

BIOC 462/491 Internship Position in Industry

# Summer or Fall 2024

**Company:** 



Montreal, QC Canada H4S2E1 https://parazapharma.com/

Project Title: Optimization of heterologous protein expression in mammalian cells

### **Project Description**:

### Background/context

Mammalian suspension cell lines have come of age for heterologous production of proteins. Transient transfection of suspension cell lines provides an easy way for research-scale protein production on short time and modest budget.

## Aim/hypothesis

The project aims at evaluating vector elements for protein yield improvement in transient transfection of commercially-available Thermo's cell line Expi293. Specifically, we will focus on elements stabilizing mRNA transcript (Backliwal et al., 2008) and maintaining a plasmid in an episome (Durocher et al., 2002).

### Methodology to be used by the student

The student will construct expression plasmids, introducing different expression-enhancing elements one by one or in combination. A gene of secreted rabbit IgG Fc heavy chain fragment crystallizable and a gene of an integral membrane protein will serve as two very different reporters for expression quantification. Depending on the progress, the student will have an opportunity to master assembly-based cloning using iVEC3 *E.coli* cell line for *in vivo* cloning, plasmid design, mammalian cell culture maintenance, mammalian cell protein expression, membrane and soluble protein quantitation, SDS-PAGE gels and Western blots.

### Relevance of the project

Many laboratories omit mammalian cell expression presuming it is expensive and inefficient. This project, together with other initiatives, will prove high speed and high yield of mammalian cell expression.

### **Contact Information:**

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#### **References:**

- Backliwal G, Hildinger M, Chenuet S, Wulhfard S, De Jesus M, Wurm FM. Rational vector design and multipathway modulation of HEK 293E cells yield recombinant antibody titers exceeding 1 g/l by transient transfection under serum-free conditions. Nucleic Acids Res. 2008 Sep;36(15):e96.
- Durocher Y, Perret S, Kamen A. High-level and high-throughput recombinant protein production by transient transfection of suspension-growing human 293-EBNA1 cells. Nucleic Acids Res. 2002 Jan 15;30(2):E9.