BCHM 421/422 – 2019/2020

Project Outline:

Pulmonary arterial hypertension (PAH) is a disease of occlusive vascular remodeling in the pulmonary circulation that causes right heart failure and death within 3-5 years of diagnosis[1]. Mutations in *BMPR2*, the gene encoding the bone morphogenetic protein (BMP) type II receptor (BMPR-II) account for 70-80% of heritable PAH and roughly 20% of seemingly idiopathic cases of disease[2]. Previous work by our group has demonstrated that signaling of the endothelial-selective[3] BMP ligand, BMP9, is impaired in PAH patients exhibiting reduced BMPR-II expression, resulting in excessive endothelial cell proliferation. We have recently performed next-generation, whole transcriptomic RNA-sequencing on pulmonary endothelial cells, with or without siRNA-mediated silencing of *BMPR2*, to identify the impact of *BMPR2* loss on the transcriptional response of these cells to BMP9 treatment. This screen has identified a number of novel coding and non-coding RNAs that could be driving the hyperproliferative endothelial response to BMP9 in PAH.

The SSP student will be responsible for examining the non-coding RNAs identified through this screen, including long non-coding and circular RNAs, to validate the results of the screen and to determine the contribution of specific RNAs to endothelial function. Techniques include the assessment of gene expression by qPCR and the silencing of candidate RNAs by siRNA, as well as functional assays examining endothelial cell proliferation, migration and apoptosis. This work will be conducted using infrastructure in the Ormiston lab (www.OrmistonLab.com), as well as the Queen's Cardiopulmonary Unit (Q-CPU), a new, $10-million facility for translational research in diseases of the heart, lung and circulatory system.

Supervisor: Mark Ormiston

Project Title: Examining the impact of non-coding RNAs on the phenotype and function of pulmonary endothelial cells following silencing of the bone morphogenetic protein type II receptor

Keywords:
1. Vascular biology
2. Bone morphogenetic protein signaling
3. Non-coding RNAs
4. siRNA silencing
5. Cell culture and functional assays

Project Goals:
1. Validate the differential transcription of non-coding RNAs in endothelial cells following *BMPR2* silencing, with or without BMP9 stimulation.
2. Use siRNA-mediated silencing to determine the contribution of specific non-coding RNAs to the endothelial functional response to BMP9.

Experimental Approaches:
Primary endothelial cell culture, siRNA knockdown, qPCR, endothelial functional assays (proliferation, migration, apoptosis).

References:
1. Benza RL, Miller DP, Barst RJ, Badesch DB, Frost AE, McGoon MD. An evaluation of long-term survival from time of diagnosis in pulmonary arterial hypertension from the reveal registry. *Chest*. 2012;142(2):448-456. doi:10.1378/chest.11-1460.
2. Machado RD, Aldred MA, James V, *et al*. Mutations of the TGF-β type II receptor BMPR2 in pulmonary arterial hypertension. *Hum Mutat*. 2006;27(2):121-132. doi:10.1002/humu.20285
3. Long L, Ormiston ML, Yang X, *et al*. Selective enhancement of endothelial BMPR2 with BMP9 reverses pulmonary arterial hypertension. *Nat Med*. 2015;21(7):777-785. doi:10.1038/nm.3877.