SPECIFIC AIMS

Congenital malformations of the genitourinary tract carry with them significant morbidity and increased risk of mortality for individual patients, and is a growing public health burden. At present, there are limited therapies available to ameliorate the progressive loss of genitourinary tissue. A comprehensive understanding of how the genitourinary organs (including kidney and urinary tract) develop in utero is necessary to effectively develop novel therapies to replace or repair injured tissue. The Genitourinary Development Molecular Anatomy Project (GUDMAP) has been extraordinarily successful at providing a high-resolution map of gene expression in the murine genitourinary system [1-8]. However, a similar description has not been available for the developing human genitourinary system, nor has it been possible to develop optimized experimental techniques to grow, expand and differentiate human genitourinary progenitor cells in vitro. These research efforts by the developmental community have been hampered by the lack of a central hub for the procurement, and distribution of high quality human genitourinary samples.

The Health Sciences Tissue Bank (HSTB) at the University of Pittsburgh has been involved in research human tissue procurement for over 18 years: collecting, maintaining and disbursing quality samples to research scientists, both in HSTB is embedded within the Department of Pathology of the University of Pittsburgh Health Systems; thus providing rapid access to very high quality tissue and biological specimens. HSTB has established consenting protocols in line with the best practices recommendations from the NIH, a strong informatics backbone to facilitate specimen procurement and annotation, and has in place robust quality control and quality assurance programs. The HSTB biorepository is fully accredited by the College of American Pathologists (CAP). HSTB has an established program accruing fetal tissues that has been IRB approved since 2005. In this calendar year, we have disbursed over samples collected from The collections can be significantly ramped up as material could have been accrued from as many as last year.

We have preliminary data that we can isolate human genitourinary tissues (kidneys, ureters and bladders) from various developmental ages (6-24 weeks). We have produced publication quality histological images of the developing urogenital organs (including kidneys and bladder), and have immunostained kidneys for endothelium, nephron progenitors and early-differentiated nephron structures and bladders for the urothelium and muscle layers. We have utilized Dynabeads® to separate distinct cellular subpopulations in the kidney including: nephron progenitors, ureteric epithelium, podocytes and endothelium, and have confirmed that we can produce high quality material that is appropriate for RNA sequencing. We propose to act as both the GUDMAP Tissue Hub and Tissue Gathering site to build upon the pre-existing HSTB and provide top quality genitourinary samples to members of the scientific community including those within GUDMAP.

Aim 1: To generate an inventory of genitourinary tissue throughout normal human development

The main goal of this aim is to develop a pipeline for the acquisition, quality control and distribution of human genitourinary samples obtained throughout development (6-42 weeks gestation). We currently have access to 6-24 week samples through the HSTB. However, for later gestational stages (25-42 weeks gestation) we have partnered with the International institute for the Advancement of Medicine. This will provide access to a novel resource for neonatal donation. We aim to collect and store a minimum of per developmental week. Each of these samples will have histology, immunohistochemistry and in situ hybridization performed to assess tissue quality, protein and RNA integrity. Furthermore, we will obtain maternal blood, urine and amniotic fluid; based on the clinical situation and ability to procure. Based on our current experience, we get these biological materials in most cases. Anonymized demographic information of each specimen will also be provided.

Aim 2: To provide fresh genitourinary tissue and biological research specimens

This aim will generate an ongoing resource to distribute fresh developmental human genitourinary samples from various stages (6-42 weeks) to the GUDMAP Atlas projects. The samples will be procured by a pathologist and inspected for mechanical damage. Samples will be collected from all qualified cases. The samples will then be subdivided based on the demand for fresh/frozen aliquots; the validation laboratory for quality control will keep a portion of each sample. The tissue samples will be immediately sent out for live cell use or immediately separated into distinct cellular populations before shipping based on researcher demands. Permissible annotating information; including demographics of each specimen, will also be provided.