A Newly Identified Chromatin Remodeling Pathway in the Lens Enables a Proof-of-Concept Gene Therapy for Posterior Capsule Opacification

Xiang, Evan (School: Shady Side Academy)

Posterior capsule opacification (PCO) is a major complication of cataract surgery, driven by the epithelial-to-mesenchymal transition (EMT). In this study, I investigated the mechanistic effects of a common class of drugs used for diabetes treatment, Dipeptidyl Peptidase-4 Inhibitors (DPP4i), to develop a proof-of-concept gene therapy for PCO by targeting a novel chromatin remodeling pathway. I show that DPP4 inhibitors attenuate the EMT through potentiation of the p38/MAPK protein ($p<0.001$), resulting in downstream alteration of chromatin topology. Notably, through Hi-C sequencing, I show that SNAI1 is inhibited by separation from its primary enhancer in-cis and SMAD2 is inhibited through compartmental shifting. However, this study also revealed that DPP4 inhibitors have significant ocular off-target effects, making DPP4i treatment non-ideal for long-term treatment. To ameliorate this, I developed a proof-of-concept gene therapy via CRISPR-Cas9 that targets the same novel functional mechanism through insertion of a cHS4 insulator, while avoiding the substantial cytotoxicity of DPP4 inhibitors. Initial lipofectamine transfection in-vitro shows similar inhibition of EMT with cell viability increasing by more than 20% ($p<0.01$). GUIDE-seq confirmed minimal off-target gRNA activity. Thereafter, a potential delivery solution was developed and proposed by packaging eSpCas9 construct ribonucleoproteins into antibody-conjugated lipid nanoparticles coated in hyaluronic acid to facilitate delivery in future murine studies. My results here uncover a novel signaling pathway in the lens through a common drug, which I then exploit to develop a therapeutic for prevention of a complication of cataract surgery.

Awards Won: