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"Mechanism of STING Signaling and Auto-Activation during Pediatric Inflammatory Disease"

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The cellular receptor STING (Stimulator of Interferon Genes) controls a human immunity pathway that allows cells to sense pathogen– and tumor–derived DNA. Uncontrolled activation of STING signaling has emerged as a direct cause of childhood autoimmunity. STING–Associated Vasculopathy with onset in Infancy (SAVI) is a rare disease caused by gain–of–function mutations in the receptor that induce constitutive signaling and systemic immune activation. Children with SAVI exhibit chronic inflammation, severe necrotizing vasculitis, and neurological impairment. Specific therapies for SAVI disease do not currently exist, and afflicted children rarely survive past childhood. Importantly, the lack of insight into the function of SAVI mutations limits treatment, and demonstrates that our current models of STING signaling are incomplete.

In spite of the clear role of STING activation in immune cell responses, STING signaling is poorly understood, including a gap in knowledge of how the receptor engages downstream factors to initiate signaling. I have used a biochemical and structural approach to understand STING activation, and my lab is now uniquely poised to determine how patient mutations cause disease. All SAVI mutations map within the receptor dimeric interface, and we hypothesize that these mutations artificially "lock" an active signaling conformation.

To test this model, we will use a genetic approach to define the protein interaction motifs required for STING to recruit downstream signaling kinases (Aim 1). Additionally, we will determine the structural impact of SAVI mutations on STING conformation and biochemically define how mutations disrupt regulation of kinase recruitment (Aim 2). The long–term goal of my lab is to understand how STING signaling can be manipulated to rationally tune human immune responses. Identifying the function of SAVI mutations will provide a unique framework to develop a new model of STING activation, and offer the opportunity to discover treatments that mitigate childhood autoimmunity disease.
Human development requires exquisite spatial and temporal control of the proteins that are expressed in a cell. Translation initiation factors are key players in protein synthesis, and although alterations to these factors are found in a wide array of genetic diseases, including cardiac or craniofacial disorders, cancer, autism, and diabetes, the molecular basis for the resulting disease is poorly understood. The overarching goal of my research is to discover how mRNA translation regulation drives the dynamic gene programs during organismal development and why childhood diseases result when this is dysregulated.

We recently discovered that certain initiation factors moonlight in roles outside of general translation to control the translation of select transcripts, thus presenting a potential mechanism by which these proteins could drive tissue-specific development. In particular, the 13-subunit eukaryotic initiation factor complex eIF3 plays a major role in balancing cell proliferation and differentiation by mediating specialized translation of ~500 mRNAs, through translation activation of cell growth regulation mRNAs such as Jun and translation repression of myogenic factors like BTG1.

These surprising findings lead to a number of important questions: How is the activity of eIF3 in specialized translation regulated? What are the mRNAs controlled by eIF3 during development? What is the subsequent effect on development when eIF3 activity is dysregulated? We will answer these questions using an integrative approach combining sequencing and computation analysis, RNA–protein biochemistry, and genetics. Our immediate goal is to provide a molecular understanding of eIF3–specialized translation during development; while our long-term objective is to address why incorrect expression of translation initiation factors leads to congenital diseases. These studies will illuminate the importance of translation regulation during development and provide insight into why detrimental childhood diseases result when this regulation fails.
Pseudomonas aeruginosa induced chronic pulmonary infection is a major cause of death in over 30,000 children affected by cystic fibrosis (CF) in the US alone. Bacteria strongly attach to lung cells via specialized protein appendages called pili and form biofilms that are difficult to eradicate, resulting in recurrent hospitalization and mortality. Currently there are no effective methods to selectively target P. aeruginosa pili to block infections.

Previous studies have suggested that pathogenic bacteria alter electrical charges on pili for cellular interaction and immune recognition. However, existing technology cannot directly study their effects. We have developed new techniques for quantitative imaging of static and dynamic charges in pili. Pili were previously considered as non-conductors. However, using these new methods, we have found that pili of soil bacteria are electrically conductive and conductivity is important for bacterial metabolism.

Using our new methods we plan to analyze a spectrum of interactions between both a host surface and a pathogen. Particularly, we will employ atomic force microscopy to measure nanoscale forces between the host–cell surface and individual P. aeruginosa and determine the role of charge interactions in bacterial infection. Our studies will provide a strong foundation to develop novel anti-microbial therapies that could suppress bacterial infections in CF by neutralizing host–pathogen charge interactions.
Improving the continuum of care for neonates born in rural settings in low- and middle-income countries is a pressing public health concern. The central hypothesis of this project is that lay midwives in Guatemala can serve as an efficient means for rapid evaluation of medical referral of neonates when indicated, if they are provided with decision support and formal linkages to the medical referral chain. In this project, we will accomplish this by designing a novel phone-based mHealth application which provides midwives with real-time decision support and access to on-call clinicians to guide them through the examination and triage of neonates in the first week of life. The mHealth implementation will be embedded within a larger quality improvement framework designed, in partnership with a local primary health care organization in Guatemala, to reduce barriers to neonatal referrals. The project has two Specific Aims:

Specific Aim 1: To develop a smart-phone based decision support system for early neonatal evaluation by lay midwives.

Specific Aim 2: To use a QI framework to evaluate the impact of the use of the decision support application by a cohort of lay midwives on improving the neonatal referral rates.

The primary outcome will be an increase in the proportion of neonates referred to a higher level of care. Secondary outcomes include the proportion of neonates receiving timely evaluation by midwives and the incidence of neonatal death/adverse events. After an iterative, adaptive design process to optimize the mHealth application, a quality improvement team will oversee the mHealth implementation over 12 months. Statistical process control methodology (control charts) will be used to track improvements in the neonatal continuum of care throughout the implementation and to document achievement of the primary and secondary outcomes.
Otitis media (OM) is the most commonly diagnosed pediatric disease and the #1 reason for antimicrobial prescription to US children. Moreover, 62% of children with OM demonstrate viral infections in their middle ear, to which antibiotics are ineffective but prescribed nonetheless. The widespread use of systemic antibiotics against a disease of such high prevalence and recurrence is believed to breed antibiotic resistance. To avoid systemic antibiotic exposure, we have developed a platform that can non-invasively overcome the impermeable barrier of the tympanic membrane (TM) and sustainably deliver antibiotics directly to the middle ear. The current application attempts to completely eliminate antibiotic usage in this prevalent childhood disease and to mitigate antibiotic resistance by using a stand-alone therapy that treats bacterial and viral infections.

I propose a therapeutic system that continuously generates hypohalites in situ, mimicking the vanadium haloperoxidase of seaweeds, using vanadium pentoxide nanowires (V2O5-NW). V2O5–NW catalyzes oxidation of halides to hypohalites using hydrogen peroxide (H2O2). Compared to haloperoxidase, V2O5–NW has better stability, high reactivity with H2O2 at very low concentration (>100 nM), and scalability for industrial production. Unlike other inorganic catalysts, V2O5–NW performs well under physiological conditions (no need for organic solvents, or extreme temperature or pH).

Given the paucity of biomedical experience with hypohalites, studies of biocompatibility, toxicity, and biodistribution (especially across the round window, and into the inner ear) will be important. We note that vanadium is present in many foods, and hypochlorite ion is generated by human myeloperoxidase during the oxidative burst of activated neutrophils. Damage to hair cells by reactive oxygen species comparable to hypochlorite has been reported at much higher concentrations (1–10 mM) than I intend to use.