Optimization of Resolubilization and Disassembly of AAV5 Virus-Like Particles

Bachelor Thesis

General Information

PhD project started 01.2025
Supervisors: Giulia Polazzo, Loulotte Waling
Starting from **January/February 2026**
Positions available: 1 BSc position
Language spoken: **English**

1. Introduction

Virus-like particles (VLPs) are nanostructures that mimic the organization and conformation of viruses, but they are noninfectious as they lack viral genetic material. Adeno-associated virus (AAV) vectors are among the most promising tools for gene therapy applications due to their safety and efficiency in gene delivery, but the production is challenging resulting in low yields and high costs. Previous research has demonstrated the potential of precipitation as capture step for VLPs. This project focuses on improving the recovery and purity of AAV5-VLPs by optimizing the resolubilization of precipitated capsid proteins and investigating potential disassembly steps.

2. Research Objectives

- Optimize the resolubilization yield of AAV5-VLPs (VP3-only) from precipitated pellets.
- Investigate the effect of disassembly treatments on VLP recovery and purity.
- Evaluate the impact of different buffer compositions on solubility and particle integrity.
- Assess yield and purity using analytical methods.

3. Methodology

- Optional expression of AAV5-VP3 capsid proteins in *E. coli*.
- Precipitation and recovery of VLPs from crude extracts.
- Screening of different resolubilization buffers and disassembly conditions (e.g., pH, salt concentration, chemical additives), automating the procedure with the liquid handling station *Freedom EVO® 200* (Tecan Group Ltd.).
- Quantitative and qualitative analysis of solubilized fractions to determine recovery efficiency and particle quality.

4. Analytics and Tools

- Robotic liquid handling station (*Freedom EVO® 200*, Tecan Group Ltd.)
- SDS-PAGE electrophoresis system, ELISA kit
- High Performance Liquid Chromatography, UV/Vis spectroscopy, Dynamic Light Scattering

5. Timeline