Abstract of proposed student project (1 page limit. This should mirror the aims page of a grant and CLEARLY indicate the student's role.)

Urothelial Cell to Sensory Neuron Signaling in Bladder Pain.

An estimated 2.7% of women globally suffer from bladder dysfunction and pain caused by interstitial cystitis/bladder pain syndrome (IC/BPS)[1]. There are no durable or effective treatments for most IC/BPS patients due to poorly understood pathophysiology limiting the development of new therapeutics. One pathophysiologic feature of IC/BPS is the disrupted function of urothelial cells[2-4]. Urothelial cells, the endothelial cells that line the bladder, provide a protective barrier to contain urine, respond to mechanical/chemical stimuli, and transmit signals to other cell types within the bladder[5]. Patients with IC/BPS have abnormal urothelial cell gene expression, cell differentiation, and cell signaling, including increased adenosine triphosphate (ATP) release after stimulation[4,6-9].

ATP plays several roles in the bladder as a signaling molecule, including bladder nociception and bladder sensory signaling[10,11]. Thus, ATP is an attractive target for intervention in IC/BPS. However, due to the heterogeneous population of cell types in the bladder wall (urothelial, interstitial, resident immune cells, and sensory afferents) that express ATP purinergic receptors sub-types and influence nociception[5], it is difficult to determine which cell/tissues and pathways to target. To address this challenge, we propose to use an innovative optogenetic approach to test the hypothesis that urothelial cell released ATP is critical for bladder nociceptive signaling (**Fig. 1**).

Optogenetics and transgenic cre mouse line development have made it possible to develop animal models to selectively modulate a specific cell type within a heterogeneous population of cells using light[12]. Recently, this technique has been used in non-neuronal cells, such as keratinocytes and colon epithelium, to delineate their role in signaling directly to sensory neurons[13,14]. We have developed a mouse model that expresses the non-selective and light-activated cation channel, channelrhodopsin2 (ChR2), in urothelial cells. With this strategy, we can stimulate urothelial cells directly and evaluate their effects on sensory signaling and nociception. Pairing these approaches with our newly developed implantable wireless light delivery devices will allow us to modulate urothelial cells in awake non-restrained animals directly[15-17]. Using this new mouse model, we will test the central hypothesis that activation of urothelial cells and subsequent release of ATP modulates sensory neuron activity and influence bladder nociception. We will test this hypothesis with the two aims:

Aim 2: Evaluate the nociceptive responses to selective activation of urothelial cells in naïve and cystitis mice. Urothelial cells can release several factors that may directly activate or sensitize nociceptive sensory neurons. Consistent with this, our exciting preliminary results indicate that urothelial stimulation enhances nociceptive responses. In this aim, we will evaluate the effect of urothelial stimulation on bladder nociception using an implantable wireless light source to activate opsins expressed in the urothelial cells locally. The results from this proposal will aid in furthering our understanding of urothelial signaling in pain detection (e.g., nociception). Further, we anticipate that the knowledge gained from this proposal will lead to more effective drug targets and contribute to the overall goal of increasing the quality of life in patients suffering from IC/BPC.

The FSVP student on this project will help perform device implantation and assessments of pain and bladder (8-10 animals) function during urothelial stimulation following implantation. There a several behavioral assays we use to assess these sensory functions and there is significant time needed for data analysis as well. The student will also remove bladder tissue following the experiment to evaluate the tissue device interface for fibrosis and inflammation.