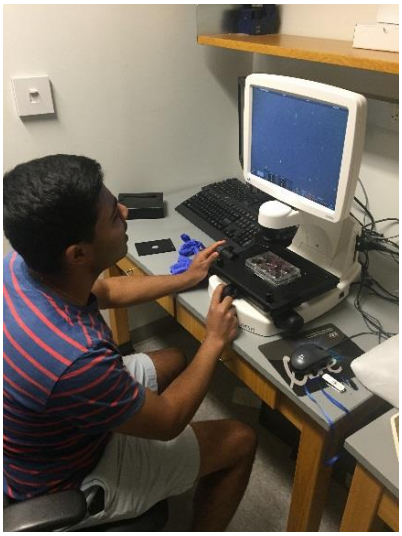

Understanding the Mechanisms of Mammalian Tissue Regeneration
Hari Magge (COL 2018)
Advisor: Thomas Leung

This summer, I worked in the lab of Dr. Thomas Leung in the dermatology department of the Perelman School of Medicine, studying the mechanisms of mouse tissue regeneration. In mice and other mammals, traumatic injuries heal with a strong fibroblast and collagen response, producing a scar. Although less dramatic, there are limited examples of mammalian tissue regeneration or wound healing without a scar. Our lab previously developed a mouse model where injured mouse ears regenerate tissue without a scar (Leung et al. 2015). My first goal addressed the molecular mechanisms that determine this switch between skin regeneration and scar formation. At injury sites, increased expression of SDF1 promotes scar formation. I wanted to find proteins that modulate SDF1 expression in wounded mice.

The second part of my project was to use CRISPR/cas9 technology to knock out a segment of the mouse genome and replace it with a green fluorescent protein (GFP) insert. I used GFP because it shows green fluorescence when exposed to light within a certain range; presence of green cells confirms success of the experiment. CRISPR/cas9 technology can be used to cut a cell's genome at a specified location, allowing for precise genome editing. I used CRISPR/cas9 technology to remove a 22 base pair sequence of the mouse genome 1.6 kb before the transcription start site for SDF1. I targeted this specific site because it is 100% conserved between mice and humans, which indicates that it may have an important role in the regulation of SDF1. In place of the removed portion, I constructed and inserted a GFP fragment. Under a fluorescent microscope, the cells that had successful replacement of the 22-base pair sequence with the GFP sequence are bright green. These green GFP positive cells will eventually be used to assess SDF1 function.

In order to attempt to accomplish the above goals, I learned how to perform Western Blots to study protein levels, undertake molecular cloning, and use CRISPR technology, which are all research mechanisms that will serve me well in my future research endeavors. My experience with PURM has given me practical experience to complement my learning in the classroom, which is why I will continue my research with Dr. Leung during the academic year.