.
Dolores Acevedo Garcia, PhD, Harvard School of Public Health
Robert Marlin, MD, PhD, Chambridge Health Alliance
Jacqueline Bhabha, JD, Harvard Kennedy School of Government
University/Harvard Law School
Jocelyn Chu, MPH, ScD, Cambridge Health Alliance
Kathy Bungay, PharmD, Tufts Medical Center

Increased immigration enforcement efforts across the country are creating a climate of fear and discrimination for both documented and undocumented immigrants. Local communities struggling to meet the demands of these new populations are creating their own immigration policies which limit access to entitlements, education, and health care for immigrant communities. Preliminary data suggests that fear of deportation is not only affecting immigrant mental health but is changing the way that immigrants access health care. While undocumented immigrants are particularly affected, documented immigrants are also experiencing problems. To further understand the impact of more intensive enforcement on immigrant health, the community of Everett has engaged Harvard researchers. Using community-based participatory research methodologies, we plan to collaboratively examine these issues, particularly focusing on undocumented immigrants, and subsequently identify potential interventions at either the clinical or policy level that may support immigrant health and well-being. We will examine the problem in-depth in one specific community, Everett, MA, from several perspectives: ambulatory and emergency department providers, immigrants and local authorities. Specifically, we aim to: 1) Explore local implementation of immigration policies and its impact on the health and health seeking behaviors among undocumented immigrants residing in Everett via multiple perspectives; 2) Identify potential interventions to support immigrant health needs, and 3) Examine the potential applicability of these interventions to other communities including Cambridge, Somerville, and Boston.

Social Networks and Socialized Risk: Understanding and Mitigating Disparities in Renal Transplantation

Principal Investigator: Douglas W. Hanto, MD, PhD, Beth Israel Deaconess Medical Center/Harvard Medical School

Co-Investigator(s): Keren Ladin, MSc, Beth Israel Deaconess Medical Center/Faculty of Arts and Sciences, Harvard University
James R. Rodrigue, PhD, Beth Israel Deaconess Medical Center/Harvard Medical School
Didier A. Mandelbrot, MD, Beth Israel Deaconess Medical Center
Martha Pavlakis, MD, Beth Israel Deaconess Medical Center
Nicholas Christakis, MD, PhD, MPH, Faculty of Arts and Sciences, Harvard University / Harvard Medical School

Project Summary: Although blacks constitute 37% of the U.S. population receiving dialysis and 35% of those on the transplant waiting list, they only receive 25% of deceased-donor kidneys and just 15% of live-donor transplants (1). Live donor kidney transplantation (LDKT) is an option for patients with ESRD, where a living donor donates an organ for transplantation to a patient. LDKT is associated with superior graft and patient survival rates and is more cost-effective than longterm dialysis or deceased donor kidney transplantation. However, among those patients who received LDKT in 2007, 66% were white and only 15% were black (2). By using social network analysis we seek to elicit the underlying causes revealing why blacks exhibit dramatically lower rates of transplantation, especially of LDKT. Objective: To explore the role of social networks in facilitating racial disparities in LDKT. Aims: Odds of finding eligible donors depend on the health, financial status, willingness to be tested (social norms), and number of persons in social network (representing pool of potential donors). We postulate that social networks of black recipients might limit life chances in two ways: (1) limiting the number of eligible donors through due social capital and poor health, (2) limiting the number of potential donors who get tested due to differences in social norms, risk perception, or risk aversion.

The Neurobiology of Appetite Regulation in Anorexia Nervosa

Principal Investigator: Elizabeth Austen Lawson, MD, Massachusetts General Hospital
Laura Holsen, PhD, Brigham and Women's Hospital

Co-Investigator(s): Anne Klibanski, MD, Massachusetts General Hospital
Jill Goldstein, PhD, Brigham and Women's Hospital
Anne Becker, MD, Massachusetts General Hospital
David Herzog, MD, Massachusetts General Hospital
Emily Stern, MD, Brigham and Women's Hospital

Disordered eating is an important problem causing illnesses ranging from self imposed starvation to obesity. Anorexia nervosa (AN) is a common eating disorder characterized by severe weight loss, many metabolic and endocrine derangements and a significant increase in mortality. Hormones that govern appetite and food intake are markedly abnormal in AN and include those secreted by the brain as well as in the gastrointestinal tract. However, the neuroendocrine and neuroanatomic pathways governing eating behavior are largely unknown. Brain imaging studies have shown that brain circuitry in individuals with eating disorders are markedly abnormal. Specific brain regions involved in eating behavior and food motivation link to areas of the brain that regulate hormone production. We propose to investigate whether appetite-regulating and stress hormones implicated in fasting, eating and visualization of food are abnormal in AN compared to healthy controls. We will use functional magnetic resonance imaging (fMRI) to investigate whether deficits in brain activity in regions implicated in appetite regulation and food motivation are abnormal and associated with alterations in appetite and stress hormones. These studies will link hormones, brain circuitry and eating motivation to enable an understanding of diseases associated with disordered appetite and food intake. Specific Aims: To examine whether: 1. Response of appetite-regulating and stress hormones to fasting, consumption of a mixed meal challenge, and visualization of food is abnormal in women with AN and those who have recovered from AN compared to healthy controls. 2. Deficits in brain activity in regions implicated in appetite regulation and food motivation (limbicfrontal circuitry) are abnormal in women with AN and those who have recovered from AN compared with healthy controls using functional magnetic resonance imaging (fMRI).

Effects of T2D Gene Variants on Induced Pluripotent Stem Cell Derived Beta Cell Function

Principal Investigator: Paul Huang, MD, PhD, Massachusetts General Hospital

Co-Investigator(s): Chad A. Cowan, PhD, Massachusetts General Hospital
Douglas Melton, PhD, Faculty of Arts and Sciences, Harvard University
Gordon Weir, MD, Joslin Diabetes Center

The goal of this proposal is to define the effects of newly identified type 2 diabetes (T2D) genes in the function of human pancreatic insulin-producing beta cells. This translational medicine study leverages an ongoing 3000 patient study in the Cardiology Division at MGH that involves collection of blood for genotyping, phenotyping for coronary disease and T2D, and provisions to call patients back. We propose here to call back patients who carry specific genetic variants that are associated with increased risk for T2D, and to obtain skin biopsies, from which we can derive induced pluripotent stem (iPS) cells. These iPS cells will be differentiated into cells with properties of pancreatic beta cells, and studied for beta cell function in terms of basal and stimulated insulin secretion. Our hypothesis is that iPS-derived beta cells from patients who carry T2D associated gene variants will show defects in insulin secretion and beta cell function. This would provide 1) proof of the concept that primary abnormalities in beta cell function can cause T2D, 2) cell lines useful to study the pathophysiology of T2D development, 3) a tractable system to test genetic, pharmacologic, and cell-based therapies for T2D. If achieved, this proposal would represent an important contribution to our understanding of the basic disease mechanisms for T2D, and may lead to novel new treatments and individualized medicine. This project is interdisciplinary and cross-institutional in that it involves clinical cardiology at MGH, genome-wide association researchers at the Broad Institute, Harvard Stem Cell Institute members involved with iPS generation and Joslin Diabetes Center experts on pancreatic beta cells and their function.

Appetite Hormone Ghrelin in the Saliva of Type-II Diabetes-Associated Periodontitis

Principal Investigator: Toshihisa Kawai, DDS, PhD, The Forsyth Institute

Co-Investigator(s): Nadeem Y. Karimbux, DMD, MMSc, Harvard School of Dental Medicine
Winston P. Kuo, DMSc, DDS, Harvard Medical School
Alexander R. Ivanov, PhD, Harvard School of Public Health
George L. King, MD, Joslin Diabetes Center
Robert J. Genco, DDS, PhD, State University of New York at Buffalo

Accumulated lines of evidence have demonstrated that periodontal disease (PD) is more severe and more prevalent in persons with type-2 diabetes mellitus (DM-II) than non-diabetics. A preliminary antimicrobial activity assay carried out in our laboratory indicated that Ghrelin, which is an appetite peptide-hormone secreted in both stomach and saliva, possesses an unreported, but strong, antimicrobial activity to periodontal pathogens. Recent studies have demonstrated that Ghrelin is secreted in saliva at significantly lower levels in DM-II patients than healthy control subjects. Therefore, we hypothesize that diminished salivary Ghrelin levels in DM-II patients would permit the outgrowth of periodontal pathogens. To gain insight into the role that Ghrelin plays in the association between PD and DM-II, we propose a study involving 100 subjects (DM-II with and without PD, non-diabetic PD patients, and healthy control, n=25, respectively). A multidisciplinary study team, composed of periodontists, immunologists, systems biologists, endocrinologists, and specialists in proteomics from across the broad spectrum of clinical and basic science disciplines at Harvard University, will then conduct a hypothesis-driven clinical study to evaluate Ghrelin salivary titers in order to assess its antimicrobial activity in relation to DM-II-associated PD. Our long-term goal is to develop a saliva-based point-of-care (POC) diagnostic system and other therapeutic approaches to ameliorate the effects of DM-II-associated PD.

Patterns of Use of Newly Approved Orphan Drugs for Rare Diseases

Principal Investigator: Aaron Kesselheim, MD, JD, MPH, Brigham and Women's Hospital

Co-Investigator(s): Jerry Avorn, MD, Brigham and Women's Hospital
Daniel Paul Carpenter, PhD, Faculty of Arts and Sciences, Harvard University
Anup Malani, JD, PhD, Harvard Law School
Arthur A. Daemmrich, PhD, Harvard Business School
Richard G. Hamermesh, MBA, DBA, Harvard Business School

The 1983 Orphan Drug Act authorized financial incentives for pharmaceutical companies to invest in basic and translational research to develop drugs for rare diseases (defined as those affecting < 200,000 patients). To date, over 325 drugs have been approved as “orphan drugs” and received the offered subsidies. However, in assessing the impact of this legislation on translational research, there has been little formal evaluation of how orphan drugs are prescribed once they are approved. Off-label use of such drugs in conditions for which efficacy evidence has not been reviewed by the FDA is common. This may be clinically appropriate if supported by sufficient evidence, but when it is not, such use may confer little benefit, put patients at risk, and lead to wasteful spending of scarce health care resources. In this pilot study, we will investigate the patterns of use of orphan drugs after FDA approval. First, we will conduct a full literature review to identify the evidence bases for potential uses of these drugs. Then, we will employ an extensive database of medication use and health care utilization including diagnoses and filled prescriptions for approximately 425,000 patients—supplemented with additional data as necessary from a large health insurance company—to evaluate the patterns of use of these orphan drugs and define the clinical and demographic characteristics of patients who are prescribed them for several specific diagnoses. We will stratify such use three ways: (a) that which is in accordance with FDA indications, (b) that which is not FDA-approved but is supported by published efficacy evidence, and (c) that which is not FDA-approved and is not supported by efficacy evidence. We will analyze the rate of uptake of orphan drugs after FDA approval, as well as their cost, in each of these strata, to answer two specific questions: first, how much orphan drug use falls into each category, and second, what are the costs attributable to each category of orphan drug use. We will draw on the multidisciplinary expertise of the co-investigators in this project to examine—from the perspectives of clinical medicine, epidemiology, regulatory policy, and health economics—the effects of the Orphan Drug Act on innovation and translational research, and its implications for subsequent uses of the orphan drugs developed.

Targeted Approach to Treatment of Malignant Melanoma

Principal Investigator: Roya Khosravi-Far, PhD, Beth Israel Deaconess Medical Center

Co-Investigator(s): David E. Fisher, MD, PhD, Massachusetts General Hospital

Abstract withheld at the request of the investigator.

Pharmacological Treatment of Rett Syndrome by Stimulation of Synaptic Maturation with IGF-1

Principal Investigator: Omar Khwaja, MD, PhD, Children's Hospital Boston

Co-Investigator(s): Michael Shannon, MD, MPH, Children's Hospital Boston
Leonard Rappaport, MD, Children's Hospital Boston
Ingrid Holm, MD, MPH, Children's Hospital Boston
Mriganka Sur, PhD, Massachusetts Institute of Technology
Matt Gregas, PhD, Children's Hospital Boston
Yaron Finkelstein, MD, Children's Hospital Boston and Hospital for Sick Children

Background: Rett syndrome (RTT) is a severe genetic form of autism in girls. Loss of functional MECP2 gene product causes impaired synaptic maturation in cortex and brainstem. Patients consequently have abnormal cortical function and autonomic/respiratory dysregulation including sudden death. There is no treatment for RTT. Work to date strongly implicates the IGF1 pathway in ameliorating symptoms of Rett syndrome (RTT) in *Mecp2*-null mice, an established experimental model for RTT. IGF1 is FDA-approved for clinical use in children. Objective: We propose to investigate the efficacy and safety of recombinant human Insulin-like Growth Factor (Increlex®, Tercica, Inc) in treating autonomic and neurodevelopmental dysfunction in girls with RTT. Aims: Thirty patients will be enrolled in a randomized double-blind, placebo-controlled cross-over clinical trial of treatment with IGF1. This design ensures all subjects have access to the trial medication and allows for secondary analyses of change in the post-treatment group. Primary and secondary outcomes: Improvement in cardiorespiratory regulation and neurodevelopmental function measured using a novel electrophysiological biomarker of autonomic/respiratory function and validated survey instruments. Hypothesis: Subjects with RTT will show sustained improvements in cardiorespiratory regulation and neurodevelopmental measures during treatment with IGF1. Significance: This pilot study will provide critical data for a broader study of pharmacological therapy to stimulate synaptic maturation in RTT and other neurodevelopmental disorders.

Is There a Cancer Developmental Signature in “Normal Tissue”?

Principal Investigator: Frank Kuo, PhD, Brigham and Women's Hospital

Co-Investigator(s): Isaac Kohane, MD, PhD, Children's Hospital Boston
Kamila Naxerova, Harvard Medical School

Prior studies of gene expression in various cancers have demonstrated the insights obtained by measuring how gene expression changes mirrored those in tissue development. In the course of these studies, some of the “normal tissues” that served as controls had developmental changes similar to the malignant tissues. We propose to use the Pathology Specimen Locator to identify breast cancer specimens that include “normal” margins and to compare the microRNA (miRNA) developmental signature as a function of distance from the malignancy. This study may affect surgical management of newly diagnosed patients with breast cancer and also offer more accurate prognoses. This entails the following specific aims: Aim 1: Obtain tissue from paraffin archives at multiple Catalyst hospitals from patients with primary breast cancer where the sample includes wide margins. Also 5 control samples (from breast reduction surgery) will be obtained. Aim 2: Extract miRNA from each of three sites of each of 10 tumors and the 5 controls sample and hybridize them with commercial miRNA microarrays. Aim 3: Identify those miRNA that are significantly differentially up and down-regulated (relative to the controls) in each of the samples. Determine if there is a gradient in the trend in the expression of these dysregulated miRNA from the tumor and through the two successively distant points outside the margin of the tumor. Aim 4: Project the dysregulated miRNA at the three locations against a previously obtained murine miRNA time-series of the developing breast. Determine if there is a developmental signature outside the histologically defined tumor and if it is in a gradient through the three locations.

Fate-mapping of Human Adipogenesis with Stable Isotope Labeling and Quantitative Multiisotope Mass Spectrometry (MIMS)

Principal Investigator: Richard Lee, MD, Brigham and Women's Hospital

Co-Investigator(s): Claude Lechene, MD, Brigham and Women's Hospital
Evan Rosen, MD, PhD, Beth Israel Deaconess Medical Center
Matthew Steinhauser, MD, Brigham and Women's Hospital
Todd Perlstein, MD, Brigham and Women's Hospital
Sam Senyo, PhD, Brigham and Women's Hospital
Erin Bohula, PhD, Brigham and Women's Hospital

With the obesity epidemic, the adipocyte is increasingly taking center stage as not only a fat storage cell, but also as a mediator of obesity-associated metabolic and cardiovascular diseases. Thus, understanding the dynamics of adipogenesis in humans is highly significant. While there is some evidence that humans maintain low levels of adipocyte turnover into adulthood, a practical approach to adipogenesis in humans has not yet been described. Our central hypothesis is that healthy adult humans maintain a low level of adipogenesis and that new adipocytes are derived from a preadipocyte precursor cell. A combination of stable-isotope labeling followed by fat biopsy analysis with a revolutionary mass spectrometry based technique will enable us to definitively answer this question. We will employ ^{13}C -oleic acid as an adipocyte-specific metabolic label. Mature adipocytes are the only cell type within adipose tissue that take up and store oleic acid. We will follow the ^{13}C -oleic acid pulse with a ^{15}N -thymidine infusion, which will label any dividing cells. The use of these safe stable isotopes with MIMS will allow the precise quantitation of the rate of adipogenesis and, more importantly, define the parent cell as a pre-existing mature adipocyte or a preadipocyte precursor cell. Our aims are: Specific Aim 1: To test the hypothesis that pulse labeling with stable isotope labeled thymidine followed by MIMS analysis will enable the quantitation of adipogenesis in human subjects. Specific Aim 2: To test the hypothesis using sequential stable isotope labeling and MIMS analysis that a preadipocyte precursor cell pool functions as the source for new adipocytes in humans.

Neural Prosthetics with Chemical Harvesting and Stimulation for Facial Nerve Reanimation

Principal Investigator: Samuel Lin, MD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Jongyoon Han, PhD, Massachusetts Institute of Technology
Rahul Sarpeshkar, PhD, Massachusetts Institute of Technology
Amr N. Rabie, MD, Beth Israel Deaconess Medical Center
Yong-Ak Song, PhD, Massachusetts Institute of Technology

Abstract withheld at the request of the investigator.

Generation of Liver Tissue *In Vitro* Using Genetic Reprogramming Technology

Principal Investigator: David Lum, PhD, Massachusetts General Hospital

Co-Investigator(s): Chad A. Cowan, PhD, Massachusetts General Hospital
Joseph P. Vacanti, MD, Massachusetts General Hospital
Sangeeta Bhatia, MD, PhD, Massachusetts Institute of Technology
Howard Pryor, II, MD, Massachusetts General Hospital
Kartik Trehan, Massachusetts Institute of Technology

Liver disease is the tenth leading cause of death in the United States, attributing to over 30,000 deaths a year. At the present time, there is no pharmaceutical agent or device that is capable of performing the myriad functions of the liver and the demand for liver transplantation continues to increasingly outpace supply. Although cell-based therapy is considered a possible alternative to transplantation, currently there is no easily attainable hepatocyte cell source available that retains liver function *in vitro*. Our objective is to define methodology that will allow the creation of expandable, patient-specific hepatocyte-like cells that are suitable for cell-based therapy and the *in vitro* modeling of liver function and disease. HNF4A is a key regulator of hepatocyte cell fate. We have used the ectopic expression of HNF4A in human adipose-derived mesenchymal stem cells (ADMSCs) to generate reprogrammed hepatocyte-like cells (rHep(HNF4A)). Our first aim is to molecularly and functionally characterize the reprogrammed hepatocyte-like cells to determine their similarity to primary hepatocytes. Given the feasibility of generating patient-specific ADMSCs, our second aim is to generate HNF4A reprogrammed, hepatocyte-like cell lines from patients with liver diseases such as inborn errors of metabolism and Hemophilia B. These cell lines will facilitate the *in vitro* modeling of many diseases. Our third aim is to assess the suitability of HNF4A reprogrammed hepatocyte-like cells in cell based therapies including cell transplantation and bio-artificial liver devices.

Polymer Bacterial Mimics as Cancer Vaccines

Principal Investigator: David Mooney, PhD, Harvard School of Engineering and Applied Sciences

Co-Investigator(s): Glenn Dranoff, MD, Dana-Farber Cancer Institute

One half of all men, and one third of all women in the US will have cancer in their lifetime, and cancer remains a major cause of death. The goal of these studies is to create polymers to both prevent and treat cancer. The utility of cancer vaccines is dependent on their ability to activate the immune system to destroy tumor cells, and the hypothesis underlying this application is that a polymer that mimics bacterial infection, while simultaneously providing cancer antigen, can effectively and quantitatively recruit, activate and disperse host dendritic cells (DCs), and the programmed dendritic cells will be capable of stimulating specific T-cell populations and eliciting a strong anti-tumor response. The Specific Aims to be addressed in the application are (1) design and develop a polymer system to allow for controlled presentation of stimulatory factors (GM-CSF and CpG oligonucleotides) to host DCs to control their recruitment to the material, and subsequent maturation, and (2) test the ability of these materials to recruit, program and enhance lymph node homing of cancer antigen-primed DCs in order to provide effective cancer vaccines against melanoma in both a prophylactic and therapeutic context. It is anticipated that the polymers developed in this project may provide a more practical and effective vaccine than the cell-based vaccines currently under development. Further, this general approach of programming cells in situ may provide a powerful new approach to cell therapy broadly.

Accelerated Contrast-enhanced Whole Heart Coronary MRI with Compressed Sensing

Principal Investigator: Reza Nezafat, PhD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Warren J. Manning, MD, Beth Israel Deaconess Medical Center
Vahid Tarokh, PhD, Harvard School of Engineering and Applied Sciences
Roger W. Brockett, PhD, Harvard School of Engineering and Applied Sciences
Mehmet Akcakaya, Harvard School of Engineering and Applied Sciences

Abstract withheld at the request of the investigator.

Evaluating Temporal Patterns of Glycemic Control in Patients with Type I Diabetes Using a New i2b2/SHRINE Cell for Automated Time Series Analysis

Principal Investigator: Daniel Nigrin, MD, Children's Hospital Boston

Co-Investigator(s): Griffin Weber, MD, PhD, Beth Israel Deaconess Medical Center/Harvard Medical School
Allison Goldfine, MD, Joslin Diabetes Center
Maryanne Quinn, MD, Children's Hospital Boston
Howard Wolpert, MD, Joslin Diabetes Center
Lori Laffel, MD, MPH, Joslin Diabetes Center
Allesandro Doria, MD, PhD, MPH, Joslin Diabetes Center

Type 1 Diabetes Mellitus affects 1.7 per 1000 individuals age 0-19 years living in the United States, and young adults with diabetes face an increased risk of premature morbidity and mortality. We aim to determine whether there are temporally related patterns between glycemic control and routine diabetes maintenance visit history. In addition, we aim to examine duration of poor glycemic control and incidence of subsequent development of diabetes-related complications. Although these are not inherently new postulates, they have traditionally been difficult to elucidate through traditional clinical trial means, as they deal with a chronic illness that requires long periods of followup, as well as transitions from institution to institution, and from pediatric to adult providers. To enable our study, we will create new functionality for the Catalyst i2b2/SHRINE system to allow it to perform time series analyses for specific cohorts of patients. We will implement this new functionality at BIDMC, CHB and JDC. In addition, we will develop tools to allow for linking of patients between institutions, to track them as they progress from pediatric care at CHB to adult care at the JDC or BIDMC for example. The temporal analysis tools we develop through this project will be implemented such that they can be applied to disease in any medical discipline, and can be extended to any institution using the i2b2/SHRINE platform.

Use of Functional Neuroimaging to Assess Effects of Viewing the Built Environment.

Principal Investigator: Nicolas Oreskovic, MD, MPH, Massachusetts General Hospital

Co-Investigator(s): Moshe Bar, PhD, Massachusetts General Hospital
Suzanne Lanyi Charles, PhD, Harvard University Graduate School of Design
Didi Tsigaridi, Harvard University Graduate School of Design

Background: The rapid rise in obesity around the world reflects substantial lifestyle changes in diet and exercise patterns. Physical activity has many known health benefits. Attempts to increase physical activity, however, often require substantial lifestyle modifications, have typically impacted a minority of the population, and tend to have had limited effectiveness. One solution to increasing the physical activity is through increased use of the built environment. Advantages to this approach include the possibility of daily exercise, accessibility to a wide swath of the population, and the ability to promote exercise without demanding a major effort on the part of the individual. Determining which built environment characteristics are most useful in promoting physical activity, however, requires a better understanding of how design is perceived by humans, consciously and unconsciously. Objective: We propose to study how various design elements in the built environment are registered by the human brain, and to determine whether certain design elements drive or deter use of the built environment. This unique study represents a novel cross-disciplinary collaboration between the fields of medicine, cognitive neuroscience, and architecture. Aims: The aims of the study are: 1) To assess via functional MRI (fMRI) whether viewing different built environment characteristics are associated with a specific pattern of brain activation, 2) To assess whether LEED streetscape characteristics identified as important in promoting walkability can activate emotion-processing or motor-function centers within the central nervous system, thus suggesting the capacity for responses to viewing built environment elements.

Utilizing Micro Array Technology as a Novel Approach to Chronic Wound Care

Principal Investigator: Dennis Orgill, MD, PhD, Brigham and Women's Hospital

Co-Investigator(s): Lifei Guo, MD, PhD, Brigham and Women's Hospital
Daniel Irimia, MD, PhD, Massachusetts General Hospital
Mehmet Toner, PhD, Massachusetts General Hospital
Doug Helm, MD, Brigham and Women's Hospital
Taylor Lloyd, Brigham and Women's Hospital

With the incidence of diabetes surging worldwide to epidemic proportions, the burden of chronic wounds is also growing with resulting enormous health-care expenses. In 2001, nearly 11 billion dollars in the U.S. was spent on care of diabetic foot ulcers and amputations. However, the medical and surgical treatment of diabetic wounds remains difficult and is often insufficient, leading to a high amputation rate among those patients. A novel approach is needed to accelerate wound healing in order for patients to maintain function and return to productive lives as well as to reduce the economic burden of wounds on our society. Adding proliferating fibroblast and keratinocyte cells directly to a wound bed has been shown to accelerate wound closure in animal studies and in limited clinical studies on chronic wounds. However, the major obstacle in translating this research to clinical application is the difficulty of applying cells onto a wound. To address the limitations of current wound care technologies, we hypothesize that local delivery and reliable uniform distribution of actively proliferating cells will accelerate wound healing further. To this end, a device is needed that will deliver cells and growth factors under precisely controlled conditions to promote the healing of chronic wounds. We propose an interdisciplinary research effort to bridge the obstacle between laboratory studies that show the effectiveness of cultured cells on chronic wounds and the clinical practice of applying and maintaining these cells on the wound bed. We aim to use current microfabrication techniques to develop a micro array chamber (M.A.C) device that can topically deliver cells to a wound. Following a pilot animal study to test this device on a diabetic mouse model, we aim to use extra-mural funding to both expand upon what we will learn from this pilot study and to conduct a future clinical study of the M.A.C device.

Clinical Implementation of a Novel, Non-invasive, Closed-loop, Patient-specific Electrographic Seizure Detection and Automatic Vagus Nerve Stimulator Activation System, with an Audible Alarm in Patients with Refractory Epilepsy

Principal Investigator: Trudy Pang, MD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Ali Shoeb, Massachusetts Institute of Technology
John Guttag, PhD, Massachusetts Institute of Technology
Steven Schachter, MD, Beth Israel Deaconess Medical Center
Elizabeth Thiele, MD, PhD, Massachusetts General Hospital

Uncontrolled seizures represent a major cause of disability, morbidity, and mortality for 1 in 4 patients with epilepsy, or over 500,000 persons in the U.S. alone. Currently, there is no means available to automatically initiate non-invasive abortive therapy at the onset of a seizure. Additionally, many patients lose awareness when a seizure begins and therefore cannot seek an environment that is safe for them in the event of a seizure. Our group has developed a prototype for a non-invasive, wearable seizure detection system equipped with an alarm. The detection system is capable of learning to recognize the changes in a patient's brainwaves associated with the onset of a seizure with high sensitivity and specificity. The audible alarm at the onset of a seizure could ameliorate the consequences of seizures by allowing time for patients to seek a safe position or to administer an acute therapy. We have further integrated this seizure detection system with the Vagus Nerve Stimulator (VNS), to trigger potentially abortive therapy by initiating VNS stimulation when a seizure is detected electrographically. The main goals of this proposal are: 1) Assess the feasibility of implementing the closed-loop automatic seizure detection, VNS stimulation in inpatients with frequent seizures; 2) Evaluate the clinical impact of closed-loop VNS stimulation on seizure severity, duration, and recovery period, as well evaluate the impact on cognitive function during electrographic seizures; 3) Develop a wireless, ambulatory, closed-loop seizure detection, VNS stimulation, and alarm-generating system that can be used in the outpatient setting.

Magnetic Resonance Imaging as a Genomic/Proteomic Expression Correlate to Characterize Renal Cell Carcinoma

Principal Investigator: Ivan Pedrosa, MD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Sabina Signoretti, MD, Brigham and Women's Hospital
Long Ngo, PhD, Beth Israel Deaconess Medical Center
Sandra Gaston, PhD, Beth Israel Deaconess Medical Center
Rupal Bhatt, MD, PhD, Beth Israel Deaconess Medical Center
Elizabeth Genega, MD, Beth Israel Deaconess Medical Center
Andrew Wagner, MD, Beth Israel Deaconess Medical Center
David Alsop, PhD, Beth Israel Deaconess Medical Center
Winston Kuo, DMSc, DDS, Harvard Medical School
Colin Brenan, PhD, Biotrove

Background: Inactivation of the VHL gene, HIF upregulation, and VEGF over-expression are molecular markers of angiogenesis in RCC. Arterial spin labeling (ASL) is a magnetic resonance imaging (MRI) method for measuring blood flow by manipulating the signal from inflowing arterial blood and provides direct evaluation of tumor perfusion. Signal intensity on blood oxygen level-dependent (BOLD) imaging, an MRI technique, is modulated by the oxygen levels and flow. Objective: We seek to identify vascular MRI measures in renal masses that correlate to molecular alterations promoting angiogenesis, including VHL inactivation and VEGF over-expression. We will also attempt to define molecular pathways that correlate with different levels of tumor perfusion *in vivo*. Aim 1: To compare MRI measurements of vascularity and hypoxia within the tumor in-vivo in RCC patients with the results of molecular assessments in the same tumor after nephrectomy. a. Average and peak tumor perfusion on ASL MRI and R2* measurements on BOLD MRI will be correlated with VHL inactivation and overall VEGF over-expression in the tumor. b. Areas of high and low perfusion and oxygenation within the mass on ASL and BOLD MRI, respectively, will be spatially co-registered and correlated to genomic/proteomics and expression of angiogenesis and hypoxia markers in the tumor. Aim 2: To develop new immunohistochemical stains for RCC that correlate with the spatially co-registered over-expressed markers of angiogenesis.

CMV-specific Cellular Immunity in Breast Milk and Protection Against Postnatal Cytomegalovirus Transmission to the Breastfeeding Infant

Principal Investigator: Sallie Permar, MD, PhD, Children's Hospital Boston/Beth Israel Deaconess Medical Center

Co-Investigator(s): Karen M. Puopolo, MD, PhD, Brigham and Women's Hospital
Mary Lucia Gregory, MD, MSc, Beth Israel Deaconess Medical Center
Helen H. Kang, Beth Israel Deaconess Medical Center
Amy Zolit, Brigham and Women's Hospital/Children's Hospital Boston

Neonatal cytomegalovirus (CMV) infection can lead to long-term neurologic consequences, most commonly hearing loss, and causes a severe infection in very low birth weight (VLBW) preterm infants. While the majority of CMV-seropositive women shed CMV in breast milk, postnatal transmission of CMV occurs in only 30 to 40% of breastfeeding infants, suggesting that immune factors in breast milk may prevent postnatal transmission. Virus-specific cellular immunity is crucial for the control of CMV viremia, but the role of CMV-specific immunity in breast milk on breast milk viral shedding and mother-to-infant transmission of CMV has not been established. In this study, we will compare the magnitude of the CMV-specific cellular immune response in breast milk to that in the peripheral blood of mothers of CMV-infected infants. Furthermore, we will define the relationship of CMV-specific cellular immunity in breast milk to CMV viral load in the breast milk of CMV-transmitting women. Finally, in a case-control study, we will compare the magnitude of the CMV-specific cellular immune response and CMV viral load in breast milk of postnatally transmitting and non-transmitting mothers of VLBW preterm infants. This study will identify the maternal breast milk immune correlates of protection against postnatal CMV infection. Defining the maternal immune correlates of protection against postnatal CMV transmission will determine the safety of breast milk feeding for VLBW preterm infants of CMV-seropositive women and form the basis for designing maternal vaccination strategies to prevent CMV transmission via breast milk.

Right Atrial 4q25 Gene Expression Signatures in Patients with Atrial Fibrillation

Principal Investigator: Tjorvi E. Perry, MD, Brigham and Women's Hospital

Co-Investigator(s): Jonathan Seidman, PhD, Harvard Medical School
Brendan Everett, MD, MPH, Brigham and Women's Hospital
Sary Aranki, MD, Brigham and Women's Hospital
Kuang-Yu Liu, PhD, Brigham and Women's Hospital
Prem Shekar, MD, Brigham and Women's Hospital
Simon C. Body, MBChB, MPH, Brigham and Women's Hospital

Atrial fibrillation (AF) is the most common sustained cardiac arrhythmia, and is associated with increased risk of stroke, congestive heart failure, and premature death. Recently, 4q25 locus variants have been associated with AF in ambulatory patient populations. We have recently shown that 4q25 variants are associated with postoperative AF after cardiac surgery. We hypothesize that right atrial 4q25 mRNA expression differs between individuals who develop postoperative AF, depending on whether or not the 4q25 risk allele is present. The principal specific aim of this proposal is to identify the association between right atrial 4q25 mRNA expression and atrial fibrillation after cardiac surgery in patients stratified by the 4q25 risk allele. Specifically, we will collect right atrial tissue samples, whole blood for DNA extraction, and extensive demographic and perioperative information from 120 patients undergoing cardiac surgery. To identify the risk allele carriers, we will genotype the single nucleotide polymorphism, rs2200733, in all patients. We will subsequently compare right atrial 4q25 mRNA expression in carriers of the risk allele, who also develop postoperative AF, with patients who do not carry the risk allele, and/or do not develop postoperative AF. This study design will allow us to identify differentially expressed, even novel, 4q25 transcripts in individuals with postoperative AF who carry the 4q25 risk allele.

A Novel Application of Transcranial Magnetic Stimulation to Assay Synaptic Plasticity as a Potential Endophenotype and Clinical Outcome Measure in Fragile X

Principal Investigator: Jonathan Picker, MBChB, MD, Children's Hospital Boston

Co-Investigator(s): Joseph Gonzalez-Heydrich, MD, Children's Hospital Boston
Alexander Rotenberg, MD, PhD, Children's Hospital Boston/Beth Israel
Deaconess Medical Center
Alvaro Pascual-Leone, MD, PhD, Beth Israel Deaconess Medical Center
Lindsay Oberman, PhD, Beth Israel Deaconess Medical Center

Fragile X (FX), the most common inherited form of mental retardation (MR), is caused by a mutation in the FX mental retardation gene. This mutation impairs the capacity of neuron-to-neuron connections to store memories. Memories are stored as neuron-to-neuron connections which strengthen (termed long-term potentiation, LTP) or weaken (termed long-term depression, LTD) with experience. LTP and LTD (LTP/LTD) are measured invasively by electrical brain stimulation, and are abnormal in mice with the FX mutation. Similar abnormalities likely underlie MR in FX patients. However, there is no clinical test of LTP/LTD in humans. In preclinical trials, drug treatments that improve learning in FX mice also improve LTP/LTD. If this is true in humans, then a clinical measure of LTP/LTD after drug treatment can be a useful method to rapidly identify effective therapies which can then proceed to longer behavioral trials to see if they can prevent MR in FX patients. A clinical measure of LTP/LTD could also be used in the same way to unblock the path to the development of therapeutics for Tuberous Sclerosis, Neurofibromatosis, and other disorders of the neurochemical pathway that is critical for normal LTP/LTD. Transcranial magnetic stimulation (TMS) is a noninvasive, safe and painless technique recently developed to measure LTP/LTD in humans. Accordingly, we propose to investigate whether TMS measures of LTP/LTD predict symptom severity in FX patients as a step toward developing a novel test of treatment efficacy in FX and other forms of MR.

Discovery of Novel Sarcomere Gene Splice Variants in Human Heart Failure

Principal Investigator: William T. Pu, MD, Children's Hospital Boston

Co-Investigator(s): David Hill, PhD, Dana-Farber Cancer Institute
Anju Nohria, MD, Brigham and Women's Hospital
Rajan Sah, MD, PhD, Brigham and Women's Hospital
Yongwu Hu, PhD, Children's Hospital Boston
Changyu Fan, MS, Dana-Farber Cancer Institute

Heart failure is a leading cause of morbidity and mortality worldwide. Current therapeutic options in heart failure are limited, so that 70-80% of heart failure patients will die within eight years. Improved treatment options require greater understanding of the pathogenesis of heart failure. Sarcomere genes provide heart muscle cells the ability to contract. We hypothesize that in heart failure alternative assembly of sarcomere gene messenger RNAs occurs (alternative splicing), and that this impairs sarcomere gene function. While studying the sarcomere gene troponin T2 (TNNT2), 4/40 transcripts analyzed from diseased hearts had altered splicing, resulting in a novel isoform with in-frame deletion of one amino acid. This splicing variant was not detected in nonfailing hearts. We hypothesize that other heart disease related splice variants exist and have escaped recognition because the transcriptomes of diseased and normal myocardium have not suitably analyzed. In this proposal, we test this hypothesis through two specific aims. First, using massively parallel sequencing we will perform deep sequencing of sarcomere transcripts from diseased and normal myocardium to identify disease-related splice variants. For detected variants, we will analyze genomic DNA to exclude potential contribution of genomic variation in sarcomere genes. Second, we will examine the effect of mechanical unloading of the failing human heart to determine if the expression of these variants is related to myocardial load. This proposal has the potential to uncover a novel disease mechanism that contributes to the pathogenesis of human heart disease.

Attention Deficit Disorder and Schizophrenia as Disorders of Neuronal Adaptation

Principal Investigator: Tommi Raij, MD, PhD, Massachusetts General Hospital

Co-Investigator(s): Donald C. Goff, MD, Massachusetts General Hospital
Jyrki Ahveninen, PhD, Massachusetts General Hospital
Matti Hamalainen, PhD, Massachusetts General Hospital
Fa-Hsuan Lin, PhD, Massachusetts General Hospital

For two stimuli presented consecutively, the brain's response to the second one is smaller ("adaptation"). Recovery time from the previous stimulus is called adaptation lifetime. Adaptation supports neuronal activation traces and core sensory-perceptual functions such as sensory memory and temporal windows of integration (TWIs). We have recently developed novel tools that map the adaptation status throughout the brain with unprecedented accuracy. We found that in healthy subjects, adaptation lifetimes are shortest in low-order sensory cortices and systematically increase towards high-order association areas, which apparently supports brain functional hierarchy. Objective. As a novel hypothesis, we suggest that attention deficit disorder (ADD) and schizophrenia reflect abnormalities in the hierarchy of adaptation lifetimes. ADD patients may have too short adaptation lifetimes, resulting in shortened TWIs and attention span. Schizophrenic patients may have prolonged adaptation lifetimes, resulting in overtly long TWIs and increased associations along with excessive top-down feedback to sensory areas, leading to sensory and cognitive deficits, paranoia, hallucinations, and avoidance of intensive stimulus environments. This model could explain many symptoms in ADD and schizophrenia and allow targeted development of a new generation of both diagnostic as well as therapeutic tools. Aims. We propose to measure the adaptation lifetimes in 8 ADD and 8 schizophrenic adults using our already existing and proven fMRI and MEG paradigms. We hypothesize that ADD is associated with shortened and schizophrenia with prolonged lifetimes compared to normal subjects.

Development of an Iterative Platform that Accelerates the Discovery and Validation of Transcription Factor Inhibitors

Principal Investigator: Alan Rigby, PhD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Andrew L. Kung, MD, PhD, Dana-Farber Cancer Institute

It is well established that a limited number of transcription factors are indeed overactive in many human cancers and that these overactive transcription factors represent obvious and direct anticancer targets. The ability to selectively target and inhibit transcription factors (TFs) through selective and specific inhibition of the TF-DNA interaction interface represents a paradigm shift in transcriptional regulation and offers a unique opportunity to reprogram the expression signature of genes downstream of these TFs, which are critical in cancer development and progression. The PIs propose that small molecule inhibitors of these TFs can be rapidly identified, validated and optimized through an iterative partnership involving a computer aided drug discovery (CADD) platform developed by the Rigby laboratory and a high throughput functional screen that will be co-developed by the PIs of this proposal. This Catalyst Pilot proposal is focused on the PIs collective expertise in structural/computational biology and *in vitro/vivo* functional evaluation, which will permit us to rapidly explore and validate small molecule inhibitors that selectively target unique chemical space, the TF-DNA interaction interface with refined selectivity and specificity. This multi-disciplinary, multi-institutional discovery platform provides an exciting new approach for the identification of novel therapies for the treatment of cancer with an initial focus on breast, prostate, leukemia, as well as pediatric cancers such as Ewing's Sarcoma.

Adoptive Cell Therapy with Anti-tumor T Cells that are Resistant to Inhibition in the Tumor Microenvironment

Principal Investigator: Jerome Ritz, MD, Dana-Farber Cancer Institute

Co-Investigator(s): Michail V. Sitkovsky, PhD, Northeastern University
Akio Ohta, PhD, Northeastern University
Dmitriy Evgenyevich Lukashev, PhD, Northeastern University
Jorgen Kjaergaard, PhD, Northeastern University
Steven Hatfield, Northeastern University
Manasa Madasu, Northeastern University
Heather Daley, Dana-Farber Cancer Institute

Although adoptive T cell therapy can eradicate tumors in mice, this approach has had limited success in patients with cancer. In preclinical models, hypoxia plays a critical role in protecting tumor cells from adoptively transferred T cells. The primary mediator of this resistance is extracellular adenosine, which accumulates in the tumor microenvironment in response to local hypoxia. Cytolytic T cells (CTL) express adenosine receptors (A2AR) and activation of A2AR leads to accumulation of cAMP and inhibition of T cell function. Blockade of A2AR facilitates tumor destruction *in vitro* and *in vivo*. We have also recently shown that ex vivo expansion of CTL in the presence of A2AR agonists results in selective expansion of adenosine-resistant CTL (AR-CTL) that mediate more effective destruction of tumors *in vivo*. The proposed studies will determine whether human CTL can be engineered to acquire resistance to extracellular adenosine. Once methods for expanding AR-CTL are established, clinical-scale manufacturing procedures will be developed and validated. These studies will focus on EBV-specific CTL, which are currently being manufactured for treatment of naso-pharyngeal carcinoma and other cancers. In these patients, EBV-specific CTL induce tumor regression but responses are seldom complete and tumors eventually progress. Future studies will evaluate the safety and efficacy of adoptive therapy with adenosine-resistant EBV-specific CTL in a Phase I clinical trial. This will represent the first-in-human evaluation of a novel method for improving the efficacy of adoptive T cell therapy and, if successful, will have broad application to the field of cellular therapy for cancer.

Establishing a Platform for Quantifying the Anti-atherosclerosis Action of HDL in Humans

Principal Investigator: Frank Sacks, MD, Harvard School of Public Health

Co-Investigator(s): Annaswamy Raji, MD, Brigham and Women's Hospital
David C. Henderson, MD, Massachusetts General Hospital
Carlos Mendivil, MD, Harvard School of Public Health

Background: A low blood concentration of high density lipoproteins (HDL) is a strong predictor of cardiovascular disease (CVD). A major function of HDL is to remove excess cholesterol from vascular cells in arteries prone to atherosclerosis, a process termed reverse cholesterol transport. Evidence is accumulating that the function of HDL may be more important to cardiovascular disease protection than its concentration in the blood. However, reverse cholesterol transport by HDL has never been quantified in humans, since no method has been devised. Objective: We plan to develop a detailed picture of normal HDL metabolism and how it is altered in several clinical settings associated with a high incidence of atherosclerosis: obesity, insulin resistance, type 2 diabetes, and antipsychotic drug treatment. Aims: During the project year, we will study 10 human subjects, 5 with normal HDL and 5 with low HDL concentrations in the clinical settings of obesity and insulin resistance. We expect that the groups will have large differences in metabolic rates and fluxes through their HDL pathways in reverse cholesterol transport. This knowledge is crucial for development and clinical interpretation of lifestyle and pharmacological interventions that raise a low HDL concentration. With the results from this study, HDL interventions could be evaluated not only by their effects on HDL cholesterol concentrations but by effects on physiology. Interventions considered promising to develop or recommend for therapy move HDL physiology toward normal settings.

M-array: A Novel Microarray Technique for Simultaneous Detection of Large and Small Mutations

Principal Investigator: Yiping Shen, PhD, Massachusetts General Hospital

Co-Investigator(s): James Gusella, PhD, Massachusetts General Hospital
Bai-Lin Wu, PhD, Children's Hospital Boston

Abstract withheld at the request of the investigator.

Pilot Trial of Bumetanide for Neonatal Seizures

Principal Investigator: Janet Soul, MD, Children's Hospital Boston

Co-Investigator(s): Frances Jensen, MD, Children's Hospital Boston
Kevin Staley, MD, Massachusetts General Hospital
Ann Marie Bergin, MD, Children's Hospital Boston
Deirdre O'Reilly, MD, MPH, Children's Hospital Boston
Sarah Barnett, MD, MPH, Massachusetts General Hospital

We hypothesize that bumetanide, added to conventional GABAergic antiepileptic drugs (AEDs), will be a safe and well tolerated medication that will reduce neonatal seizures caused by acute neurologic disorders, compared with conventional antiepileptic drugs alone. The hypothesis is derived from recent basic science research performed by Harvard researchers showing that bumetanide has a novel age-specific mechanism of action to reduce seizure activity by blocking a chloride transporter that is highly expressed in the newborn brain, thereby enabling currently used GABAergic medications to exert an inhibitory effect on immature neurons. This pilot trial will be a translational research study and the first trial of a novel antiepileptic medication to be studied in newborns in decades. Bumetanide is a commercially available drug that has been used for over 30 years in newborns and children as a diuretic. We propose a cross-institutional translational research study at Children's, Massachusetts General and Brigham and Women's Hospitals, to bring this novel AED from the laboratory to the bedside for the treatment of neonatal seizures. The primary aims of this Phase I/II trial are to determine the safety and pharmacokinetics of bumetanide in newborns with refractory seizures due to hypoxic-ischemic encephalopathy, stroke and intracranial hemorrhage. The pharmacokinetic and safety data obtained in the proposed pilot study are needed to design a multicenter Phase III trial to determine the efficacy of bumetanide for the treatment of neonatal seizures. This pilot study will be the critical and obligatory first step towards testing this promising novel agent in a hypothesis-driven multicenter trial.

An Inhalable Vaccine Delivery Platform for Effective and Efficient Immunization

Principal Investigator: Amit Srivastava, PhD, Children's Hospital Boston

Co-Investigator(s): Jose Gomez-Marquez, PhD, Massachusetts Institute of Technology – Edgerton Center

Christopher Hug, MD, PhD, Children's Hospital Boston

Background: Delivering vaccines by inhalation is recognized as a viable global health application and a safer alternative to injections. In the developing world, needles and syringes are often misused and reused, due to absence of trained medical personnel and inadequate biohazardous waste disposal facilities. Although the demand for a safe and painless alternative to needles and syringes is great, there are no convenient and field-deployable aerosol devices currently on the market. Objective: In this project, we aim to develop AEROVAX – a portable, needle-free inhalation platform for vaccine and drug delivery in a rapid, safe, painless and cost-effective manner under the extreme conditions of healthcare's last mile; the device works without electricity, overcomes cold chain issues and does not require medically-trained personnel to operate. Aims: (1) Optimize this inhalation platform using the off-the-shelf, efficacious and widely used measles and MMR vaccines by iterative multi-parameter analysis of aerosols generated by the device. (2) Concurrent device development using the dual principles of appropriate global health device design: participation of local end user community in the design process and interim in-country field testing.

The Resting Brain for Neurosurgical Planning

Principal Investigator: Steven Stuffelbeam, MD, Massachusetts General Hospital

Co-Investigator(s): Randy Buckner, PhD, Faculty of Arts and Sciences, Harvard University
Joseph Madsen, MD, Children's Hospital Boston
Emad Eskandar, MD, Massachusetts General Hospital

Background: The best hope for survival in patients with malignant brain cancer or epilepsy is resection of as much of the lesion as possible, while preserving as much normal functioning brain as possible. Currently, presurgical mapping of epilepsy and tumor patients requires a multilayered approach that involves several different invasive procedures and tests. Objective: Develop a method to replace these tests with a single non-invasive test that would map the entire brain—including the motor system, language systems, and memory systems—all in about 20 minutes, and without the need of patient active participation. Aims: We hypothesize that essential areas of cortex can be accurately mapped based on intrinsic functional connectivity using functional connectivity magnetic resonance imaging (fcMRI). Therefore patient compliance will not be required and a single, simple procedure could be used for many functional mapping needs. This project will also lead to a new language and memory lateralization technique replacing the traditional invasive Wada test and invasive brain mapping that both require a conscious and cooperative patient. Furthermore, by assessing the disruption of functional connectivity, the brain lesions and epileptogenic cortex may be identified. This project proposes a completely new approach for presurgical mapping for patients with brain lesions. If successful, it will decrease neurosurgical morbidity. This project is a joint effort of researchers and clinicians from MGH, Harvard University, and Children's Hospital Boston.

Microbiota Associated with Dental Decay Around Fixed Orthodontic Appliances

Principal Investigator: Anne C. R. Tanner, BDS, PhD, The Forsyth Institute

Co-Investigator(s): Andrew Sonis, DDS, Children's Hospital Boston
Natalia Chalmers, DDS, PhD, The Forsyth Institute
Ralph Kent, ScD, The Forsyth Institute
Bruce Paster, PhD, Harvard School of Dental Medicine/The Forsyth Institute

Fixed or bonded orthodontic appliances can result in rapid accumulation of dental plaque around the brackets. This bacterial accumulation increases risk of tooth demineralization, which leads to 'white spot' caries lesions (dental decay). Despite preventive measures, 50% of orthodontic patients at the Children's Hospital experience this problem. Dental plaque associated with caries represents a unique microbial ecosystem with high levels of acid-producing bacteria. Recent studies have revealed new cariogenic bacteria, in addition to *Streptococcus mutans*, particularly in the *Bifidobacterium/Scardovia* genera and unnamed, non-*mutans* *Streptococcus* species. The long-term goal of this proposal is to understand the cause of white spot lesions associated with orthodontic treatment in order to prevent these lesions. The objective of this application is to determine prevalence and association of bacterial species with white spot lesions. The central hypothesis is that white spot lesions are caused by specific acid-producing bacterial species. This hypothesis will be tested in two specific aims: 1) microarray identification of bacteria in white spot lesions, 2) quantification of *S. mutans*, *Bifidobacteria* and new caries-associated species in white spot lesions. This innovative approach combines comprehensive evaluation by microarray with targeted identification of key species by quantitative Real-Time PCR and Fluorescent In Situ Hybridization. At the conclusion of the proposed study, it is expected that new pathogens for white spot lesions will be identified. The proposed research is significant because it will improve our ability to predict development of white spot lesions, and indicate which species are risk indicators of future disease and must be suppressed for effective therapy. Interventions based on risk would prevent these lesions and the costly restorative treatments required to repair them.

Method of Inducing Brown Adipogenesis by BMP-7 as an Anti-obesity Therapy

Principal Investigator: Yu-Hua Tseng, PhD, Joslin Diabetes Center

Co-Investigator(s): Aaron Cypess, MD, PhD, Joslin Diabetes Center
Andrew White, MD, Beth Israel Deaconess Medical Center
Gerald Kolodny, MD, Beth Israel Deaconess Medical Center
Tim Schulz, PhD, Joslin Diabetes Center
Matthew Palmer, PhD, Beth Israel Deaconess Medical Center

Obesity is occurring at epidemic rates in the US and worldwide. This impacts the risk and prognosis of many diseases, including diabetes, cardiovascular disorders, cancers and others. Current clinical approaches for treatment of obesity have thus far met limited success, so the search for new therapies for obesity is urgently needed. Emerging evidence suggests that brown fat is normally present in humans and can most likely be induced and function in the context of adaptive thermogenesis. Promoting brown fat differentiation is therefore potentially a very attractive way to counteract obesity. We have recently discovered that bone morphogenetic protein (BMP)-7 significantly increases brown adipocyte differentiation, thermogenesis, and weight loss in mice, leading to the hypothesis that BMP-7 can be used as an anti-obesity therapy by increasing brown fat-mediated energy expenditure. To test this hypothesis, we propose to (1) determine the efficacy of pharmaceutical grade, recombinant human BMP-7 in murine preclinical studies, (2) test the brown adipogenic effect of BMP-7 in human adipose progenitor cells, and (3) ultimately analyze brown fat mass and energy metabolism in patients receiving BMP-7 treatment. The proposed research is innovative and represents a unique endeavor to combine interdisciplinary expertise to advance public health and improve quality of lives.

Dried Blood Spot-based HIV RNA Monitoring and Sensitive Genotyping During Directly Observed Antiretroviral Therapy in Haiti

Principal Investigator: Athe Tsibris, MD, Massachusetts General Hospital

Co-Investigator(s): Louise Ivers, MD, Brigham and Women's Hospital/Partners in Health

The use of effective antiretroviral therapy for HIV-infected patients in Haiti can result in short-term treatment outcomes that are similar to those seen in Western nations. The lack of viral load monitoring, however, complicates patient's and clinician's dedicated efforts to achieve and sustain durable viral suppression. When HIV viral load and resistance testing are not available, directly observed therapy is one strategy to improve patient adherence, maximize the chances of viral suppression, and minimize the emergence of resistance. The effect of such treatment programs on rates of viral suppression and HIV resistance is, however, unknown. Furthermore, the prevalence of primary HIV-1 drug resistance in Haiti and the genotypic patterns of resistance emerging during therapy have not previously been assessed. We hypothesize that directly observed antiretroviral therapy will improve rates of viral suppression and limit the development of HIV drug resistance, relative to historical controls. We propose to establish a pilot cohort of HIV-1 infected Haitian subjects initiating directly observed antiretroviral therapy and, using dried blood spots as source material, determine the proportion of subjects that achieve virologic suppression. We will use standard and sensitive (multiplex allele-specific PCR) HIV genotyping assays to investigate the cohort prevalence of HIV-1 reverse transcriptase and protease inhibitor resistance mutations at baseline and at the time of virologic failure. This proposal will provide data of importance to both individual patients and for the programmatic planning of antiretroviral therapy regimens for one of the largest HIV service providers in Haiti.

Structurally-reinforced Antigens for HIV Vaccination

Principal Investigator: Loren Walensky, MD, PhD, Dana-Farber Cancer Institute

Co-Investigator(s): Joseph Sodroski, PhD, Dana-Farber Cancer Institute
Jeffrey Supko, PhD, Massachusetts General Hospital
Greg Bird, PhD, Dana-Farber Cancer Institute
Alisa Perry, Dana-Farber Cancer Institute
Navid Madani, PhD, Dana-Farber Cancer Institute
Amy Princiotta, Dana-Farber Cancer Institute
Xiaoying He, PhD, Massachusetts General Hospital
Nela Zvereva, Massachusetts General Hospital

Abstract withheld at the request of the investigator.

Microbiome Aspects of Necrotizing Enterocolitis in Premature Infants

Principal Investigator: W. Allan Walker, MD, Massachusetts General Hospital

Co-Investigator(s): Katherine E. Gregory, PhD, RN, Boston College/Brigham and Women's Hospital
Linda J. Van Marter, MD, MPH, Children's Hospital Boston/Brigham and Women's Hospital
Bruce Paster, MD, Harvard School of Dental Medicine/The Forsyth Institute

Necrotizing enterocolitis (NEC), an inflammatory gastrointestinal disease, is among the most significant complications of prematurity. It is both life-threatening and associated with significant long-term morbidity. Research on NEC has explored the inflammatory cascade and factors related to enteral feeding, yet, the pathogenesis remains poorly understood. Defining the intestinal microbiota in premature infants who are at risk for NEC is an innovative approach that will lead to a better understanding of the pathogenesis of the disease and thus, an improved ability to develop disease prediction strategies and clinical interventions. The premature gut is poorly colonized with protective bacteria at birth and thus, is vulnerable to disease. Disease risk is amplified by the routine administration of antibiotics. In sum, NEC may evolve because the protective effect of colonizing bacteria is absent, resulting in infants who are susceptible to gut pathogens and prone to acquire pathogens in the hospital environment. The project aims include: (i) conduct feasibility of microbiome analyses using fecal matter obtained from premature infants and (ii) study differences in the microbiome of premature infants who develop NEC from those who do not develop NEC. The central hypothesis is that the fecal elimination of specific human microbiota will differentiate a population of premature infants who develop NEC from those who do not develop this devastating gastrointestinal disease. The use of stool for microbiome analysis has the advantage of a noninvasive approach that will not deplete the premature infant of a limited blood volume. A multidisciplinary team working across Harvard Catalyst affiliates will achieve the aims.

Posture-Dependent Imaging of Ventilation in an Open-Access MRI Scanner

Principal Investigator: Ronald L. Walsworth, PhD, Faculty of Arts and Sciences, Harvard University

Co-Investigator(s): José G Venegas, PhD, Massachusetts General Hospital
Bruce R. Rosen, MD, PhD, Massachusetts General Hospital
Ross W. Mair, PhD, Faculty of Arts and Sciences, Harvard University
Matthew S. Rosen, Faculty of Arts and Sciences, Harvard University
Rachel N. Scheidegger, Massachusetts Institute of Technology/Faculty of Arts and Sciences, Harvard University
Dan Chonde, Faculty of Arts and Sciences, Harvard University

The effects of body posture on the regional distribution of pulmonary ventilation and perfusion in both healthy subjects and asthmatics has been a topic of considerable recent study. Positron emission tomography (PET) has been used to directly assess the topographical distributions of pulmonary ventilation and perfusion. Magnetic resonance imaging (MRI) of inhaled hyperpolarized He gas provides exquisite anatomical detail with the significant benefit of not employing ionizing radiation. However, no study has quantified the effect of body posture on the topographical location of ventilation defects or regions of perfusion heterogeneity. Importantly, conventional PET and MRI scanners preclude studies when the subject is upright, despite the known significant variation in lung function when upright compared to horizontal. The central objective of this study is to measure, for the first time, the effect of postural changes (upright vs horizontal) on lung ventilation in a small population of healthy subjects. This is a crucial step towards carrying out systematic studies of the effect of posture in asthmatics, with the potential for significant impact on the treatment of those with chronic lung diseases, the obese, and children. We have designed and built a first-of-its-kind open-access human-research MRI scanner for this purpose. This scanner permits subjects' lungs and lung function to be imaged after inhaling hyperpolarized He gas, while sitting, reclining, or lying horizontally. To enable this research program to be conducted effectively, the imager will be relocated from the Center for Astrophysics to the MGH Martinos Center for Biomedical Imaging.

Pharmacogenetics of Asthma Treatment in a Generalizable Population

Principal Investigator: Ann Wu, MD, MPH, Harvard Medical School/Department of Ambulatory Care and Prevention

Co-Investigator(s): Kelan Tantisira, MD, MPH, Brigham and Women's Hospital
Scott Weiss, MD, MS, Brigham and Women's Hospital
Robert Davis, MD, MPH, Kaiser Permanente Georgia and Emory University
Jianjun Wu, MD, PhD, Harvard Medical School/ Department of Ambulatory Care and Prevention

Over 3 billion dollars have been spent on the Human Genome Project, but few applications of the genetic knowledge from this endeavor have been introduced into clinical practice. This grant offers research that will bridge the gap between basic genetic research and actual health benefits for patients. Genetic tests currently in early development hold promise to greatly improve the management of asthma, the most common chronic disease of children. Approximately 20% of children with asthma do not respond to inhaled corticosteroids, the most commonly used controller medication for asthma. We have developed a new pharmacogenetic test that we believe will help clinicians identify such patients and individually tailor their management. The critical next step in translating this research product into practice is to carry out the research to validate this test's clinical utility in large, population-based settings. The goal of this study is to (i) determine the feasibility of obtaining specimens from patients who belong to a large multi-specialty medical group for the purposes of genetic research, and then (ii) evaluate the effectiveness of our pharmacogenetic test that predicts whether an asthma patient will respond to inhaled corticosteroids in this generalizable population.

Identification and Functional Assessment of HIV-1 Cellular Reservoirs on a Single Cell Level

Principal Investigator: Xu Yu, MD, MSc, Massachusetts General Hospital

Co-Investigator(s): J. Christopher Love, PhD, Massachusetts Institute of Technology/The Broad Institute
Yuan Yong, Massachusetts Institute of Technology

HIV-1 can cause a latent or a lytic infection of host cells. While lytically infected cells actively produce HIV-1 particles and thus contribute to the extent of HIV-1 viremia, latently infected cells provide a silent reservoir for HIV-1 that is protected against currently available antiretroviral drugs and HIV-1-specific immune responses. This reservoir therefore represents the major barrier to eliminating the infection. Despite the critical relevance of this latently infected cell population, there are currently no technologies available that allow for the identification and enumeration of these cells. In addition, no acceptable technological solution exists for the identification and isolation of lytically HIV-1 infected cells. Therefore, it is currently entirely unclear how many latently or lytically infected cells circulate in a given infected individual, and what immunophenotypic or functional genetic such cells have. The PIs here propose to combine resources from the Broad Institute (MIT/Harvard) and the AIDS Research Center at MGH to use a novel chip-based microengraving assay to isolate individual cells from HIV-1 infected persons into microwells and subsequently test their latent or lytic infection status on an individual cell level, using ultrasensitive single cell/well detection techniques. This novel, collaborative experimental approach will allow specifically identifying, isolating and characterizing latently infected cells, and will thus be important for addressing a host of questions in HIV-1 pathogenesis that remain unanswered so far and for developing targeted treatment strategies to eliminate this critical population of latently infected cells and thus eliminating HIV-1 entirely.

The Role of Endothelial to Mesenchymal Transition (EndMT) in the Congenital Hypoplastic Left Heart Syndrome (HLHS)

Principal Investigator: Elisabeth Zeisberg, MD, Beth Israel Deaconess Medical Center

Co-Investigator(s): Ivan Melnychenko, MD, Children's Hospital Boston

Hypoplastic left heart syndrome (HLHS) is the leading cause of death among all forms of congenital heart disease in babies. It is characterized by abnormal development of the left-sided cardiac structures, associated with severe endocardial fibrosis, obstructed blood flow from the left ventricular outflow tract and arrested growth of the left heart. Little is known about the pathogenesis of HLHS in which endocardial fibrosis is a key feature. The main mediators of fibrosis are fibroblasts but their origin has never been studied in the setting of HLHS. Endothelial to Mesenchymal Transition (EndMT) is a new mechanism of fibroblast recruitment in chronic heart disease, which was first described by Dr. Elisabeth Zeisberg. EndMT also occurs during heart development, when the septa and valves are formed from the endothelial cells of the endocardium. Dr. Melnychenko has developed an animal model to mimic the pathologies, including endocardial fibrosis, involved in HLHS by performing heterotopic transplantation of neonatal rat hearts into adult rats. We propose to test our hypothesis, that endocardial fibrosis in HLHS is caused by endothelial cells of the endocardium undergoing EndMT, by utilizing Dr. Melnychenko's animal model in transgenic mice in which cells of endocardial origin are irreversibly tagged with YFP (Aim 1). We further propose to test in the animal model, if inhibition of EndMT by BMP-7, a molecule shown to prevent EndMT in chronic heart disease, inhibits endocardial fibrosis as it occurs in HLHS (Aim 2). Lastly, we plan to analyze cardiac tissue from children with HLHS with respect to EndMT and to the BMP-7 receptor, to test the potential of targeting EndMT as a new therapeutic strategy for HLHS (Aim 3).