NSURP Project 1

Title: Effect of TRPV4 channel mutation on RNA splicing in patients with metatropic dysplasia

Mentor: Vicky Funanage, PhD

Project Description:

Background:
Pathogenic TRPV4 mutations have been identified in a number of musculoskeletal conditions, including dominant brachyolmia, Kozlowski type spondylometaphyseal dysplasia, metatropic dysplasia (MD), Maroteaux type spondylo-epiphyseal dysplasia, and parastremmatic dysplasia, as well as in several neurological disorders, including hereditary motor and sensory neuropathy type IIC, congenital spinal muscular atrophy Charcot-Marie-Tooth 2C (CMT2C), and scapuloperoneal spinal muscular atrophy (SPSMA).

Hypothesis:
The transient receptor potential vanilloid 4 (TRPV4) cation channel mediates calcium influx in tissues in response to physical, chemical, and hormonal stimuli. Differential expression of splice variants based on cell type may play a role in pathology of metatropic dysplasia.

Specific Aim:
To determine the effect of TRPV4 mutations in fibroblast, chondrocyte, and bone cell lines from control and MD patients on expression of known five splice TRPV4 mRNA variants. In addition, characterize the effect of the differing TRPV4 splice variants and severity of the metatropic dysplasia.

Methodology:
Our cohort of MD patients have previously been characterized by sequencing of their DNA (blood or tissue), but the effect of TRPV4 mutation on mRNA expression in these patients needs to be studied further. There are five known TRPV4 splice variants, previously characterized only in tracheal epithelial cells. This study will be two-fold: 1) to first determine if any of the alternative splice variants are found in cell lines established from control and MD patient skin, cartilage, and bone cells; 2) characterize any effect of the TRPV4 mutation on expression of the mRNA variants.
Title: Validity and reliability of automated hearing test in children

Mentor: Kyoko Nagao, PhD

Project Description:

Background:
Hearing loss is a very common problem among children. The audiometric hearing test is a standard test to evaluate peripheral hearing sensitivity. Recently many computer-based programs became available to perform automated hearing tests outside the audiology clinic, such as in schools. However, the reliability and validity of automated tests have not been well-established for pediatric population.

Hypothesis:
Previous literature largely based on adults suggests that automated audiometry performed outside of an audio booth can provide accurate hearing screening results. We expect that automated audiometric results will be comparable to the conventional hearing results in children. However, hearing threshold differences between two tests may be larger in younger children.

Specific Aim:
The aim of the study is to evaluate the validity and reliability of automated hearing screening tests among children compared to regular hearing tests performed in an audiological setting.

Methodology:
Children (typically developing and children with hearing loss) (age range 6 to 12 years) will be recruited from Nemours/ Alfred I. duPont Hospital for Children for this study. All participating children are required to be able to engage in a regular hearing test. Their hearing sensitivity will be tested at 0.5, 1, 2, 4, and 8 kHz with both automated hearing screening application and conventional audiometric test. We will employ a counterbalanced design to control order effects. A 2-way contingency table will be created to compare the performance of the automated test to the standard diagnostic pure-tone audiometry.
NSURP Project 3

Title: Antimicrobial stewardship interventions in a large primary care network

Mentor: Lori Handy, MD

Project Description:

Background:
Antibiotic use is common in pediatric primary care, with 1 of every 5 pediatrician visits resulting in an antibiotic prescription. While antibiotics are needed to treat infections, they often are prescribed for conditions that don't require antibiotics, or a medication that is too broad for the condition is prescribed. This leads to antibiotic resistance in the population, and adverse effects for the patient such as allergic reactions, gastrointestinal upset, or Clostridium difficile infections.

Hypothesis:
Substantial variation in use of broad-spectrum antibiotic prescriptions will be identified as baseline measures prior to antimicrobial stewardship interventions, after controlling for patient and provider characteristics.

Specific Aim:
To explore variability in use of broad-spectrum antibiotic prescriptions within an outpatient pediatric practice network in order to identify targets for an antimicrobial stewardship intervention

Methodology:
We will conduct a retrospective cohort study of children prescribed antibiotics in our pediatric practice-based research network, followed by a pilot study of three ASP interventions. The student will utilize encounter- and patient-level data in the EHR to determine rates of broad spectrum antibiotic prescriptions to provide specific targets for ASP interventions.
NSURP Project 4

Title: Improving outcomes from pediatric cardiac arrest (ICU-RESUS)

Mentor: Shirley D. Viteri, MD

Project Description:

Background:
This study is focused on improving the outcomes from pediatric cardiac arrests that occur within the Pediatric Intensive Care Unit and Cardiac Intensive Care Unit. Part of the improvement process will include likely implementation of the ICU-RESUS bundle which focuses on practicing cardiopulmonary resuscitation, a key skill that all pediatric healthcare providers should possess.

Hypothesis:
1. There will be at least a 27% relative increase in survival with favorable neurological outcome at hospital discharge following implementation of the ICU-RESUS bundle.
2. There will be at least a 50% relative increase in the proportion of minutes with high quality CPR provided during ICU cardiac arrest resuscitations.

Specific Aim:
Specific Aim 1. Evaluate the effectiveness of the ICU-RESUS interventional bundle to improve survival with favorable neurological outcome at hospital discharge in children treated for an ICU cardiac arrest.
Specific Aim 2. Evaluate the effectiveness of the ICU-RESUS interventional bundle to improve the quality of CPR provided by ICU healthcare providers for children during an ICU cardiac arrest.

Methodology:
Patients who receive cardiopulmonary resuscitation will be identified by the treating clinicians and will notify the study team. Data collection will include obtaining patient characteristics from the electronic medical record, obtaining hemodynamic variables from bedside monitors, and CPR quality variables from defibrillators. Implementation of the ICU-RESUS bundle will include bedside point of care cardiopulmonary resuscitation training and monthly multi-disciplinary debriefings of actual cardiac arrests.
Title: Value of diffusion-weighted imaging for diagnosis of acute appendicitis in children

Mentor: Grace Guo, MD

Project Description:

Background:
Acute appendicitis remains the most common acute surgical condition of the abdomen and is the most commonly misdiagnosed condition in the pediatric age group. MRI has become widely used in children and does not require ionizing radiation and iodinated contrast media.

Hypothesis:
Diffusion-weighted (DW) MRI is a noninvasive technique capable of probing the micro-environment of tissue by measuring water movement. If water molecules are restricted in their motion because of cell membranes or, in the case of free fluid, by high viscosity, the signal intensity is high. We hypothesize that DWI performed alone will be an effective diagnostic technique for acute appendicitis as conventional MRI with DWI.

Specific Aim:
The aim of our study is to determine the accuracy of DWI for the diagnosis of acute appendicitis in children comparing with conventional MRI.

Methodology:
Over 500 appendix MRI examinations from November 26, 2013, through March 30, 2016 will be retrospectively reviewed. Two groups of patients (one with conventional MRI including DWI and another one with DWI only), all of who have conventional MRI studies and DWI, and who have confirmed clinical findings, positive or negative for appendicitis will be reviewed. Four radiologists will be divided in two teams of reviewers. Each team will be given a unique set of deidentified cases consisting of both conventional MRI with DWI and DWI only. Each team will receive different cases. Then, each team will read the other team’s set of cases. Sensitivity, specificity and accuracy will be calculated for combined conventional MRI with DWI and DWI only for the depiction of acute appendicitis.
Title: Can occupational therapy tests detect change in hand use in children with cerebral palsy?

Mentor: Jennifer Ty, MD and Nancy Lennon

Project Description:

Background:
Children with cerebral palsy (CP) demonstrate disorders of movement that often lead to limitations in hand use and reduced ability to perform daily tasks of childhood. Occupation therapy (OT) is an important component to advance fine motor skill performance in the school aged child with CP. The QUEST is a standardized tool intended to evaluate the quality of upper limb movement in children with CP and has evidence of good reliability and validity. The SHUEE is a video-based tool used to evaluate pathological movement components and the extent to which the affected hand is used in task completion. Both the QUEST and the SHUEE are intended to guide treatment decisions and evaluate outcomes in an objective and reproducible manner. The aim of this study is to examine the ability of the QUEST and the SHUEE to detect change in hand use in a group of children with CP who’ve had OT, splinting, Botox, or surgery.

Hypothesis:
1. The SHUEE will demonstrate good ability to detect change in upper limb position after Botox or Surgery and in spontaneous arm use after OT.
2. The QUEST will demonstrate fair to good ability to detect changes in grasp pattern and arm support after hand splinting or OT.

Specific Aim:
The specific aim of this study is to examine the ability of the QUEST and the SHUEE to detect change in hand use in a group of children with CP who’ve had OT, splinting, Botox, or surgery at AIDHC.

Methodology:
A retrospective chart review will identify patients with CP age 2 to 18 who have performed QUEST or SHUEE testing before and after an upper limb intervention here at AIDHC. We anticipate a sample of 30 patients will meet the criteria for data analysis. Standardized response means (SRM) and effects sizes will be calculated to measure responsiveness of the QUEST and SHUEE. We will examine patient age and classification, intervention time frames and strategies, and sub-test domains to look for responsiveness trends.
NSURP Project 7

**Title:** Define the binding parameters that govern the sensitivity of chimeric antigen receptors to tumor antigens

**Mentor:** Zhengyu (Mark) Ma, PhD

**Project Description:**

**Background:**
As a targeted immunotherapy approach, adoptive T cell therapy (ACT) redirects T cell toxicity to tumors and holds great promise for treating pediatric cancers such as leukemia and brain tumors. A key component in ACT is the chimeric antigen receptor (CAR) that when expressed on T cells, binds to tumor antigen highly expressed on cancer cells and signals T cells to kill. Since most tumor antigens are also expressed in certain normal tissues at low levels, a major challenge in ACT is to fine tune the sensitivity of CARs so that only tumor cells expressing high levels of tumor antigen are targeted by T cell toxicity.

**Hypothesis:**
We hypothesize that CARs with moderate binding strength (affinity and avidity) to tumor antigens have the suitable sensitivity to mediate potent T cell attack on tumor expressing high levels of tumor antigens while sparing normal tissues expressing tumor antigens at low levels.

**Specific Aim:**
We will construct CARs that bind to the tumor antigen Her2 with varying binding affinity and valency (as monomers or dimers). We will determine the sensitivity of CARs to Her2 using cell lines expressing Her2 at varying levels.

**Methodology:**
Using intracellular staining and flow cytometry, we will determine T cell cytokine production and cytotoxicity in response to target cells expressing Her2 at different levels at the single cell level.
**NSURP Project 8**

**Title:** Genetics of rare pediatric diseases

**Mentor:** Katia Sol-Church, PhD

**Project Description:**

**Background:**
The lab works in collaboration with Nemours clinicians to discover new genes causing dysmorphology syndromes, as well as discover biomarkers that can predict clinical outcomes (genotype/phenotype/metagenomic correlations) for other pediatric disorders.

**Hypothesis:**
Our hypothesis is that most rare disorders are caused by mutations found in the coding regions of important genes, and that the microbiome may at time have a modifying effect on gene/environment interactions.

**Specific Aim:**
Our primary aim is to find causative variants in children with dysmorphology syndromes that are seen by clinicians in the Division of Medical Genetics. We have assembled a series of patients with syndromes of unknown etiology. The student will join the multidisciplinary team to analyze and validate hits discovered by whole exome sequencing and targeted next generation sequencing. Additional projects will include 16S ribosomal gene sequencing to determine how the metagenome may influence disease susceptibility in other pediatric disorders.

**Methodology:**
All basic and advanced molecular technologies will be available for this project including as needed. This includes nucleic acid isolation and quantitation, DNA sequencing and genotyping, profiling using STR analysis and data visualization using biocomputing analysis.
**Project Description:**

**Background:**
Siblings of children with cancer are exposed to significant stress after their brother or sister’s diagnosis. Concern about the ill child, disruptions in family roles and routines, decreased contact with family members, as well as added demands for care giving or other responsibilities in the home are common.

**Hypothesis:**
These unique challenges leave siblings of children with cancer at risk for acute and long-term psychosocial difficulties. Although severe psychopathology is rare, several reviews suggest that some siblings exhibit symptoms of anxiety, depression, post-traumatic stress, lower quality of life and disruptions in academic and social functioning. Siblings can also demonstrate resilient outcomes. We hypothesize that there are a set of identifiable trajectories of distress experienced by siblings of children with cancer and that specific risk and protective factors are associated with these trajectories.

**Specific Aim:**
1) To identify and describe the various trajectories of sibling adjustment to childhood cancer (cancer-related traumatic stress symptoms) that manifest during the first two years following diagnosis.
2) To determine what factors (e.g., demographic variables, cancer- and treatment-related variables, parental distress, parent-child relationship, family functioning, social support, extended family/school/community engagement) distinguish siblings with resilient adjustment trajectories from those with sustained or escalating distress.

**Methodology:**
Approximately 200 families of children with cancer with a healthy sibling between the ages of 8 and 17 will be recruited for participation from the enrollment rosters of SuperSibs powered by Alex’s Lemonade Stand Foundation. Eligible families will be within 1 year of diagnosis at the time of enrollment and participants (one parent and one sibling per family) will complete a survey at three time points within the first two years post-diagnosis. The survey includes measures of cancer-related traumatic stress, positive and negative mood, parental monitoring and warmth, family functioning, perceived social support, extended family support, and engagement in school and community activities, as well as measures of demographics and cancer diagnosis and treatment-related factors.
NSURP Project 10

Title: In-vitro studies of the fetal gubernaculum.

Mentors: Julia Barthold, MD and Jason Gleghorn, PhD

Project Description:

Background:
The “testicular dysgenesis syndrome” (TDS) comprises 2 reproductive phenotypes considered fetal in origin: spermatogenesis defects and testicular germ cell tumor (TGCT), and 2 birth defects: cryptorchidism and hypospadias. Controversy exists regarding the degree to which risk factors are shared among these disorders. Cryptorchidism is one of the most commonly encountered birth defects, occurring in 2-4% of boys, but genetic risks seem much weaker than for other TDS phenotypes, with population data suggesting the importance of the maternal environment, and only moderate heritability of the trait. The increasing concern that ubiquitous endocrine-disrupting chemical (EDC) exposure contributes to TDS susceptibility is plausible and is a potential public health concern.

Hypothesis:
The gubernaculum directs fetal testicular descent via swelling and migration. Activation of the androgen receptor (AR) plays a pivotal role in this process having its affect directly on the fetal gubernaculum. We hypothesize that functional gubernaculum-testis co-culture models will provide insight into AR signaling mechanisms and the potential additive anti-androgenic activity of endocrine-disrupting chemical (EDC) mixtures in vitro.

Specific Aim:
Develop static and dynamic co-culture models to study AR signaling targets in the fetal gubernaculum.

Methodology:
Partnering with the Gleghorn Lab at University of Delaware, we will develop a fetal gubernaculum-testis co-culture model in soft agar, and a microfluidic platform to study pressure-induced gubernaculum inversion. After growth in culture, gubernacula will be processed for analysis. The student will be involved in various experimental protocols including observation with the potential for hands-on microdissection of fetal tissue; 3D organ culture and imaging of cultured tissues; tissue clearing, confocal imaging and 3D image analysis to determine muscle/mesenchymal volume ratio and AR+ cellular density; and/or measurement of fetal organ stiffness using micropipette aspiration.