Large-Scale Production of Exosomes for Targeted Therapeutics

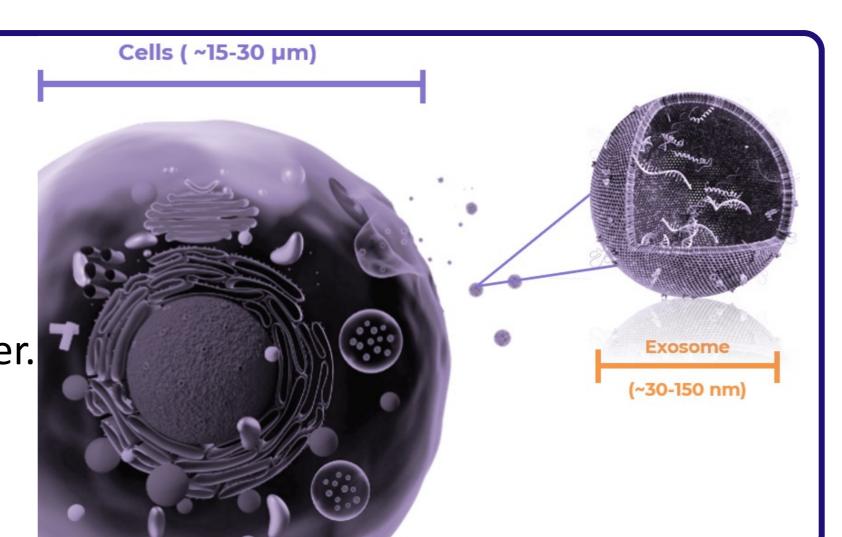
Rebecca Heather, Meryck Mucenski, Rigel Tormon, Phuong (Christina) Trinh, Helen Zhang

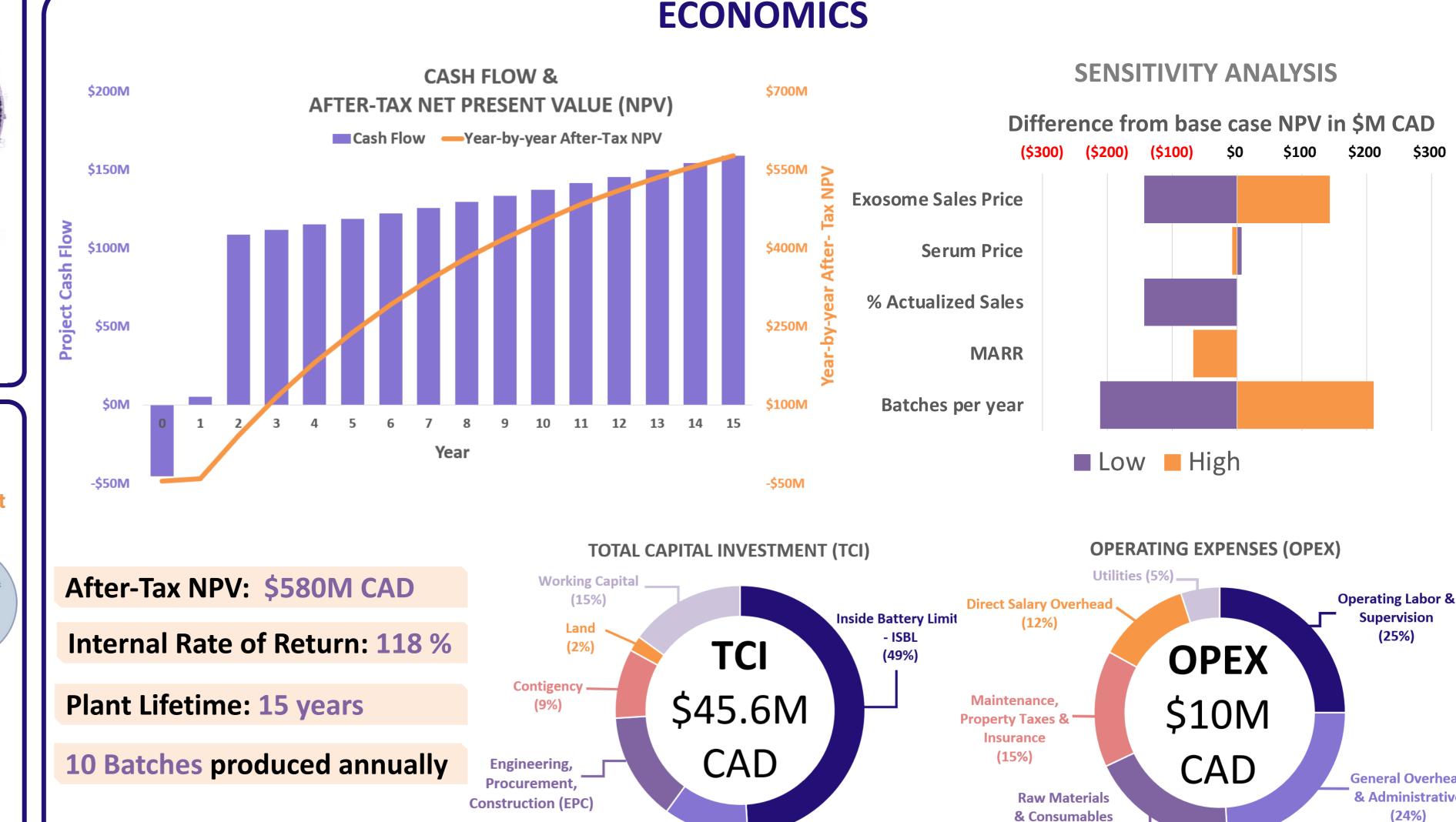
Department of Chemical and Petroleum Engineering

INTRODUCTION^{1,2,3}

Exosomes are extracellular vesicles that are secreted by cells for **cellular communication**. They carry **bioactive cargo** like nucleic acids, proteins, and other metabolites and can precisely deliver them to recipient cells, making them an effective vehicle for **targeted drug delivery** and **disease diagnostics**.

Currently, clinical trials are exploring their use in various diseases, such as diabetes, stroke, and cancer. However, current exosome production methods are cost-ineffective, labour-intensive, and lack scalability. This is where EXOVIA comes in, as we are designing an economically viable process for producing exosomes on a large scale, in our facility based in Calgary.



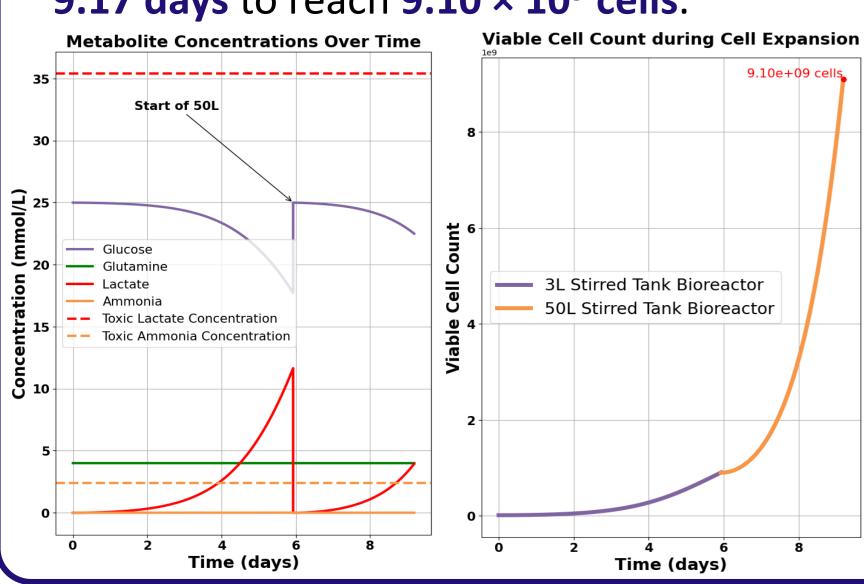


PROCESS OVERVIEW Cryovial T-flask 3L Stirred-tank Bioreactor Bioreactor Bioreactor Filtration Ultrafiltration Formulated Product WSC Upstream ×3 (in parallel) Downstream

UPSTREAM⁴

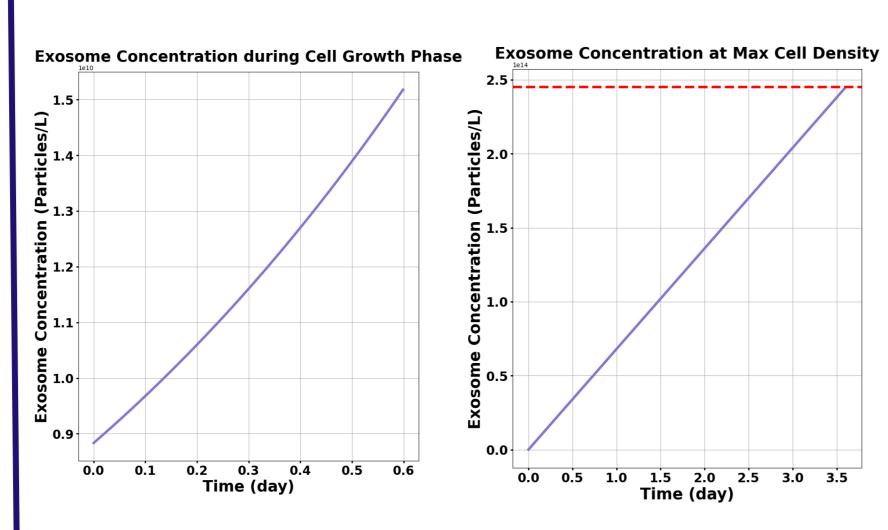
Cell Expansion

Two stirred-tank bioreactors are run in batch mode to multiply the cells and reach the target cell count for seeding onto the hollow fiber bioreactor. This process takes **9.17 days** to reach **9.10** × **10**⁹ cells.



Exosome Production

The hollow fiber bioreactor will take 22.2 days to process one batch with a total of 6 collections where each will have 2.72 × 10¹³ exosomes. One full batch will produce 1.632 × 10¹⁴ exosomes.

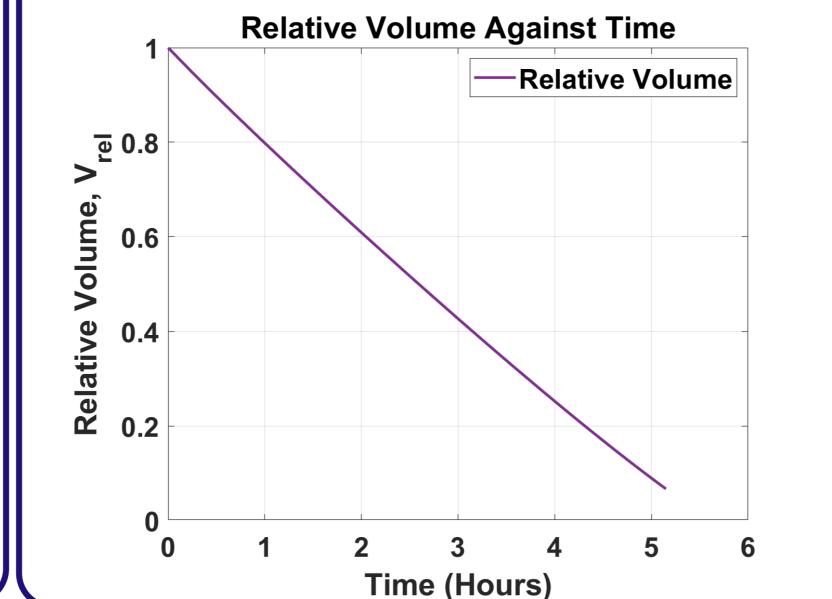


DOWNSTREAM^{5,6}

Tangential Flow Filtration

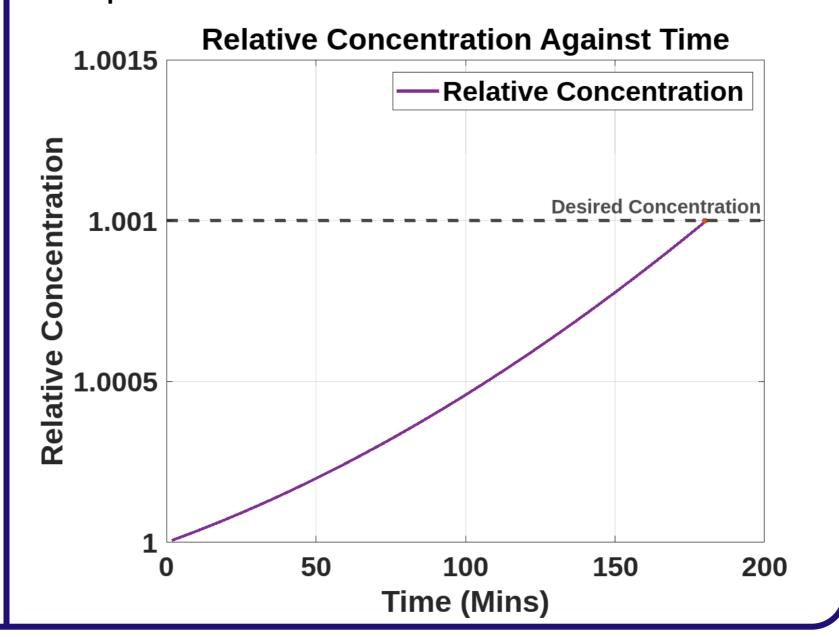
A hollow fiber cartridge will remove excess media and any contaminants smaller than 35 nm in diameter.

The volume will be decreased by 93.33%.



Ultrafiltration

This ultrafiltration device provides the final removal of any contaminants less than 20 nm, purifying the exosome product. One sample from this unit creates 34 doses.



PRODUCT

SCHULICH

School of Engineering

EXOVIA produces 10 high-quality annual batches using a Quality by Design approach and adheres to Good Manufacturing Practices, supplying 14,500 vials for 57 clinical trials across the U.S. and China, capturing a major share of the global exosome trial market.



Acknowledgement

Lonza

We thank **Dr. Michael Kallos** for his consistent guidance and support for the duration of this project. We would also like to extend our gratitude to our industry sponsor, **Dr. Krishna Panchalingam**, from Lonza for his time, expertise, and industrial insights.

ENVIRONMENT, HEALTH, AND SAFETY⁷

Key Safety Hazards:

- Contamination
- Equipment Failure or Malfunction
- Operator Error
- Deviations in Operating Conditions
- Biohazardous Materials & Waste

Solid Waste 38%

Safeguards & Mitigations:

- Operator Training & PPE
- Controls, Sensors, & Alarms
- Equipment Maintenance & Inspection
- HVAC Ventilation
- cGMP Practices
- Clean Rooms

REFERENCES

- 1. Kalluri, R., & LeBleu, V. S. (2020). The biology, function, and biomedical applications of exosomes. Science, 367(6478) eaau6977. https://doi.org/10.1126/science.aau6977
- Exosomes Market Size, Share & Growth Analysis Report, 2030. (n.d.). Grand View Research.
- https://www.grandviewresearch.com/industry-analysis/exosomes-market
 Krueger, T. E. G., Thorek, D. L. J., Denmeade, S. R., Isaacs, J. T., & Brennen, W. N. (2018). Concise Review: Mesenchyma Stem Cell-Based Drug Delivery: The Good, the Bad, the Ugly, and the Promise. STEM CELLS Translational Medicine, 7(9), 651–663. https://doi.org/10.1002/sctm.18-0024
- Lawson, T., Kehoe, D. E., Schnitzler, A. C., Rapiejko, P. J., Der, K. A., Philbrick, K., Punreddy, S., Rigby, S., Smith, R., Feng Q., Murrell, J. R., & Rook, M. S. (2017). Process development for expansion of human mesenchymal stromal cells in a 50L single-use stirred tank bioreactor. Biochemical Engineering Journal, 120, 49–62.
- https://doi.org/10.1016/j.bej.2016.11.020
 Tangential Flow Filtration | Diafiltration & concentration at once? How? (n.d.), Rocker.
- https://www.rocker.com.tw/en/application/tangential-flow-filtration/
- Chen, Y.-S., Lin, E.-Y., Chiou, T.-W., & Harn, H.-J. (2019). Exosomes in clinical trial and their production in compliance with
- good manufacturing practice. Tzu an wedical Journal, 32(2), 113–120. https://doi.org/10.4103/tcmj.tcmj_182_19
 7. Collins, C. H., & Beale, A. J. (Eds.). (2015). Safety in Industrial Microbiology and Biotechnology. Butterworth-