Biomedical Collaborative Pilot Grants
Cycle 3

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 3 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, 61 grants were awarded of up to $50,000 per funded proposal to support a one-year pilot research project.

Funding decisions for the third cycle of Biomedical Collaborative Pilot Grants were announced in June 2010.

For more information about these awards, see the following news article.

Experimental Approach to Genotype-Phenotype Relationships in CADASIL Pathogenesis

Principal Investigator: Cenk Ayata, Massachusetts General Hospital

Co-Investigator(s): Spyros Artavanis-Tsakonas, Harvard Medical School

Cerebral Autosomal Dominant Arteriopathy, Subcortical Infarcts, and Leukoencephalopathy syndrome (CADASIL), the most common heritable cause of stroke and vascular dementia in young adults, is a microvasculopathy linked to mutations in the *NOTCH3* gene. In adult brain, *NOTCH3* is expressed exclusively by vascular smooth muscle. Age-dependent smooth muscle degeneration and granular osmiophilic material accumulation are characteristic in CADASIL, leading to strokes, leukoaraiosis and dementia; however, their mechanisms are poorly understood. Specifically, it is not known whether CADASIL mutations cause loss-of-function or pathological gain-of-function. We recently showed that *Notch3* knockout worsens stroke outcome via cerebrovascular mechanisms, and now have a unique opportunity to define the cerebrovascular dysfunction and stroke phenotype by employing novel mouse models expressing Notch3 R1031C or C455R mutations found in CADASIL patients. While the former mutation is clinically associated with classical CADASIL, the latter presents with very early onset strokes and severe leukoaraiosis, thus allowing genotype-phenotype correlation in an experimental setting. We will test whether CADASIL mutations: (1) disrupt cerebrovascular function: Using sophisticated optical imaging tools, we will investigate neurovascular coupling and cerebral blood flow autoregulation; (2) worsen stroke outcome: Established models will be employed to assess neurological function and infarct volume after stroke. Expressing these mutations on a *Notch3* knockout background will further assess whether mutant proteins rescue/worsen the *Notch3* null phenotype, specifically addressing the loss- or gain-of-function. By doing these, we aim to define the broad outlines of novel genotype-phenotype relationships in CADASIL, and build the foundations for future mechanistic studies in search of therapeutic targets.

An Interdisciplinary Approach to Developing and Testing Evidence-based Mental Health Interventions for War Affected Youth in Sierra Leone

Principal Investigator: Theresa Betancourt, Harvard School of Public Health

Co-Investigator(s): Adeyinka Akinsulure-Smith, The City College of New York
Nathan Hansen, Yale University School of Medicine
John Weisz, Judge Baker Children's Center

Of the many dangers of war, abduction and child soldiering are considered particularly pernicious to child mental health and development. In Sierra Leone, where children and youth were widely involved during a decade of conflict, the long term psychosocial adjustment and social reintegration of former child soldiers is of great concern. From 2002-2008, Dr. Betancourt (PI) conducted the first ever prospective longitudinal study of male and female former child soldiers. Our analyses of factors contributing to risk and resilience indicate that community-based interventions promoting social support, healthy coping, and positive community engagement are critical for assisting war-affected youth. However, little research has been conducted to identify, develop or validate interventions that are effective for this population.

We propose an interdisciplinary collaboration incorporating best practices and evidence from existing intervention research targeting violence-affected youth. With pilot funding from the Harvard Catalyst Program we aim to:1) Review, select and culturally adapt evidence-based modules from trauma-focused interventions for use with youth in Sierra Leone; 2) Develop a manualized intervention to provide psychosocial support and culturally appropriate mental health services; 3) Assess the feasibility, efficacy and acceptability of the adapted intervention.

The results of this study will inform the development of a future NIH R01 application to implement a randomized controlled trial of the adapted intervention. Ultimately, this intervention would not only aim to respond to the current need in Sierra Leone, but would also contribute to the evidence base on mental health interventions to assist young people exposed to war violence.

Identification of Variants Responsible for Bicuspid Aortic Valve Disease

Principal Investigator: Simon Body, Brigham and Women's Hospital

Co-Investigator(s): Eric Isselbacher, Massachusetts General Hospital
Robert Levine, Massachusetts General Hospital
Christine Seidman, Brigham and Women's Hospital
Jon Seidman, Harvard Medical School
Susan Slaugenhaupt, Massachusetts General Hospital

1% of Americans are born with a bicuspid aortic valve (BAV), with consequent risk of aortic stenosis. Patients with BAV have a markedly increased risk of thoracic aortic dissection and aneurysm, independent of the severity of BAV and aortic stenosis. 30% of individuals undergo surgical repair of BAV or thoracic aortic disease.

The genetic etiology is characterized by heritability estimate of 89% and 6-10 fold increased risk in first degree relatives. However, much of the disease is sporadic, without characteristic Mendelian inheritance. *NOTCH1* variants have been associated with BAV in three family kindreds but further replicated association of other causal variants have not been made in other kindreds or in those with sporadic disease.

We have currently collected DNA and medical history from 155 patients who responded to a mailing to a pilot cohort of 280 patients who had previously undergone aortic valve replacement (AVR) at BWH. By February 21st, we will have mailed an additional 492 BAV patients who have previously undergone AVR at BWH and 502 BAV patients who have previously undergone AVR at MGH. From these 1274 patients, based upon current returns from the pilot cohort, we anticipate receiving DNA and medical histories from at least 500 BAV patients.

This study aims to:
1. Perform whole exome sequencing (WES) in 10 individuals with sporadic BAV.
2. Identify genes that overexpress variants in BAV patients that are rare or absent in reference non-BAV genomes,
3. Sequence identified genes in an additional 46 patients to identify variation specific to BAV disease.
4. Genotype BAV-associated variants in acohort of 500 patients with BAV and population-based controls.
5. Identify variants that are over-expressed in patients with BAV.

This study may potentially indicate methodologies to establish the biologic mechanisms involved in BAV. It may also identify established drugs or drug targets that modify the pathways in BAV that might be valuable in modifying the occurrence of thoracic aortic aneurysm and dissection.

Adenosine Induced Stress-rest CT in Patients at High Risk of Coronary Artery Disease

Principal Investigator: Thomas Brady, Massachusetts General Hospital

Co-Investigator(s): Brian Ghoshhajra, Massachusetts General Hospital
Udo Hoffmann, Massachusetts General Hospital
Raymond Kwong, Brigham and Women's Hospital
Ahmed Tawakol, Massachusetts General Hospital

Recent studies demonstrate that benefits of coronary revascularization are limited to patients with coronary stenosis who demonstrate impaired myocardial perfusion. However, conventional stress testing has limited ability to accurately identify patients with hemodynamically significant CAD. In addition, none of these non-invasive techniques provides direct information regarding coronary anatomy necessitating referral to invasive coronary arteriography. Hence, a single non-invasive technique that provides accurate data on coronary stenosis and myocardial perfusion is required.

Cardiac CT can rule out the presence of significant CAD, however the ability to detect hemodynamically significant CAD is reduced by the presence of coronary calcification in high risk patients. We recently demonstrated that CT can identify adenosine stress-induced myocardial perfusion defects in patients with accuracy comparable to nuclear myocardial perfusion imaging (MPI). When combined with coronary stenosis assessment, stress CT had a diagnostic accuracy greater than nuclear MPI.

The proposed pilot study seeks to validate myocardial perfusion and delayed enhancement imaging using a novel low radiation (~3 mSv), dual energy CT system. Stress CT with delayed enhancement will be obtained in 15 patients at high risk or with known CAD and compared to 3T cardiac MR data, the clinical standard for myocardial perfusion and delayed enhancement imaging. This research represents a new collaboration between investigators at MGH and BWH. If successful, this research should lead to several NIH grant proposals including a prospective randomized multicenter clinical trial to assess the ability of a comprehensive cardiac CT exam to optimally manage patients at high risk and/or with known CAD.

Biomarkers of Cardiac Allograft Vasculopathy

Principal Investigator: David Briscoe, Children's Hospital Boston

Co-Investigator(s): Elizabeth Blume, Children's Hospital Boston
Kevin Daly, Children's Hospital Boston
Michael Givertz, Brigham and Women's Hospital
S. Ananth Karumanchi, Beth Israel Deaconess Medical Center
Alan Packard, Children's Hospital Boston

Allograft rejection characteristically involves cellular and humoral immune responses against the graft. The major cause of late mortality after cardiac transplantation is cardiac allograft vasculopathy (CAV). The diagnosis of CAV relies on expensive and invasive tests, such as coronary angiography and intravascular ultrasound, which are most sensitive in advanced disease.

Biomarker analysis has focused on the assessment of T-cell activation and regulatory responses. However, the alloimmune activation that characteristically occurs following transplantation is not always associated with development of chronic rejection/CAV. Graft endothelial cells (EC) are the primary targets of the allogeneic response, and EC injury/repair occurs in association with rejection. We plan to test the hypothesis that the earliest biomarker of CAV is an EC based response.

We propose: (1) To determine whether the assessment of arrays of angiogenic factors are predictive of the development of CAV in a cohort of over 100 patients, (2) To determine if PET scan could serve as a sensitive test for early EC injury in a murine model of chronic rejection. In support of our planned study, we performed pilot arrays on 18 patients (9/9 with/without CAV). Of 55 angiogenesis related molecules studied, multivariate logistic regression indicated that 6 are highly predictive and independently associated with CAV (all $P<0.01$). Our objective is to determine if biomarkers of endothelial injury/repair and PET scan can be used as sensitive, non-invasive markers of CAV. In the future, we plan to employ this approach to develop early therapeutic interventions for the treatment of CAV.

Kisspeptin as an *In Vivo* Probe of GnRH Neuronal Function: Application to the Evaluation of Delayed Puberty

Principal Investigator: Yee-Ming Chan, Children's Hospital Boston and Massachusetts General Hospital

Co-Investigator(s): Stephanie Seminara, Massachusetts General Hospital

Human sexual maturation is triggered by increasing secretion of the master reproductive hormone GnRH at puberty. Signaling by the neuropeptide kisspeptin has recently been found to be essential for normal sexual maturation. We have demonstrated that a single dose of kisspeptin potently stimulates GnRH-induced LH release in healthy adult volunteers (IND 74,877, ClinicalTrials.gov NCT00914823). This and other data indicates that kisspeptin resides directly upstream of GnRH in the reproductive endocrine cascade.

We propose to apply kisspeptin administration to the evaluation of adolescents with delayed puberty. Most adolescents with delayed puberty have constitutional delay of puberty (CDP), in which puberty occurs late but otherwise normally. In contrast, some adolescents with delayed puberty have a permanent defect in GnRH release or action. To date, there has been no method to assess GnRH neuronal function *in vivo*, and so it is difficult to distinguish CDP from GnRH deficiency on initial presentation; typically, the definitive diagnosis can only be made retrospectively. Thus, additional tools are needed for the evaluation of delayed puberty.

Because kisspeptin directly and potently stimulates GnRH release, we will use kisspeptin as the first-available probe of GnRH neuronal function. Adolescents who exhibit an impaired response to kisspeptin are likely to have permanent GnRH deficiency, whereas adolescents who exhibit a normal response to kisspeptin are likely to have CDP. Prospective identification of adolescents with permanent GnRH deficiency will avoid delays in definitive treatment and prevent the psychosocial distress and risk for low bone mineral density associated with untreated GnRH deficiency.

Longitudinal Investigation of Pathological Changes in a Mouse Model of Ocular Hypertension (Glaucoma) by *In Vivo* Retinal Imaging

Principal Investigator: Dong Chen, Schepens Eye Research Institute

Co-Investigator(s): Clemens Alt, Massachusetts General Hospital
Dean Cestari, Massachusetts Eye and Ear Infirmary
Kin-Sang Cho, Schepens Eye Research Institute
Charles Lin, Massachusetts General Hospital
Nadja Tajouri, Massachusetts Eye and Ear Infirmary

Glaucoma is a neurodegenerative disease of retinal ganglion cells causing vision loss and eventually blindness. While the underlying mechanism of such disease remains unclear, elevation of intraocular pressure (IOP) is a well-known risk factor. Several studies have suggested that mechanical injury and retinal ischemia-and-perfusion injury may participate in the degeneration of retinal ganglion cells in glaucoma. However, the retinal changes of glaucomatous or ocular hypertension eye have to date been examined mainly by histological examination. Investigating the time course of cellular responses to ocular hypertension may provide important information that can be helpful in developing strategies for protecting retinal ganglion cells from degeneration.

Our group has developed a novel ocular hypertension model where microbeads are injected into the anterior chamber of the mouse eye. Following the microbeads blocking Schlemm's canal, an effluent of aqueous humor in the anterior chamber, IOP is elevated, resulting in degeneration of retinal ganglion cells and their axons. Here, we propose a collaborative effort to investigate retinal changes in ocular hypertension *in vivo*, using an adaptive-optics scanning laser ophthalmoscope that was developed specifically for high resolution (2-3 μm) mouse retinal imaging by Dr. Charles Lin's laboratory. It is capable of optically resolving axons and retinal ganglion cells with their dendrites. The goal of this study is to gain a detailed understanding of the temporal and spatial changes of retinal ganglion cell morphology and retinal vasculature in response to ocular hypertension. The results will aid design a strategy for protection of retinal ganglion cells.

Phosphatidylcholine Transfer Protein Inhibitors for the Management of Type 2 Diabetes

Principal Investigator: David Cohen, Brigham and Women's Hospital

Co-Investigator(s): Gregory Cuny, Brigham and Women's Hospital

Phosphatidylcholine transfer protein (PC-TP, a.k.a. StARD2) binds phosphatidylcholines and catalyzes their intermembrane transfer and exchange *in vitro*. The structure of PC-TP comprises a hydrophobic pocket and a well-defined head-group binding site. Our studies have revealed key regulatory roles for PC-TP in lipid and glucose metabolism. Notably, *Pctp-/-* mice are sensitized to insulin action, exhibit more efficient brown fat-mediated thermogenesis and are protected against atherosclerosis. Considering the therapeutic potential of targeting PC-TP, we undertook a high throughput screen to identify small molecule inhibitors. We utilized a fluorescence quench assay to measure phosphatidylcholine transfer activity and screen 114,752 compounds. This exercise identified 6 inhibitors of PC-TP activity with IC50 values that ranged from 4.1 – 95.0 μM under conditions of the *in vitro* assay. Extensive structure activity studies have identified active moieties for two of the most potent inhibitors. Testing in cell culture systems has revealed their capacity to enhance insulin signaling at concentrations as low as 50 nM. In an interdepartmental collaboration, the current proposal seeks to test PC-TP inhibitors for activity *in vivo*. Compounds will be tested for their influence on lipid and glucose metabolism in mice. Their administration to *Pctp-/-* mice will insure that the observed effects are due to inhibition of PC-TP. The compounds will also be tested in common diabetic mouse models. It is anticipated that PC-TP inhibitors may ultimately prove to be of value in the management of type 2 diabetes and atherosclerotic cardiovascular diseases.

Engineering Resistance to Epileptic Seizures by Metabolic Regulation

Principal Investigator: Nika Danial, Dana-Farber Cancer Institute

Co-Investigator(s): Gary Yellen, Harvard Medical School

Abstract withheld at the request of the investigator.

The Use of Positron Emission Tomography–computed Tomography (PET–CT) to Visualize the Substrate of Sudden Cardiac Death

Principal Investigator: Stephan Danik, Massachusetts General Hospital

Co-Investigator(s): Barrett Conor, Massachusetts General Hospital
Sanjeev Francis, Massachusetts General Hospital
Quynh Truong, Massachusetts General Hospital

The composition of tissue responsible for ventricular tachycardia (VT) in patients with structural heart disease is due to viable myocardium interspersed with scar and fibrotic tissue. Presently, because this tissue cannot be reliably identified in patients at risk for sudden cardiac death, most individuals with an ejection fraction of less than 35% receive prophylactic implantation of an implantable cardioverter defibrillator (ICD). In addition, many patients in whom ICDs have successfully terminated VT require catheter ablation to prevent recurrent discharges. While preliminary data suggests that cardiac magnetic resonance imaging has the potential to achieve this endpoint, most patients who are at risk for sudden cardiac death already have an ICD implanted that preclude its use. Using a porcine model of chronic healed myocardial infarction, electroanatomical mapping (EAM) will be performed to electrically characterize the scar and surrounding border zone that contain the electrical surrogates of ventricular tachycardia. Positron emission tomography–computed tomography (PET–CT) will then be performed, and the images will be integrated with the voltage map to create a three dimensional reconstruction of the heart. Histopathological analysis of each of the hearts will be performed to confirm the identification of normal, viable, and infarcted myocardium initially identified by PET-CT and EAM. The goal of this project is to characterize the electrical characteristics of tissue that contains the substrate for ventricular tacharrhythmias which could then be used in patients to aid during catheter ablation of VT as well as to help stratify patients at risk for sudden cardiac death.

Conformations of Phosphorylated Tau as a Novel Biomarker of Alzheimer's Disease

Principal Investigator: Jane Driver, Brigham and Women's Hospital and Veterans Affairs Boston Healthcare System

Co-Investigator(s): Kun Ping Lu, Beth Israel Deaconess Medical Center

We propose to demonstrate that the ratio between different conformations of phosphorylated tau in cerebrospinal fluid (CSF) is a novel and early biomarker for Alzheimer's disease (AD). Tau phosphorylated at T231 (pT231-tau) is a promising biomarker, but levels are quite variable and tend to decline as AD progresses. We have discovered that pT231-tau exists in two conformations, cis- and trans-, and that conversion between these isomers is regulated by the unique enzyme Pin1. We have developed an innovative technology to generate the first antibodies that can distinguish between these conformations. In a mouse model of AD, only cis but not trans pT231-Tau dramatically accumulates, and this is effectively reversed by Pin1 over-expression. In human brains, only cis pT231-tau is elevated in mild cognitive impairment and further accumulates with disease progression. Moreover, the cis/trans ratio of pT231-Tau in CSF was elevated in advanced AD with little inter-individual variation. We thus hypothesize that the cis/trans ratio of pT231-tau may be a novel biomarker for early detection and progression of AD. To test this hypothesis, we will quantify cis and trans pT231-tau in the ventricular CSF of patients with autopsy proven AD in various Braak stages from a brain bank of nearly 300 individuals available from our collaborator Dr. Neil Kowall. We will determine the sensitivity and specificity of the CSF cis/trans ratio of pT231-Tau for predicting the diagnosis of AD by comparing patients with AD to those with other types of dementia and no dementia.

Fibroblast Growth Factor 21 as a Biomarker for Fatty Liver Disease

Principal Investigator: Jody Dushay, Beth Israel Deaconess Medical Center

Co-Investigator(s): Nezam Afdhal, Beth Israel Deaconess Medical Center
Michelle Lai, Beth Israel Deaconess Medical Center
Robert Lenkinski, Beth Israel Deaconess Medical Center
Eleftheria Maratos-Flier, Beth Israel Deaconess Medical Center

Nonalcoholic fatty liver disease (NAFLD) is a leading cause of chronic liver disease. A subset of individuals with NAFLD progress to nonalcoholic steatohepatitis (NASH), which increases risk for cirrhosis and carcinoma. It is important to monitor patients closely for progression to NASH. Repeated liver biopsies pose risk, therefore monitoring typically includes transaminase levels and/or ultrasound, both of which lack sensitivity and specificity. There is currently no biomarker for NAFLD.

Fibroblast growth factor 21 (FGF21), a member of the FGF superfamily, is expressed predominantly in the liver and secreted into the circulation. In rodents and humans, FGF21 is elevated in diabetes, obesity, and insulin resistance. The Maratos-Flier lab has shown that FGF21 plays a key role in lipid oxidation in rodents. FGF21 levels increase significantly with fasting and ketogenic diet and decrease with refeeding in mice. Administration of FGF21 to obese, diabetic rodents and nonhuman primates improves glycemia and causes weight loss.

We recently discovered that serum levels and hepatic mRNA expression of FGF21 are significantly increased in NAFLD. We did not find expression in human adipose tissue, suggesting that serum elevation is due to increased hepatic expression. In lean humans FGF21 does not appear to be nutritionally regulated. Based on these findings, we believe FGF21 may be a biomarker for fatty liver.

This pilot study will define the relationship between serum and hepatic FGF21 levels in obese individuals with and without NAFLD. We will also use NMR spectroscopy to confirm that FGF21 is a biomarker of hepatic lipid accumulation.

Injectable Matrix-embedded Endothelial Cells for Vascular Therapy

Principal Investigator: Elazer Edelman, Brigham and Women's Hospital

Co-Investigator(s): Eytan Abraham, Brigham and Women's Hospital
Natalie Artzi, Massachusetts Institute of Technology
Boaz Mizrahi, Children's Hospital Boston
Charles Vacanti, Brigham and Women's Hospital

Vascular disease is the leading cause of mortality in the USA. To facilitate revascularization of stenotic vessles, over 1 million angioplasty procedures are performed annually in the USA. The most significant limitation of angioplasty remains the high rates of restenosis post procedure, as well as denudation of the endothelial cells (EC) layer, which impedes vascular homeostasis. Restenosis after angioplasty or stent implantation occurs in approximately 50% of treated vessels. We propose to investigate the use of a novel minimally invasive injectable matrix embedded endothelial cell (MEEC) platform to minimize restenosis and to restore the EC layer and function post angioplasty.

Endothelial cells (EC), the major regulatory cells of the blood vessel, have emerged as a key component of the vascular response to injury. We postulate that supplementing the adventitial endothelium with MEEC may provide control over the response to vascular injury. Indeed, open surgical placement of perivascular matrix embedded allogenic endothelial cells has been shown to inhibit intimal thickening, stenosis and to facilitate EC layer restoration. While open field surgery is amenable to direct adventitial placement of matrix embedded EC, the ability to apply MEEC by using a minimally invasive injectable formula, will enable the utilization of the MEEC platform for post angioplasty procedure treatment.

Treatment Resistant Geriatric Depression in Primary Care: Is NAAG (N-Acetylaspartyl-glutamate), Measured by Proton Magnetic Resonance Spectroscopy (1H-MRS) at 4 Tesla, a Predictor of Treatment Response?

Principal Investigator: Brent Forester, McLean Hospital

Co-Investigator(s): Bruce Cohen, McLean Hospital
Anne Fabiny, Cambridge Health Alliance
Eric Jensen, McLean Hospital
Caitlin Ravichandran, McLean Hospital

Geriatric Depression is associated with significant morbidity and mortality and reduced social, physical and cognitive functioning. Although depression is often recognized and treated in the primary care setting, accurate diagnosis and adequate treatment, especially in those who remain treatment non-responsive after an initial medication trial, remains challenging. Furthermore, current treatments are limited by the lack of biomarkers to assess new therapies. Magnetic resonance spectroscopy (MRS) offers a noninvasive method for investigating metabolic and neurotransmitter changes associated with geriatric depression. Previous proton (1H) MRS studies indicate that glutamate levels are abnormally high in the cerebral cortex in patients with depression. Medications that selectively reduce glutamatergic neurotransmission have antidepressant activity in animal models and in depressed patients. N-Acetylaspartylglutamate (NAAG) blocks presynaptic glutamate release and postsynaptic glutamate transmission. Reduced NAAG levels may, therefore, be associated with excessive glutamatergic activity and serve as a marker for depression and treatment non-response. Memantine is an antagonist at the NMDA glutamate receptor and has demonstrated antidepressant effects in open label studies of younger adults. We propose to measure and determine whether NAAG levels predict response to treatment with Memantine in older adults with depression who are refractory to current therapeutic interventions. This study may provide guidance for novel therapeutic interventions based on glutamatergic abnormalities in treatment resistant geriatric depression. Funding will enable a collaboration between a tertiary care geriatric psychiatry program and Neuroimaging Center at McLean Hospital and a primary care geriatrician at the Cambridge Health Alliance serving an aging community population with significant rates of depression.

Neural Pre-markers of Developmental Dyslexia in Infants with a Family History of Developmental Dyslexia

Principal Investigator: Nadine Gaab, Children's Hospital Boston

Co-Investigator(s): Ellen Grant, Children's Hospital Boston

Developmental dyslexia (DD) is the most prominent specific learning disabilities in school-age children with a strong genetic basis. Neuroimaging studies have revealed functional and structural differences within various brain regions in school-age children and adults with DD. Our proposed study will investigate whether the observed differences in structural neuroanatomy (within regional gray matter and white matter tracks) can already be observed in 40 infants with compared to without a family history of DD. We aim to acquire structural T1 brain images using magnetic resonance imaging as well as and diffusion tensor imaging in infants (age 5-9 months; 20 with and 20 without a family history of DD). Children will be recruited through already established contacts within the greater Boston community (e.g.; Alumni of the local dyslexia schools) and the Developmental Medicine Center at Children's Hospital Boston. Differences in whole-brain and regional gray matter indices as well as white matter tracts between children with compared to without a family history of DD will be examined. Identifying possible early neural pre-markers of developmental dyslexia will be essential for the development and implementation of early remediation programs. Furthermore, will help to evaluate and improve early identification tools for children at risk and will lead to the implementation of preventive strategies and the refinement of diagnostic criteria. Furthermore, it may diminish the clinical, emotional and social impact of DD.

Effect on Acetaminophen Metabolism by Liquid Formulations: Do Excipients in Liquid Formulation Prevent Production of Toxic Metabolites?

Principal Investigator: Michael Ganetsky, Beth Israel Deaconess Medical Center

Co-Investigator(s): Barbara LeDeuc, Massachusetts College of Pharmacy and Health Sciences
Mark Bohlke, Massachusetts College of Pharmacy and Health Sciences
Robert Lipton, Beth Israel Deaconess Medical Center
Steven Salhanick, Beth Israel Deaconess Medical Center
David Williams, Massachusetts College of Pharmacy and Health Sciences

Acetaminophen (APAP) poisoning is the most frequent cause of acute hepatic failure in the United States. Toxicity requires cytochrome P-450 bioactivation of APAP. Children are less susceptible to APAP toxicity; the current theory is that they have different metabolism than adults. However, children's liquid preparations of APAP contain excipients which have been shown to inhibit APAP bioactivation *in vitro* and in rodents. Children tend to ingest liquid preparations, which could potentially explain their decreased susceptibility instead of an intrinsically different metabolism. Further, our review of Poison Center epidemiologic data shows that liquid preparations are less toxic in adults. Our hypothesis is that excipients in liquid preparations inhibit the bioactivation of APAP. The design is a pharmacokinetic cross-over study in humans. Healthy adult subjects will be recruited for administration of therapeutic doses of APAP in capsule and liquid formulations. Plasma via a heplock will be collected at serial time points up to 8 hours and assayed for APAP and its metabolites. After a washout period, subjects will receive the same dose of APAP in the alternate preparation. The pattern of metabolites, indicating the activity of the bioactivating enzymes, will be compared. A significant difference in P-450 metabolites will support the hypothesis and provide preliminary data for studies in patients who have ingested potentially toxic doses of APAP. Ultimately, this work could support development of novel antidotal therapy for APAP overdose. This project will support ongoing collaborative efforts between Medical Toxicologists at BIDMC and scientists at the MCPHS Division of Pharmaceutical Sciences.

Novel Approach to Pulmonary Vein Ablation using Transcervical Flexible Endoscopy

Principal Investigator: Denise Gee, Massachusetts General Hospital

Co-Investigator(s): William Brugge, Massachusetts General Hospital
David Milan, Massachusetts General Hospital
Christopher Morse, Massachusetts General Hospital
David Rattner, Massachusetts General Hospital
Brian Turner, Massachusetts General Hospital

Atrial fibrillation (AF) is the most common arrhythmia in the US. Antiarrhythmic drugs are often first-line therapy but effectiveness varies. Surgical approaches are time-consuming and invasive. Catheter-based techniques use energy sources to create circumferential lesions around the pulmonary vein (PV)–atrial junction. Unfortunately, there is a >5% complication rate and lengthy fluoroscopic exposure times are required. Thermal energy also increases the risk of local complications due to overheating, tissue coagulation, and variable temperature distribution in treated tissue. Irreversible electroporation (IRE) is a modality in which microsecond electrical pulses are applied to generate a destabilizing electric potential causing nanoscale defects in the lipid bilayer. Permanent nonthermal transmural damage is produced within fractions of a second. Given the increasing role for flexible endoscopy in the field of surgery and our experience in transesophageal Natural Orifice Translumenal Endoscopic Surgery (NOTES), we propose a study to explore PV ablation using a) transcervical flexible endoscopy through a small cervical incision with standard energy modalities and b) IRE—a new modality that might overcome limitations inherent in existing thermal energy sources. Transcervical PV ablation will allow access to the posterior mediastinal compartment and direct visualization of the PVs. It promises to be potentially faster and less morbid than other surgical or catheter-based procedures. Prototype endoscopic catheters that can deliver radiofrequency (standard) and IRE (experimental) ablation will be used to compare these two modalities. The success of transcervical PV ablation or of IRE would be a groundbreaking discovery that could change the face of AF therapy.

Identification of Novel NF-κB Regulators in Lymphomagenesis

Principal Investigator: Benjamin Gewurz, Brigham and Women's Hospital

Co-Investigator(s): Elliott Kieff, Brigham and Women's Hospital
Margaret Shipp, Dana-Farber Cancer Institute

Nuclear Factor kappa B (NF-κB) is a family of transcription factors at the crossroads of innate and acquired immunity, allergy, and oncogenesis. Immune receptors differentially activate NF-κB through 'canonical' or 'non-canonical' signal transduction pathways. Viruses and cancer cells exploit NF-κB to drive cell proliferation and inhibit apoptosis. Epstein Barr Virus (EBV)encoded Latent Membrane Protein 1 (LMP1) uses two cytoplasmic domains to constitutively activate non-canonical and canonical NF-κB. LMP1 is expressed in most EBV associated malignancies, mimicking NF-κB hyperactivation states present in non-EBV infected Diffuse or Mediastinal Large B-Cell Lymphomas (DLBCL and MLBCL). DLBCL and MLBCL frequently evolve sustained NF-κB activation through genetic lesions in both canonical and non-canonical NF-κB pathways. Important cellular regulators of NF-κB activation remain to be identified, and RNAi allows the first comprehensive genetic analysis of both mammalian NF-κB pathways. We recently completed a genomewide siRNA screen of canonical NF-κB activation by LMP1. We identified more than 100 validated novel positive (putative oncogene) and negative (putative tumor suppressor) regulators of canonical NF-κB, many of which are enzymes. We will expand the candidate gene pool of NF-κB regulators through a screen of the LMP1/non-canonical NF-κB pathway. We propose to then screen a large collection of DLBCL and MLBCL tumor samples for the presence of genetic lesions or aberrant expression levels in the strongest NF-κB screen hits. This new collaboration will enhance understanding of lymphomagenesis and will likely identify rational diagnostic and therapeutic lymphoma targets.

Development of Novel Anti-tuberculosis Agents that Inhibit Protease ClpP1P2

Principal Investigator: Alfred Goldberg, Harvard Medical School

Co-Investigator(s): Eric Rubin, Harvard School of Public Health

Tuberculosis remains one of the leading causes of death from infectious diseases worldwide. *Mycobacterium tuberculosis* (*Mtb*) has become increasingly resistant to the available antibiotics. Therefore identifying new drug targets, specifically Mtb enzymes that are essential for viability, and developing inhibitors of their functions is an important approach to combat this devastating disease. Prof. Eric Rubin and co-workers have shown by genetic approaches that a proteolytic enzyme, ClpP, is essential for the viability of Mycobacteria and for infection in mice. Since ClpP is not present in the cytoplasm of mammalian cells, where protein breakdown occurs by very different systems, specific inhibitors of ClpP should not affect the functioning of human cells. Therefore, the ClpP protease is a highly attractive target for drug development.

Until now, attempts to isolate a functional mycobacterial ClpP were unsuccessful. Prof. Goldberg's lab has for the first time found conditions to express and isolate the active ClpP enzyme complex and to define its unique properties: *Mtb* ClpP is a two-ring tetradecameric complex composed of 7 ClpP1 and 7 ClpP2 subunits (ClpP1 and ClpP2 are products of two different *Mtb* genes). A breakthrough in developing an enzymatic assay for Mtb ClpP1P2 was a discovery of a group of short peptides that dramatically stimulate its activity against peptide and protein substrates. This discovery has allowed us to find optimal conditions for high throughput assay of this enzyme. Our major goal in seeking support from the Catalyst Program will be to utilize the facilities of the ICCB to screen for small molecule inhibitors that might serve as lead compounds in a drug development program, to screen libraries of inhibitors of related proteases, and together with Dr. Rubin's lab, to explore their possible effects on growth of Mycobacteria.

Molecular Targets in Well-differentiated Liposarcoma

Principal Investigator: Alejandro Gutierrez, Dana-Farber Cancer Institute

Co-Investigator(s): Christopher Fletcher, Brigham and Women's Hospital
Jonathan Fletcher, Brigham and Women's Hospital
A. Thomas Look, Dana-Farber Cancer Institute
Chandrajit Raut, Brigham and Women's Hospital

Well-differentiated liposarcoma (WDLS) is one of the most common human sarcomas, however its molecular pathogenesis remains poorly understood, due largely to the absence of a suitable animal model or cell lines in which to study this disease. We have developed a zebrafish model of WDLS, induced by expression of a constitutively active *Akt2* transgene in mesenchymal progenitors of *p53*-mutant zebrafish, in which WDLS develops at a mean age of 11 weeks and closely resembles the human disease pathologically. Given that oncogenic pathways are well-conserved between zebrafish and humans, the finding that Akt activation induces WDLS in the zebrafish strongly suggests that this pathway is central to human WDLS pathogenesis. Furthermore, this model represents an ideal system in which to functionally characterize the molecular abnormalities underlying WDLS. In this proposal, we will examine primary human WDLS specimens for genetic evidence of PTEN-PI3K-AKT activation by array CGH and sequencing, which would have immediate clinical relevance given that AKT pathway inhibitors are currently in clinical development. We will then use the zebrafish model system to examine the ability of individual genes within the 12q13-15 amplification characteristic of WDLS (MDM2, CDK4, HMGA2, GLI1) to accelerate tumor onset, thus identifying the pathogenic genes driving selection for this amplification, which will represent novel therapeutic targets. These proposed studies will bring together Harvard investigators with genetic, pathologic, and clinical expertise in human liposarcoma together with expert zebrafish cancer biologists, establishing new collaborations that will allow the zebrafish model of WDLS to be harnessed to its fullest potential.

Harnessing Neural Plasticity to Prevent Psychosis

Principal Investigator: Christine Hooker, Faculty of Arts and Sciences, Harvard University

Co-Investigator(s): Matcheri Keshavan, Beth Israel Deaconess Medical Center

The aim of this proposal is to identify structural and functional neural changes as a result of a cognitive remediation intervention in people at risk for developing psychosis. Schizophrenia is a debilitating psychiatric disorder characterized by deficits in cognition and social functioning. Early identification and prevention is imperative. Prior research shows that the neurocognitive deficits that characterize patients with established schizophrenia, also affect people at risk (AR) for schizophrenia, including impairments in memory, attention, and social cognition. Prospective studies show that these neurocognitive deficits in at risk individuals predict symptomotology, functional status and conversion to full blown psychosis. At the present time, it is unknown whether targeted cognitive and social-cognitive training (TCST) as an intervention for people at risk for psychosis could improve functional outcome and/or reduce the risk of conversion. The current proposal will use fMRI and behavioral methods to investigate whether TCST in people at risk for psychosis improves the efficiency of neural mechanisms that support cognitive and social-cognitive skills. This project will bring together the expertise of Drs. Keshavan and Seidman at BIDMC/HMS and Dr. Hooker at Harvard University/FAS. Drs. Keshavan and Seidman are leaders in the identification and treatment of individuals at risk for psychosis. Dr. Hooker is a junior investigator who has expertise in using fMRI to investigate neural changes as a result of cognitive intervention in chronic schizophrenia. This Catalyst grant provides the opportunity to bring together different skill sets to address a major mental health problem.

Improving the Diagnostic Accuracy of Oncologic Positron Emission Tomography (PET) Imaging of the Liver

Principal Investigator: David Israel, Brigham and Women's Hospital

Co-Investigator(s): Georges El Fakhri, Massachusetts General Hospital

Our ultimate goal is to improve the diagnostic accuracy of PET in hepatic tumors. PET is widely used for the diagnosis and staging of cancers. The presence of liver metastases is often a question of critical importance in the care of cancer patients but lesion detection is hindered by the variable and heterogeneous appearance of the liver, which often has high background tracer uptake.

Performance in lesion detection varies widely with the type of scanner and protocol used, body habitus, and image processing. We hypothesize that improved detection accuracy can be achieved if this variability is better quantified and understood.
We propose to develop a methodology for objective assessment of hepatic images to accurately predict the likelihood that a finding is malignant. We will develop objective metrics of image quality to estimate levels of confidence in detected lesions and use them to determine the optimal acquisition and processing scheme with the best detection accuracy.

We will computationally "insert" known lesions into imaging studies obtained from cancer patients, and study the effects of hepatic background characteristics, acquisition protocol, body habitus and image processing on the accuracy of detection of lesions by human and mathematical observers. Our studies will make use of an extensive and unique archive of over 20,000 PET scans and correlative clinical information, including a large number of sequential scans in patients with known hepatic metastases and patients without known liver lesions.
This will provide the preliminary results needed for a proposal to be submitted for NIH-R01 funding.

An Initial Trial of Enteral Fish Oil Supplementation in the Treatment of Intestinal Nutrition-associated Liver Disease in Patients with Intestinal Failure

Principal Investigator: Tom Jaksic, Children's Hospital Boston

Co-Investigator(s): Megan Brenn, Children's Hospital Boston
Christopher Duggan, Children's Hospital Boston
Kathy Gura, Children's Hospital Boston
Daniel Kamin, Children's Hospital Boston
Clifford Lo, Children's Hospital Boston
Mark Puder, Children's Hospital Boston
Chi-fu Yang, Harvard Medical School

The goal of this proposed study is to examine the effect of enteral omega-3 supplementation in improving liver function among patients who have intestinal failure-associated liver disease (IFALD). Intestinal failure-associated liver disease commonly affects children with intestinal failure and who are on parenteral nutrition. Clinical data suggests that intravenous omega-3 fatty acids are hepatoprotective. It is currently unknown whether enteral omega-3 fatty acids can further reduce liver inflammation. The primary aim is to examine the efficacy of omega-3 supplementation, when compared to placebo, on normalizing liver function, as measured primarily by amino alanine transferase (ALT). This study will be conducted as a randomized, double-blind, controlled trial. The intervention group will receive a fish oil supplement consisting of EPA and DHA. The control group will receive an olive oil supplement. Study will be for 1 year. Currently we have received conditional IRB approval pending FDA approval of our proposed fish oil supplement. This first-ever pilot study of enteral fish oil in children with intestinal failure will bring together researchers from surgery, gastroenterology, medicine, nutrition, pediatrics and pharmacology.

Impact of a Company-Based Sleep Apnea Screening, Diagnostic and Treatment Program on Truckers' Health and Safety

Principal Investigator: Stefanos Kales, Cambridge Health Alliance and Harvard School of Public Health

Co-Investigator(s): Charles Czeisler, Brigham and Women's Hospital
Atul Malhotra, Brigham and Women's Hospital
Chunbai Zhang, Harvard School of Public Health and Brigham and Women's Hospital

This pilot connects several Harvard affiliates across different disciplines: occupational health and sleep medicine, intersecting at the public health issue of obstructive sleep apnea (OSA) in transportation; along with a trainee looking to bridge these fields. Fatigue/sleepiness account for 20-30% of vehicular crashes, and annually in the US, accidents involving trucks/buses kill over 5,000 persons and cause in excess of 100,000 serious injuries. The most common medical cause of excessive daytime sleepiness is OSA, and commercial drivers have a high prevalence of OSA (17-28%). Thus, identifying drivers with OSA and effectively treating them should decrease fatalities/injuries and improve drivers' health. On the basis of its accident investigations, the National Transportation Safety Board has urged Federal Transportation agencies to adopt mandatory OSA screening. However, all such regulations are only proposed. We have teamed with a large trucking company with its own mandated approach to OSA screening, diagnosis and treatment. We seek to demonstrate with data gathered in conjunction with the company that comprehensive OSA screening, diagnosis and treatment with compliance monitoring can be delivered in the work setting, while reducing the rate of motor vehicle crashes and driver injuries; reducing absenteeism and turnover among truck drivers with previously undiagnosed and untreated OSA; and improving driver health outcomes with respect to diabetes and cardiovascular diseases. The major goal of our pilot would be to jumpstart our trainee's research career in occupational sleep medicine and develop preliminary data to support future proposals related to this important collaboration.

Improving Breast Cancer Diagnosis and Care in the State of Mexico

Principal Investigator: Nancy Keating, Harvard Medical School

Co-Investigator(s): Julio Frenk, Harvard School of Public Health
Felicia Knaul, Harvard Medical School
Elena Kouri, Harvard Medical School
Larry Schulman, Dana-Farber Cancer Institute

Breast cancer is the leading cause of cancer deaths among Mexican women and the second leading cause of death among Mexican women aged 30-54. Strategies to improve diagnosis and care in resource-poor areas, and evaluations of these strategies, are crucial to address this growing problem.

The proposed work reflects a new collaboration among researchers across the Harvard community and the Harvard Global Equity Initiative. The Ministers of Health in the states of Jalisco and Morelos have committed to improve breast cancer diagnosis and treatment by 1) expanding training opportunities for local health promoters, 2) providing infrastructure to survey health promoters, 3) providing access to existing data, and 4) supplementing existing data with qualitative research though key informant interviews and focus groups.

We will assess the ability of local efforts to expand screening services, train of health promoters and clinicians, and increase support from the Jalisco Cancer Institute (JCI) and National Cancer Institute (NCI) to local secondary hospitals to improve breast cancer knowledge, screening, early diagnosis, and treatment in Jalisco and Morelos. Specifically, we will explore the following specific aims (1) Develop a survey module to assess breast cancer knowledge and awareness among the states' general population; (2) Assess breast cancer knowledge among a community health care promoters before and after focused breast cancer training; (3) Assess stage at diagnosis of newly diagnosed breast cancers; and (4) Assess the capability of physicians in secondary hospitals to treat breast cancer with support of physicians at the JCI and the NCI.

Genomic Evolution of Bacteria During the Pathogenesis of Urinary Tract Infection in Humans

Principal Investigator: Roy Kishony, Harvard Medical School

Co-Investigator(s): Marc Cendron, Children's Hospital Boston
Alexander McAdam, Children's Hospital Boston

Infections of the urinary tract by pathogenic *E. coli* are a main cause pediatric morbidity. The pathogen moves sequentially through the bladder (cystitis), the kidneys (pyelonephritis) and can eventually reach the bloodstream, provoking sepsis and risk of death. These different compartments present distinct challenges to which bacteria need to adapt. It is likely that the pathogen's adaptation to these stresses largely determines the outcome of the infection. Yet, it is currently unknown how uropathogenic *E. coli* adapts to these different selection pressures during the infection. How do bacteria evolve as they move between compartments of the human body? Is adaptation compartment-specific? Which genes are under the strongest selective pressure in each compartment? These are fascinating fundamental questions with important medical consequence. To study the evolution of pathogenic *E. coli* during colonization of the human urinary tract, we will: (1) build a library of multiple clinical isolates from each individual patient, representing multiple time points in the same compartments, or multiple compartments at a given time; (2) reveal compartment-specific and compartment-general phenotypic adaptation by quantitatively measuring growth in environments representing challenges anticipated in the human body; (3) use high throughput sequencing technologies to identify the genetic changes that underlie adaptation. These results will point to specific bacterial genes under selection in the different compartments of the human body and could suggest novel targets for "compartment specific" therapeutics.

Effect of Psychosocial Stress and Oxytocin on Peripheral Blood Transcriptome

Principal Investigator: Sek Won Kong, Children's Hospital Boston

Co-Investigator(s): Laura Kubzansky, Harvard School of Public Health

Social relationships are pervasively impaired in neuropsychiatric disorders like autism spectrum disorders, while positive social relationships are associated with better health. Recent research suggests a key role of a nine-amino-acid neuropeptide, oxytocin in both coordinating positive social interactions and mitigating responses to social stress. Intranasal administration of oxytocin has been shown to modulate human social interactions although the mechanisms remain unclear. In human studies investigation of CNS tissue is infeasible, however examination of peripheral blood gene expression may provide insight. Other work has shown that pro/anti inflammatory pathways are perturbed in response to psychosocial stress, but effects of oxytocin treatment on blood gene expression have not yet been studied. Building on work suggesting oxytocin mitigates effects of social stress, we hypothesize that oxytocin treatment will reduce or normalize stress-related gene expression changes. The goal of this project is to establish a monitoring system to evaluate this question. This project has two specific aims: 1) To identify gene expression signature of oxytocin administration under basal conditions; 2) To develop a gene expression signature of acute psychosocial stress. For Aim 1 we will recruit 20 healthy males and employ a placebo-controlled double-blind study. For Aim 2 we will recruit 10 healthy males and use a validated protocol to induce social stress. In both samples, blood gene expression will be monitored at baseline and 1 hour after application of intranasal oxytocin/placebo or stress induction. We expect the gene expression signatures from oxytocin treatment group and from individuals under stress will be negatively correlated.

HIV-1 Viral Diversity in the Intestinal Mucosa of Individuals with Progressive, Treated, and Spontaneously Controlled Infection

Principal Investigator: Douglas Kwon, Massachusetts General Hospital

Co-Investigator(s): Nina Lin, Massachusetts General Hospital
Mary Sabatini, Massachusetts General Hospital
Blair Wylie, Massachusetts General Hospital

Abstract withheld at the request of the investigator.

Characterizing Spatiotemporal Variations in Traffic-Related Air Pollution in Cambridge and Somerville, Massachusetts

Principal Investigator: Jonathan Levy, Harvard School of Public Health

Co-Investigator(s): Rex Britter, Massachusetts Institute of Technology
Sam Lipson, Cambridge Health Alliance
Scot Martin, Harvard School of Engineering and Applied Sciences
Timothy McAuley, Consulting for Health, Air, Nature, & Greener Environment (CHANGE)
David Sittenfeld, Museum of Science, Boston
Matt Welsh, Harvard School of Engineering and Applied Sciences
Wig Zamore, Somerville Transportation Equity Partnership

Traffic-related air pollution contributes significantly to mortality and morbidity within urban areas, but few studies have adequately characterized spatiotemporal variations of primary mobile source pollutants for use in epidemiological applications or risk management. Previous studies have either utilized only fixed-site monitors resulting in inadequate spatial coverage, mobile measurements that are labor-intensive and unsustainable over time, and/or dispersion models that are highly uncertain at fine spatiotemporal scales. In this study, we will test real-time sensors on stationary and mobile platforms that can collect, synthesize, and analyze air quality data. Ideally, both platforms will measure nitrogen oxides, carbon monoxide, carbon dioxide, wind speed/ direction, temperature/humidity, and geographic location. Mobile sensors will be deployed using predefined monitoring protocols utilizing multiple modes of transport (e.g., bicycles, cars, pedestrians). Sampling routes and locations will be selected by academic and community partners to include various traffic patterns, potential hotspots, and sites with more vulnerable populations. Wireless networking methods will be used to upload and synthesize the monitoring data, leveraging where possible the CitySense (www.citysense.net) network in Cambridge and Somerville. These data will be analyzed using generalized additive mixed models to predict concentrations as a function of traffic volume, topography, meteorology, and central site monitoring data. Data collected will be evaluated for its statistical interpretability, reliability, and sustainability along with other defined attributes. This unique collaboration among environmental scientists, computer scientists, public health experts, and community representatives will provide the foundation for future studies of health outcomes and disparities, public education and outreach efforts, and policy interventions.

Extracellular Domain of DDR2, a Cell Membrane Receptor Tyrosine Kinase, as a Potential Therapeutic Agent for Osteoarthritis

Principal Investigator: Yefu Li, Harvard School of Dental Medicine

Co-Investigator(s): Christopher Evans, Beth Israel Deaconess Medical Center

Osteoarthritis (OA) is a global heath problem. Currently, there are no effective therapeutic agents for the treatment of the disease. Results from our recent investigations suggest that the activation and up-regulated expression of discoidin domain receptor 2 (DDR2), as the result of the interaction of the extracellular domain (ECD) of the receptor with native type II collagen, may be one of the crucial steps in the articular cartilage degeneration, eventually leading to OA. Importantly, data from our studies indicate that the reduced expression of DDR2 attenuates articular cartilage degeneration in the knee joints of genetic and non-genetic forms of mouse OA models. Based upon the above-mentioned observations, we hypothesize that the ECD of DDR2 can inhibit the activation and up-regulated expression of the receptor in chondrocytes by interrupting the interaction of DDR2 with native type II collagen, thus delaying the articular cartilage degeneration. To test this hypothesis, in this grant application we propose to perform the following experiments: 1) Investigate the role of soluble ECD of DDR2 in modulating the activation and expression of the receptor in chondrocytes. 2) Examine the efficiency in delivery of a gene product by a lentiviral vector in mouse knee joints. 3) Investigate the effect of the ECD of DDR2 on articular cartilage degeneration in the knee joints of genetic and non-genetic mouse OA models.

Results from this application will be critical for determining whether the ECD of DDR2 can be used as a therapeutic agent for the treatment of human OA.

Targeting of HIV-1 CTL Epitope/MHC Class I Complexes by Novel TCR-like Monoclonal Antibodies

Principal Investigator: Mathias Lichterfeld, Massachusetts General Hospital

Co-Investigator(s): John Christopher Love, Massachusetts Institute of Technology

During HIV-1 infection, viral proteins are degraded into small peptides that are presented by MHC complexes on the surface of cells. This process of antigen presentation serves two major functions: (i) Recognition of such peptide/MHC complexes allows for the priming and expansion of HIV-1-specific T cells, which are an important component of the adaptive immune response against HIV-1, and (ii) viral peptide presentation serves as the predominant way by which HIV-1-specific T cells can selectively recognize HIV-1 infected cells, and initiate their elimination by immune-mediated mechanisms. Despite these critical roles of HIV-1 peptide presentation, there is currently no technological method available that allows visualizing the frequency, intensity and specificity of viral peptides that are presented *in vivo*. Being able to analyze the process of HIV-1 antigen-presentation would significantly enhance our understanding of T cell mediated immune activity against HIV-1, and can lead to new pharmaceuticals that specifically target HIV-1 infected cells. The PIs here propose a novel, highly-innovative chip-based microengraving technology that can identify monoclonal antibodies recognizing given HIV-1 peptide/MHC complexes using a rapid, high-throughput screening process. Briefly, a polyclonal library of genetically-engineered antibody-producing yeast cells will be individually placed in >500,000 nanowells on a novel microchip. Subsequently, protein microarrays will be printed from the supernatant of each nanowell and interrogated with recombinant peptide MHC class I complexes. Identified antibodies can then be used as research tools for analyzing antigen presentation in primary human cells, and will be developed into novel drugs for the therapeutic targeting of HIV-1 infected cells.

HIV and Malignancy in Botswana: A Prospective Study of Incidence, Toxicity, and Outcomes

Principal Investigator: Shahin Lockman, Brigham and Women's Hospital

Co-Investigator(s): Scott Dryden-Peterson, Brigham and Women's Hospital
Dianne Finkelstein, Massachusetts General Hospital
Rajesh Gandhi, Massachusetts General Hospital
Tendani Gaolathe, Botswana Harvard AIDS Institute
Ann LaCasce, Dana-Farber Cancer Institute
Heluf Medhin, Non-Communicable Disease Control Programme, Ministry of Health, Botswana
George Seage, Harvard School of Public Health

Most cancer deaths occur in resource-limited settings. The lifetime risk of dying from cancer by age 65 is nearly twice as great in Africa as in developed nations, with cancers associated with infection and poverty joining cancers of increasing prosperity. Epidemiologic studies from developed nations suggest that HIV infection increases risk for non-AIDS-defining cancers, but conclusions have been limited by confounding (by social and lifestyle factors). Sub-Saharan Africa has the highest HIV prevalence rates in the world, but data on cancer in Africa are sparse. Botswana, with the only national cancer registry in Africa, centralized oncologic care, an intense, generalized HIV epidemic (with 25% of adults infected with HIV-1), and widely available HIV treatment, provides a unique opportunity to study the interaction between HIV infection, antiretroviral therapy, and cancer. We will initiate a prospective observational cohort in Botswana to achieve 3 specific aims: 1) describe spectrum of cancer diagnoses, patient characteristics, and 6-month survival for patients diagnosed with malignancy, 2) determine the prevalence of HIV infection and the median CD4 count for patients presenting for oncologic care (and association of HIV infection with specific types of cancer), and 3) compare the rate of treatment-limiting toxicity between HIV-infected patients taking or not yet taking antiretroviral therapy. Approximately 625 patients presenting with cancer at Princess Marina Hospital, 65% of nationally reported cancers, will be enrolled, HIV-tested, and followed prospectively (in clinic and at home). Data from this pilot cohort will be used to secure longer-term funding to further develop the project's potential.

ACL injuries: Setting Priorities for Care, Policy, Research

Principal Investigator: Elena Losina, Brigham and Women's Hospital

Co-Investigator(s): Jeffrey Katz, Brigham and Women's Hospital
Mininder Kocher, Children's Hospital Boston

In the US 200,000 persons tear their anterior cruciate ligament (ACL) each year and half of them undergo ACL reconstruction surgery. ACL injury puts people at risk for the early onset of knee osteoarthritis. Despite much research on this topic, many questions remain about the best ways of treating ACL injuries. Does surgery reduce the long-term risk of ACL injury on development of knee OA? Is prevention of ACL injury cost-effective? Is prevention of subsequent knee injury and OA in persons who have had an ACL injury effective and cost-effective?

Computer simulation models are an ideal way to study these problems. We propose to add an ACL injury component to a comprehensive computer simulation model of knee OA prevention and management. The model will enable us to examine the effects of ACL injuries on costs and quality of life over the life course of affected individuals. We propose two specific aims:

I. To incorporate ACL injuries into the Osteoarthritis Policy Model

II. To conduct a series of policy analyses addressing critical issues in ACL injury management:
-- To forecast the long-term clinical and economic consequences of ACL injuries
-- To estimate the cost-effectiveness of surgical and non-surgical management for ACL injuries
-- To evaluate cost-effectiveness of injury prevention programs

This study will provide guidance to clinicians and policy makers on efforts to reduce pain and disability and to improve the quality of life for people who have had or are at risk for sustaining ACL injuries.

Neurobiological Effects of Childhood Adversity: A 20-year Prospective Study

Principal Investigator: Karlen Lyons-Ruth, Cambridge Health Alliance

Co-Investigator(s): Pia Pechtel, Faculty of Arts and Sciences, Harvard University
Martin Teicher, McLean Hospital

Childhood adversity has been identified as the root preventable cause for a range of health difficulties (depression, heart diseases, obesity) and maladaptive behaviors (substance abuse, suicide attempts, risk behaviors) in adulthood. Despite its pressing role, little is known about the neurobiological effects of dysfunctional parenting and disorganized attachment - common forms of early adversity. This is due to the interdisciplinary divide between developmental psychologists studying the nuances of infant attachment, and researchers in psychiatric neuroimaging focusing on changes in brain structure/function associated with psychopathology. However, animal models have shown that alterations in parenting behavior can lead to epigenetic modifications of the glucocorticoid receptor gene, change the expression of trophic factors, and alter trajectories of brain development. Understanding how parenting affects brain development provides crucial insights necessary to devise treatment programs to preempt psychopathology. The proposed study will examine brain structure/function in 20 adults who participated, since infancy, in a prospective longitudinal study on parenting and attachment. We hypothesized that dysfunctional parenting and disorganized attachment will produce enduring effects on brain development leading to differences in gray matter volume, fiber-tract integrity and resting-state functional connectivity. This inter-institutional collaboration merges disparate expertise and resources, including a large preexisting neuroimaging database on various forms of childhood trauma. Merging this knowledge will result in a more profound and predictive biopsychosocial synthesis regarding the relationship between parenting and health. This pilot grant would assist the ultimate goal of identifying clinically-relevant, morphometric changes to tailor interventions to prevent the emergence of psychiatric consequences following childhood adversity.

Red Blood Cell Derived Microparticles in Malaria

Principal Investigator: Matthias Marti, Harvard School of Public Health

Co-Investigator(s): Natasha Barteneva, Immune Disease Institute
Alexander Ivanov, Harvard School of Public Health

Plasmodium falciparum causes the most severe form of malaria with over two million deaths every year. The morbidity and mortality of the disease can be attributed to the red blood cell stages of the parasite. The clinical manifestations of severe malaria are directly correlated with the induction of strong pro-inflammatory type-1 immune responses. Microparticles (MPs) have been identified as important pro-inflammatory triggers in human malaria. The cellular sources of these MPs are poorly defined. Recent data in the mouse malaria model suggest red blood cell derived MPs as the main component in the plasma. We hypothesize that RBC derived MPs are responsible for the high levels of circulating inflammatory mediators seen in patients with severe malaria.

In this pilot study, we will characterize RBC derived MPs in *P.falciparum*. We will define different subpopulations of MPs derived from *in vitro* cultured parasite infected RBCs by flow cytometry using a series of glycolipid, RBC and parasite markers. Next, we will establish a protocol for the purification of MP populations, either by FACS enrichment or biochemically. These pilot experiments will provide the basis for detailed lipidomic and proteomic analysis to identify the parasite and host components of red blood cell derived MPs. The involved parasite molecules have great potential as targets for interventions against malaria, and as biomarkers for severe disease.

New Technologies for Microbial Identification in Clinical Samples

Principal Investigator: Danny Milner, Brigham and Women's Hospital

Co-Investigator(s): Michael Chou, Harvard Medical School
Daniel Schwartz, Harvard Medical School

The rapid identification of pathogens is of critical importance because each pathogen requires different therapeutic approaches which are often incompatible with one another. The BWH Department of Pathology provides microbial testing to identify pathogens in a number of different clinical sample types. These tests use a variety of serological, culture-based and molecular methods – some of which take a long time. While nucleic acid sequencing should theoretically be able to provide definitive identification of all classes of pathogens, it is performed in only rare instances and can be quite expensive for multiplex panels of sequencing reactions. Conversely, culture methods can be relatively inexpensive; however, not all pathogens can be easily identified in this manner. For this Catalyst pilot study, we propose to develop assays which use new targeting technologies for panels of viral pathogens. Results generated may be used as preliminary data for further clinical and epidemiological studies.

Effects of a Food Preservative on Glucose Homeostasis

Principal Investigator: Vamsi Mootha, Massachusetts General Hospital

Co-Investigator(s): Belinda Lennerz, Children's Hospital Boston
David Ludwig, Children's Hospital Boston
Scott Vafai, Massachusetts General Hospital

Abstract withheld at the request of the investigator.

Melatonin and Prostate Cancer: A Biomarker Study Among Men in the Reykjavik Cohort

Principal Investigator: Lorelei Mucci, Harvard School of Public Health

Co-Investigator(s): Charles Czeisler, Brigham and Women's Hospital
Matthew Freedman, Dana-Farber Cancer Institute
Steven Lockley, Brigham and Women's Hospital
Meir Stampfer, Harvard School of Public Health

The International Agency for Research on Cancer identified night shift work as a probable human carcinogen. One proposed mechanism is through suppression of levels of the light-sensitive hormone melatonin, produced at night by the pineal gland, due to nocturnal light exposure associated with night work. Shift work data have led to the hypothesis that other factors that alter melatonin levels could impact cancer initiation and progression. Although this hypothesis has been studied in some detail in breast and other cancers among women, there is scant human data examining the melatonin hypothesis in prostate cancer.

We propose a biomarker study of melatonin levels, germline variants, and pineal gland structure in relation to prostate cancer risk and progression nested in the Reykjavik Cohort of men residing in Reykjavik, Iceland during 1967 to 2008. This rich epidemiological study with detailed physiologic measures, biospecimens, and follow-up over 40 years is virtually untapped with respect to cancer studies. The following aims will be evaluated.

The aims for the proposed study are: 1-) Using a nested case-control study, we will determine whether men with high levels of the major metabolite of melatonin, 6-sulfatoxymelatonin, are at lower risk of prostate cancer, particularly aggressive forms of disease. 2-) Using existing GWAS data, we will identify genetic variants that influence urinary levels of melatonin; and 3-) Using prospectively assessed MRI brain images, we will quantify pineal gland volume and extent of calcifications, under the hypothesis that reduced gland volume or increased calcification is associated with lower melatonin levels, thereby increasing prostate cancer risk and progression.

Cool Comply: Patient Support Optimization

Principal Investigator: Kristian Olson, Massachusetts General Hospital

Co-Investigator(s): Aya Caldwell, Massachusetts General Hospital
Anne Goldfeld, Immune Disease Institute
Jose Gomez-Marquez, Massachusetts Institute of Technology
Amy Smith, Massachusetts Institute of Technology

Abstract withheld at the request of the investigator.

Nanowire Microarray Platform for Discovery and Delivery of Cell Reprogramming Factors

Principal Investigator: Hongkun Park, Faculty of Arts and Sciences, Harvard University

Co-Investigator(s): Thorsten Schlaeger, Children's Hospital Boston

Controlled generation of user-defined, patient-specific cell types through cellular reprogramming will one day revolutionize clinical medicine. Unfortunately, despite three decades of stem cell research, it is still not possible to produce significant amounts of clinically safe and therapeutically relevant, patient-specific cells by *in vitro* differentiation. This particular fact is a testament to the need for novel and effective, transgene-free cellular reprogramming and targeted differentiation strategies. We propose to develop a transformative new technology that enables the direct and efficient introduction of multiple cell-fate effectors into virtually any cell type and subsequently use this technology to derive hematopoietic stem cells, from both mouse embryonic stem cells and human induced pluripotent stem cells. At the core of the technology is a vertical silicon nanowire platform that enables the introduction of bioactive macromolecules directly into a cell's cytoplasm through the process of physical penetration. This transfection method is extremely efficient, even with primary cells that are notoriously difficult to transfect or transduce by conventional methods. Moreover, this technique does not affect cell viability or functionality and is compatible with multiplexing (more than one type of molecule can be introduced simultaneously into the same cell). Finally, since the nanowires are presented on flat surfaces, the method is compatible with microarray technology. We anticipate that the approach and methods developed in the proposed research effort can be generalized to a wide variety of cell types and may ultimately help uncover new routes to cellular reprogramming in a much broader fashion.

Hyperpolarized Noble Gas MRI Program at Harvard

Principal Investigator: Samuel Patz, Brigham and Women's Hospital

Co-Investigator(s): James Butler, Harvard School of Public Health and Brigham and Women's Hospital
N. Stuart Harris, Massachusetts General Hospital
Hiroto Hatabu, Brigham and Women's Hospital
Iga Muradyan, Brigham and Women's Hospital
Michael Patz, Harvard Medical School
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Matthew Rosen, Faculty of Arts and Sciences, Harvard University
Ronald Walsworth, Faculty of Arts and Sciences, Harvard University
George Washko, Brigham and Women's Hospital

Hyperpolarized noble gas MRI has demonstrated regional maps of pulmonary function. This noninvasive technology has generated considerable excitement because it can potentially fill a significant gap in clinical diagnostic methods. Expertise is needed in multiple disciplines to make progress: polarization physics to “hyperpolarize” either 3He or 129Xe gas, MRI physics, pulmonary physiology, and radiologists and pulmonologists. We have established a unique Harvard-based hyperpolarized noble gas program that incorporates all of these elements with investigators from BWH, MGH, Harvard Smithsonian Center for Astrophysics, HSPH and HMS. We seek funding for an initial collaborative study, the results from which will be used to seek further extramural support. We plan to study High Altitude Pulmonary Edema (HAPE), whose mechanism is currently not completely understood. A low inspired fraction of oxygen results in a hypoxic pulmonary vasoconstriction (HPV) response as well as elevated pulmonary artery pressure PAP. It is believed that the HPV response is heterogeneous, leading to heterogeneous perfusion (Q). Proton MRI has demonstrated a heterogeneous Q after exposure to hypoxia, however the relative contribution of alveolar partial pressure of oxygen (PAO2) and HPV to the observed heterogeneity in Q has not been studied. We will use both hyperpolarized 3He and proton MRI to determine this. A second component of this study will use proton MRI to determine whether or not areas with high Q, which are subjected to higher PAP and higher mechanical stress do in fact go on to develop edema as was proposed by Hultgren more than 30 years ago.

Breathing Abnormalities During Seizures

Principal Investigator: Milena Pavlova, Brigham and Women's Hospital

Co-Investigator(s): Sanjeev Kothare, Children's Hospital Boston

Sudden unexplained death (SUDEP) is 40 times more common in epilepsy patients than in the general population. Death often occurs in sleep and may be related to ventilatory abnormalities, cardiac arrhythmias or a combination of factors. Understanding the mechanisms of SUDEP will help in the prevention of this devastating complication.

Our goals are to evaluate: 1. Respiratory pattern, before, during, and after an EEG-documented seizure; and 2. Determinants of hypoxemia based on seizure characteristics, respiratory function, and patient demographics. We hypothesize that: 1. Increased minute ventilation, as well as particular locations of seizure origin, specifically mesial temporal and frontal locations, will be associated with more frequent central apneas; 2. Obstructive apneas will occur ictaly and post-ictally; 3. Patients with prior lung disease will have more severe seizure-related respiratory abnormalities.

We propose to monitor respiratory effort, airflow, carbon dioxide, oxygenation, body position, and EKG in relation to seizures in children and adults. Our proposed study requires a multidisciplinary approach, as expertise from epileptology, sleep medicine, and pulmonology are needed. Additionally, as age-related developmental maturation may have an important role, we propose to study both children and adults in the same protocol. We propose a collaboration between the sleep and epilepsy programs at Children’s Hospital and Brigham and Women’s Hospital with input from adult and pediatric pulmonologists from each hospital. The two epilepsy programs monitor over 450 patients every year. Thus, we anticipate being able to study 100 patients in one year to obtain meaningful data to substantiate a larger funding proposal.

Staphylococcus aureus Colonization and Infection in the Neonatal Intensive Care Unit

Principal Investigator: Karen Puopolo, Brigham and Women's Hospital

Co-Investigator(s): Jean Lee, Brigham and Women's Hospital
Bruce Paster, The Forsyth Institute

Despite advances in neonatal intensive care, very-low birth weight, prematurely-born infants continue to experience high rates of infection-related morbidity and mortality. Abnormal colonization with pathogenic bacteria acquired in the neonatal intensive care unit (NICU) not only increases the risk of invasive bloodstream infection, but may also influence the short-term development of non-infectious neonatal morbidities and the long-term risk of immune-based inflammatory disorders. A weekly infection control screening program for nasal or rectal carriage of methicillin-resistant *Staphylococcus aureus* (MRSA) at the Brigham and Women's Hospital NICU has revealed abnormally high rates of colonization with methicillin-sensitive *Staphylococcus aureus* (MSSA) among premature infants. MSSA is also the most frequently isolated pathogen in cases of hospital-acquired infection among these infants. The specific aims of the proposed study are to determine the molecular background of neonatal MSSA isolates, and the clinical characteristics of the colonized and infected infants. The study will utilize microbial genomic methods to determine the longitudinal relatedness of MSSA isolates; microarray-based methods to determine the relationship between MSSA colonization and the overall composition of the infant's oropharyngeal bacterial community; and epidemiologic methods to correlate specific neonatal morbidities with colonization status. Information gained from this study will be used to develop specific strategies to prevent pathogenic Staphylococcal colonization and invasive infection among premature infants.

The Association Between Mycoplasma Genitalium and Preterm Delivery at an Urban Community Health Center

Principal Investigator: Hope Ricciotti, Beth Israel Deaconess Medical Center

Co-Investigator(s): Sarah Averbach, Beth Israel Deaconess Medical Center
Jordan Dimitrakov, Children's Hospital Boston
Michele Hacker, Beth Israel Deaconess Medical Center
Timothy Yiu, Harvard Medical School

Background: Preterm delivery (PTD) is one of the leading causes of neonatal morbidity and mortality; yet the majority of PTD is idiopathic. Several reproductive tract infections are associated with PTD, and some data suggest that *Mycoplasma genitalium*, a relatively common cervical bacterium, may also play a role. *M. genitalium* colonization has been associated with cervicitis, pelvic inflammatory disease and endometritis. The Dimock Center's data indicates that in 2008, 17.8%, or 25 of 140 deliveries were less than 2500 grams. This is higher than the national average of 8.1%.

Objective: The purpose of this study is to determine the prevalence of cervical *M. genitalium* colonization and whether it is associated with PTD among women at an urban community health center.

Methods: This is a prospective cohort study of pregnant women at the Dimock Center in Roxbury, MA, which serves population at high risk for low birth weight and PTD. Given the state of knowledge about *M. genitalium*, this will be a pilot study of 100 women. Demographic, medical and obstetric data will be gathered, and cervical and urine samples will be collected at the initial prenatal visit and at 35 to 36 weeks of gestation. *M. genitalium* testing will be done using PCR.

Outcomes: (1) prevalence of cervical *M. genitalium* colonization among women at high risk for low birth weight and PTD (2) sensitivity of PCR to detect *M. genitalium* in mid-stream urine samples compared with cervical samples, and (3) risk of preterm labor, PTD, and low birth weight neonates among women with antepartum *M. genitalium* colonization compared to women without colonization.

Efficient Access to 18F-DOPA: A Diagnostic Tool for Parkinson's and Cancer

Principal Investigator: Tobias Ritter, Faculty of Arts and Sciences, Harvard University

Co-Investigator(s): Thomas Brady, Massachusetts General Hospital
Umar Mahmood, Massachusetts General Hospital
Ji-Quan Wang, Massachusetts General Hospital

Currently, many promising [18F] PET tracers cannot be accessed because there is no general chemical reaction available to make [18F]-carbon bonds. We have developed a new fluorination reaction that can make carbon-fluorine bonds in complex molecules. Our fluorination reaction has a larger scope than any other fluorination reaction developed by chemists or Nature. In this proposal we propose to develop a [18F] version of our fluorination reaction and apply it to the synthesis of the PET tracer F-DOPA. Conceptually, our project may result in a new way to make PET tracers and may significantly increase the efficiency for PET tracer synthesis for molecular imaging.

Discovering Compounds that Overcome Differentiation Arrest in Acute Myeloid Leukemia

Principal Investigator: David Scadden, Massachusetts General Hospital

Co-Investigator(s): Lee Rubin, Faculty of Arts and Sciences, Harvard University
David Sykes, Dana-Farber Cancer Institute

Acute myeloid leukemia (AML) arises from two types of mutations: those promoting proliferation, and those inhibiting differentiation. AML in adults is a devastating disease, with a 5-year survival of 25%. In contrast, a small subset of AML - acute promyelocytic leukemia – has a 5-year survival of 80% due to the discovery of therapies that promote differentiation. Developing similar differentiation therapies in the remaining 90% of AML has been hindered by inadequate model systems. We have developed a novel model for defining biologically relevant compounds that overcome differentiation arrest due to specific leukemogenic alleles. Human leukemic oncoproteins were rendered conditional by fusion with the hormone binding domain of the estrogen receptor. Primary murine bone marrow cells expressing green fluorescent protein (GFP) under the control of the lysozyme promoter were transduced with these conditional oncoproteins. The resulting myeloblast cell lines express GFP only when the oncoproteins are inactivated and the cells permitted to terminally differentiate, thereby providing a system in which to test for compounds which can also inactivate these critical oncoproteins. These cell lines have the advantage of being derived from primary marrow, available in unlimited supply, capable or normal maturation, and bearing a built-in marker of differentiation. We have demonstrated the feasibility of screening in 384-well format with a 2-log dynamic range of fluorescence between positive and negative controls. We plan to use this unique model system to identify compounds capable of overcoming the differentiation blockade of human leukemogenic oncoproteins.

Utilization and Cost Efficiency of Observation Care for Emergency Department Patients with Chest Pain in Massachusetts

Principal Investigator: Jeremiah Schuur, Brigham and Women's Hospital

Co-Investigator(s): Carlos Camargo, Massachusetts General Hospital and Brigham and Women's Hospital
V.G. Narayanan, Harvard Business School
Arjun Venkatesh, Brigham and Women's Hospital
James Ware, Harvard School of Public Health

Rationale: Chest pain (CP), a leading reason for emergency department (ED) visits, frequently results in hospital admission, leading to significant health care costs. Observation care of ED CP patients is efficient compared to inpatient care, yet there has been little study of its use across healthcare systems.

Specific Aims: 1) To describe hospital-level variation in utilization of observation care for ED patients with CP, according to several performance measures after adjustment for patient characteristics and disease severity; 2) To determine if hospitals with higher utilization of observation care for ED patients with CP are more cost-efficient; and 3) To determine if hospitals with an ED-based observation unit (EDOU) use observation care more efficiently than hospitals without an EDOU.

Methods: Retrospective cohort study of all ED visits in Massachusetts from 2008-2009. ED visits will be identified from administrative case-mix datasets and linked to a recent statewide survey that identifyied major features of EDs. Using the Clinical Classification Software to group ICD-9 codes, patients with nonspecific CP, coronary atherosclerosis & other heart disease, or acute myocardial infarction will be identified. Risk-standardized rates of performance measures will be calculated using multilevel models, adjusted for disease severity and facility type. Costs will be described at the hospital level using cost-to-charge ratios derived from publicly available hospital financial reports.

Significance: The study addresses the largely untested association between use of observation services, availability of EDOU, and important clinical and financial outcomes. Success will support a federal application to expand the study to more states; thereby improving generalizability.

A Novel Computer Algorithm to Predict Visual Field Function Based on Structural Imaging in Glaucoma Patients

Principal Investigator: Lucy Shen, Massachusetts Eye and Ear Infirmary

Co-Investigator(s): Louis Pasquale, Massachusetts Eye and Ear Infirmary
Andy Tsai, Children's Hospital Boston

Glaucoma, the second leading cause of blindness in the US, is characterized by progressive and irreversible damage of the optic nerve and associated visual field loss. The treatment of glaucoma is dependent on early and accurate diagnosis of the disease. The standard of care for glaucoma include structural assessment, which is not sensitive at detecting early damage, and functional assessment via visual field testing, which may be sensitive, but is subjective, time-consuming, and often unreliable. Spectral-domain OCT provides objective, reliable, and quantitative structural evaluation of the macula and optic nerve in an efficient and noninvasive manner. A complex relationship exits between the structural information obtained by SD-OCT and the functional data generated from visual field testing. We aim to develop a novel training-based computer algorithm using a coupled principal component analysis approach to predict the outcome of the visual field testing based on the SD-OCT scans. Our preliminary cross validation study showed good correlation between predicted and measured visual field parameters (R-squared=0.7). We are a team of two glaucoma specialists and one radiologist with expertise in medical image processing. We plan to collect a series of visual field and structural data from 50 subjects at the Massachusetts Eye and Ear Infirmary, in order to refine the algorithm for better visual field prediction and for accurate diagnosis of early glaucoma. This algorithm, when combined with the use of SD-OCT, has the potential to eliminate the need for visual field testing, and may streamline the diagnosis and management of glaucoma.

A Novel Seizure Prediction System Based on Modulations of Pre-ictal Neurodynamics

Principal Investigator: Catherine Stamoulis, Beth Israel Deaconess Medical Center

Co-Investigator(s): Rebecca Betensky, Harvard School of Public Health
Bernard Chang, Beth Israel Deaconess Medical Center

Approximately 3 million people in the US suffer from epileptic seizures and 30-40% of them have pharmacologically intractable seizures which severely affect their safety and quality of life. Alternative treatments, including brain stimulation, remain sub-optimal as they rely on robust seizure prediction, a very difficult problem due to the high variability of pre-seizure neural activity in the brain. Prediction methods based on potentially chaotic brain dynamics prior to clinical onset require long, often impractical electroencephalographic (EEG) recordings. Machine learning algorithms require the identification of robust pre-ictal features, a difficult problem given seizure inhomogeneity even for individual patients. Methods based on waveform and spectral parameters also have proved to be highly inconsistent. Nevertheless there is electrophysiological evidence of pre-ictal changes in baseline neurodynamics, possibly reflecting impending seizures, which may be used for prediction. However, a different computational approach is necessary. We propose to develop a seizure prediction system with no *a priori* knowledge of seizure dynamics and features, using 1) short pre-ictal and baseline EEGs and 2) information and network coordination measures estimated spatio-temporally from these EEGs. Fifty patients with at least two seizures each will be analyzed, to develop and validate the method. In limited preliminary studies we have shown significant pre-ictal changes in channel entropy, inter-channel mutual information and relative EEG phase at high frequencies, all detectable prior to seizure onset. The proposed system will have a significant impact on the clinicians' ability to predict and prevent disabling epileptic seizures.

Discovery and Validation of Wilms Tumor Markers Using Urine Proteomics

Principal Investigator: Hanno Steen, Children's Hospital Boston

Co-Investigator(s): Alex Kentsis, Dana-Farber Cancer Institute
Elizabeth Mullen, Dana-Farber Cancer Institute
Carlos Rodriguez-Galindo, Dana-Farber Cancer Institute

Wilms tumor is the most common kidney cancer of childhood and remains difficult to treat, with limited means to identify patients at risk for therapy failure or disease relapse. A variety of renal tumor markers are known to exist, but none have sufficient sensitivity and specificity for clinical use. Recently, we developed advanced mass spectrometry approaches to identify several thousands of distinct proteins in human urine, including novel and accurate diagnostic markers of human disease. Leveraging the already collected specimens of the national Children's Oncology Group Renal Tumor Bank, we propose to use high accuracy mass spectrometry urine proteomics to examine the composition of urine of patients with Wilms tumor in order to discover and validate novel and accurate disease markers. Such markers may be used to guide initial risk stratification, to facilitate early diagnosis and treatment, to monitor for residual disease in the course of treatment in order to identify patients at risk of relapse, and potentially to discover novel biologic pathways and therapeutic targets. This work will establish a collaborative research program among experts in clinical oncology, proteomics, and experimental therapeutics. Such a paradigm of translational, cross-disciplinary science promises to revolutionize the diagnosis and treatment of pediatric kidney tumors, with far reaching implications for the screening, diagnosis, and treatment of a wide variety of cancers, and molecular understanding of kidney cancer.

Genetics of Gene Expression in Preeclampsia

Principal Investigator: Barbara Stranger, Brigham and Women's Hospital

Co-Investigator(s): Thomas McElrath, Brigham and Women's Hospital

Preeclampsia (PE) is one of the most common disorders of pregnancy, affecting 3-8% of women in the developed world and up to 10% worldwide. PE pregnancies pre-dispose offspring towards below-average birth weight and preterm delivery, as well as maternal and infant morbidity and mortality. Through heritability studies, PE has been shown to have a strong genetic component, although only a small number of risk-inducing polymorphisms have replicated robustly, and little is known about their mechanisms of action. While small-scale studies have identified differentially expressed genes in placenta of PE cases versus controls, none of these studies has evaluated the role of genetic variation contributing to transcriptional variation. The objective of this research application is to identify genetic variants contributing to PE though effects on gene expression in maternal decidua and placenta. We propose to define the genome-wide transcriptional profile of paired maternal and placental tissues from early-onset preeclamptic mothers and normotensive control mothers. Maternal and fetal cord blood will be subjected to genome-wide genetic profiling, including both single nucleotide polymorphisms and copy number variation. This dataset will permit identification of genes that differ in expression level between maternal decidua and placenta of healthy and preeclamptic mothers, but more importantly, we will use the tools of expression quantitative trait locus (eQTL) mapping to identify genetic variants associated with expression levels, and to test for significant interaction with disease status. These findings will provide important preliminary data for an R01 proposal to apply a systems biology approach to the study of PE.

Therapeutic Potential of Endothelin Receptor Antagonism in LAM

Principal Investigator: Andrew Tager, Massachusetts General Hospital

Co-Investigator(s): Manuela Funke, Massachusetts General Hospital
Elizabeth Henske, Brigham and Women's Hospital

Abstract withheld at the request of the investigator.

Liver Engineering Using Whole Organ Bioscaffolds

Principal Investigator: Khashayar Vakili, Children's Hospital Boston

Co-Investigator(s): Heung Bae Kim, Children's Hospital Boston
Martin Yarmush, Massachusetts General Hospital

Each year approximately 27,000 people die from liver disease and 3000 people die on the transplant waiting list. This is a result of a critical shortage of organs. One approach to overcome this shortage is to engineer organs for transplantation. A major hurdle in engineering a complex organ such as the liver is recapitulating its structure which contains hepatocytes, cholangiocytes, blood vessel arranged in a complex 3-dimensional structure. One of the major advancements in the field of tissue engineering has been the use of extracellular matrices. Recently, a novel technique has been described in which a whole organ is decellularized leaving the extracellular matrix as a bioscaffold which continues to maintain the 3-dimensional structure of the organ. In early experiments, liver bioscaffolds have been successfully re-populated with hepatocytes and endothelial cells. Re-populated livers have been shown to synthesize liver specific proteins and bile salts while maintained on an *ex vivo* perfusion system. In addition, early *in vivo* experiments have demonstrated short-term (8 hours) viability of the re-populated liver graft in a rat model. However, long term survival of the re-populated liver bioscaffolds has not been demonstrated. Our proposal seeks to i) Refine the rat transplant model in order to maintain longer *in vivo* graft survival ii) Assess the growth of the liver bioscaffold in the setting of partial native hepatectomy which creates a regenerative environment.

The establishment of the long term viability of the re-populated liver bioscaffold *in vivo* will be an important step in the development of engineered livers.

Clinical Imaging of Capillary Malformations Using Optical Frequency Domain Imaging

Principal Investigator: Benjamin Vakoc, Massachusetts General Hospital

Co-Investigator(s): Brett Bouma, Massachusetts General Hospital
Marilyn Liang, Children's Hospital Boston
Jennifer Lin, Brigham and Women's Hospital

Capillary malformations (CM) present as pink or red lesions at birth and are frequently found on the face and neck. These malformations result from vascular ectasia in the superficial dermis. While benign, they present a measurable psychological burden on young adults and children. Current treatment utilizes vascular-targeting lasers such as pulsed-dye lasers (PDL) to destroy the blood vessels selectively. While PDL therapy has proven benefit to most patients, a small but significant subset of patients respond poorly. The properties of the CM microvasculature that predispose some patients to poor PDL therapy response are poorly understood. We have recently developed optical frequency domain imaging (OFDI) instrumentation and techniques for highly sensitive microvascular imaging in murine tumor models. Key features of the technology include a large imaging field, deep tissue penetration to 2 mm, and entirely endogenous contrast mechanisms. In this pilot grant, we propose to use OFDI to characterize the response of CM to PDL therapy. First, we will develop an imaging head to adapt to the existing OFDI system to operate in a dermatology clinic. Second, in a pilot study, we will image patients prior to and after PDL therapy. Through this work, we expect to establish OFDI as a powerful tool for imaging vascular lesions in the skin, providing the foundation for further research and clinical studies.

Acute Metabolic Influences on the Natriuretic Peptide System

Principal Investigator: Thomas Wang, Massachusetts General Hospital

Co-Investigator(s): Kenneth Bloch, Massachusetts General Hospital
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The heart synthesizes a family of hormones known as the natriuretic peptides (NPs). These molecules have a variety of salutary effects, including natriuresis, vasodilation, and inhibition of cardiac hypertrophy. Accordingly, the NP system plays a key role in the compensatory response to cardiac pressure and volume overload. We previously showed that obesity is associated with lower NP levels, which may predispose obese individuals to hypertension and left ventricular hypertrophy. The underlying mechanisms have not been established, however. Recently, we found that NP levels rise rapidly following weight loss surgery, suggesting that fat mass may not be the primary mediator of the obesity/NP association. Further, we demonstrated that mice fed a high-fat diet had rapid suppression of left ventricular NP expression, also supporting the role of humoral mediators. It is critical to recapitulate these findings in humans, to understand the time course of NP responses, and to determine whether carbohydrates, fat, or both stimulate these responses. Such data are best obtained from detailed metabolic studies using dietary interventions. This represents a new direction for our group, but one that is key for extending our initial findings from epidemiologic and laboratory settings. We propose to assess NPs and NP-related metabolites before and after administration of isocaloric high-carbohydrate and high-fat meals, in both lean and obese individuals. The proposed study will form the basis for further work in this area, to more fully understand the interaction of metabolism, the NP system, and cardiovascular structure/function. These areas of future discovery have important public health implications, given the rising burden of obesity and cardiovascular disease.

Assessing Brain Connectivity Disruption in Tuberous Sclerosis Complex

Principal Investigator: Simon Warfield, Children's Hospital Boston

Co-Investigator(s): Deborah Burstein, Beth Israel Deaconess Medical Center
Ellen Grant, Children's Hospital Boston
Mustafa Sahin, Children's Hospital Boston
Benoit Scherrer, Children's Hospital Boston

Tuberous sclerosis complex (TSC) is an autosomal dominant disease characterized by the presence of benign tumors, called hamartomas, which can affect virtually every organ system of the body, including the brain (known as cortical tubers). Epilepsy is common, and over 40% of patients with TSC have intellectual disability with 25-50% of TSC patients being diagnosed with autism spectrum disorder. The prognosis for these children varies tremendously across individuals. The cause of neurological deficits in TSC patients is a key unresolved question, but recent studies suggest they are due to white matter abnormalities. Previous work suggests that in the TSC brain there may be a miswiring of neuronal connections that are independent of the benign tumors and that these wiring disruptions may contribute to the development of neurological symptoms in TSC patients. High resolution animal MRI of mouse models of TSC will enable us to investigate the microstructural changes in the brain caused by TSC. We propose to create a new research team by bringing together experts in neurostructural characteristics of the TSC mouse model, and human diffusion MRI. Joint diffusion MRI analysis and histological analysis of mouse brains will enable us to determine the precise cause of changes in the diffusion MRI signal. This will enable us to infer the cause of changes of the diffusion signal in human MRI. This will enable the utilization of diffusion MRI as a biomarker for prognosis, for evaluating the success of particular interventions and for assessing response to therapy in ongoing clinical care.

Genetic and Environmental Predictors of Trauma Related Outcomes in Hurricane Katrina Survivors

Principal Investigator: Mary Waters, Faculty of Arts and Sciences, Harvard University

Co-Investigator(s): Karestan Koenen, Harvard School of Public Health
Jordan Smoller, Massachusetts General Hospital

The search for predictors of morbidity and resilience following trauma and disaster has become a focus of intense research.However, studies to date have relied on retrospective assessment of pre-trauma characteristics, a fundamental methodologic limitation. This interdisciplinary collaboration brings together Harvard faculty with expertise in sociology, psychiatry and genetics and builds on an unique longitudinal study of Hurricane Katrina survivors to identify gene-environment interactions predicting outcomes following disaster-related trauma. Prior to Hurricane Katrina, Waters and colleagues assessed 1019 low-income parents on baseline demographic and health information, including measures of social support and psychological distress. The sample was surveyed using repeated measures, as well as a new module on Hurricane experiences and posttraumatic stress disorder (PTSD) in 2006, one year after the hurricane and again in 2009, four years after the storm. (72% response rate). The study focuses on how pre-hurricane resources, capacities and systems—including mental and physical health, social networks, economic resources—affect adjustment after the trauma. We apply here to begin a new collaboration among Waters, a sociologist at FAS who is an expert on migration and young adulthood, Jordan Smoller a psychiatric geneticist at MGH, and Karestan Koenen, an epidemiologist with expertise in PTSD and genotype-environment interaction at HSPH. We seek funds to collect and analyze DNA to explore whether specific genetic polymorphisms modify the effect of social and individual environmental exposures on post-hurricane adjustment including PTSD. This effort would represent the first-ever prospective study of genetic and environmental predictors of trauma-related outcomes.

Using Mobile Electronic Protocols to Improve the Quality of Care for Newborns in Rural Tanzania

Principal Investigator: Kim Wilson, Children's Hospital Boston

Co-Investigator(s): Tyler Hartman, Children's Hospital Boston
Marie McCormick, Harvard School of Public Health
Marc Mitchel, Harvard School of Public Health
Jonathan Spector, Massachusetts General Hospital

Finding innovative means to deliver effective health care to newborns in low-income countries is essential to meeting the child survival goals of MDG4. Existing evidence based guidelines for newborns are not widely implemented as a high percentage of newborns are born in primary health settings where most health workers have limited knowledge of neonatal case management. Mobile electronic protocols, which use the small computer in mobile phones to provide decision support in following healthcare protocols, have the potential to greatly expand the capacity to deliver high quality health care to newborns. This technology has proven successful in improving quality of health care by primary health workers in AIDS screening and in care for older ill children.

We aim to develop and field test mobile e-protocols for the newborn period, and conduct a pilot study of feasibility and efficacy. We will compare compliance with established guidelines by clinicians during a period of standard care vs. care delivered using the e-protocols in three facilities in Tanzania. We will use an observation checklist to evaluate key elements of established guidelines. We will conduct a qualitative assessment of perceptions of the e-protocols. The project will inform the design of a larger clinical trial to assess the effect of mobile electronic decision support and record keeping on newborn survival.

Exposure to Biomass Smoke During Pregnancy: A Pilot Study to Examine the Role of Placental Damage

Principal Investigator: Blair Wylie, Massachusetts General Hospital

Co-Investigator(s): Majid Ezzati, Harvard School of Public Health
Wafaie Fawzi, Harvard School of Public Health
Drucilla Roberts, Massachusetts General Hospital

One half of the world's women are exposed to smoke from biomass fuel cooking fires. Smoke from biomass fuels like dung, wood, or charcoal contains numerous pollutants including particulate matter and carbon monoxide, many of which are shared with cigarette smoke.

Preliminary epidemiologic investigations suggest that maternal biomass smoke exposure during pregnancy decreases birth weight. These studies have suffered from a lack of detailed exposure assessment and biologic samples. This has restricted our ability to investigate the mechanisms of hazardous effects. Our catalyst research evaluates the potential for using the placenta, a window into the maternal-fetal interface, to make major advances in understanding the pathophysiology of impaired fetal growth resulting from biomass smoke exposure.

In this pilot of 20 never-smoking Tanzanian pregnant women (15 cooking with biomass, 5 with natural gas or electricity), we will directly measure maternal particulate matter and carbon monoxide exposures during pregnancy and correlate this exposure with the degree and pattern of placental damage observed on histopathology. We will further compare histopathologic features of these placentas with those of 5 US smokers. The study unites experts in maternal-fetal medicine, environmental health, placental pathology and epidemiology in a new collaboration and fosters mentorship of a junior faculty member as principal investigator.

To our knowledge, no study has examined the placental pathology of biomass smoke-exposed pregnancies. If successful, this pilot work will be foundational to further research aimed at understanding how biomass smoke impairs fetal growth, a leading risk factor for infant mortality in the developing world.

Development of New Molecular Genetic Tests for Neurological Diseases

Principal Investigator: Winnie Xin, Massachusetts General Hospital

Co-Investigator(s): Michael Chou, Harvard Medical School
Katherine Sims, Massachusetts General Hospital
Joseph Thakuria, Massachusetts General Hospital

The Neurogenetics DNA Diagnostic Laboratory (NDDL) at the MGH Center for Human Genetic Research currently provides clinical genetic testing services for a number of neurological and metabolic diseases. At present, all clinical genetic sequencing labs use traditional Sanger-based DNA sequencing technology to provide mutation analysis of mutant alleles of single genes or small panels of genes on a per patient basis. Screening for mutations in large numbers of candidate disease genes can be cost prohibitive and can take months to analyze.

Recent developments in high throughput sequencing technology have dramatically reduced the cost of sequencing resulting in several recent publications of early whole genome and exome data. The Church lab has developed technology that can harness the power of these next-generation sequencing platforms to economically sequence targeted sets of genes in human samples. However, this has yet to be tested in clinical samples.

For this pilot study, we propose to apply and validate these new targeting technologies for sets of candidate genes frequently involved in neurodegenerative and neurodevelopmental disorders including Amyotrophic Lateral Sclerosis, Neuronal Ceroid Lipofuscinoses, Primary Dystonia and mental retardation. Because of the NDDL's current involvement in testing for these diseases, hundreds of IRB approved patient samples are available to be used as controls for this study. Results generated may be used as preliminary data to justify use of this approach for future clinical testing, and sequencing tools generated may enable future research studies that require sequencing sets of candidate genes in many patients with familial and sporadic disease.

The Development of a SIV/Rhesus Monkey Penile Mucosal Transmission Model

Principal Investigator: Wendy Yeh, Beth Israel Deaconess Medical Center

Co-Investigator(s): Keith Mansfield, Harvard Medical School

Although an HIV vaccine represents our best hope to combat the global AIDS pandemic, its discovery has been elusive. One of the barriers to developing an effective AIDS vaccine is the lack of an animal transmission model that mimics natural routes of mucosal infection in humans. Our objective is to develop a novel simian-immunodeficiency virus (SIV)/rhesus monkey penile infection model that can serve as a preclinical tool to evaluate intervention strategies that prevent HIV-1 infections and provide a framework to enable fundamental discoveries in acute mucosal pathogenesis in the male genital tract. We will first establish a model of penile transmission and characterize the clinical outcome of SIV infection in rhesus monkeys that acquired SIV via the penile mucosa. We will then validate this model for HIV-1 vaccine and pathogenesis research by characterizing the transmitted viral variants. Finally, we will explore the acute pathogenesis of viral infection and dissemination in the male genital mucosa using this penile infection model. Through a new research partnership, this proposal brings together expertise across two institutions with unique strengths in virology, immunology, pathology, and veterinary medicine. This work represents the synergy of skill sets of two experienced researchers and may yield important insights on HIV-1 pathogenesis that can guide the design of effective prevention strategies to block HIV-1 mucosal transmission. These pilot data will form the basis of a NIH R01 application exploring the mechanisms of HIV-1 mucosal pathogenesis and testing vaccination/microbicide strategies to prevent mucosal transmission in the male genital tract.