

# BCHM 421/422 Project – 2023/2024

## Project Outline

**Supervisors:** Dr Andrew Craig & Dr Pierre-Olivier Gaudreau

**Project Title:** Develop a lung cancer/T cell co-culture model for testing new combination cancer immunotherapies

**Project Goals:** The goals of this project are to:

- 1) Establish mouse lung cancer cell lines expressing Ovalbumin and a fluorescent reporter for Granzyme B release from cytotoxic T cells
- 2) Test effects of mitotic kinase PLK4 inhibitor in combination with immune checkpoint inhibitor anti-PD1 in lung cancer cells co-cultured with cytotoxic T cells

### **Experimental Approaches:**

- 1) Student will learn how to safely maintain sterile cultures of mouse lung cancer cell lines.
- 2) Student will learn how to transduce mouse lung cancer cells with a lentivirus encoding both chicken ovalbumin (OVA, a model tumour antigen) and a Förster resonance energy transfer (FRET) reporter encoding CFP/YFP separated by a Granzyme B cleavage site (see Ref 1).
- 3) Student will learn how to isolate Ova-specific T cells from splenocytes of OT-1 transgenic mice using a magnetic separation system (2).
- 4) Student will establish conditions for co-cultures with the above cell types in absence or presence of mitotic kinase PLK4 inhibitors and anti-PD1 immune checkpoint inhibitor.
- 5) Student will learn how to use flow cytometry and confocal microscopy to measure the ability of T cells to kill cancer cells in the above culture models (3).
- 6) Some background readings on the topic (4, 5).

### **References:**

1. [G Sharma, CM Rive and RA Holt \(2019\) Rapid selection and identification of functional CD8+ T cell epitopes from large peptide-coding libraries, Nature Communications, 10, 4553.](#)
2. <https://www.stemcell.com/products/easysep-mouse-t-cell-isolation-kit.html>
3. <https://dbms.queensu.ca/cytoflex>
4. <https://pubmed.ncbi.nlm.nih.gov/27461776/>
5. <https://pubmed.ncbi.nlm.nih.gov/33777739/>