Advancing Gene Therapy Through Large-scale Viral Vector Production

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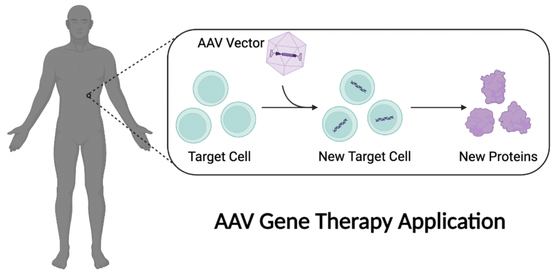
AAVantage Therapeutics

INTRODUCTION

1 in 12
~ 3 Million
Canadians suffer from rare genetic diseases

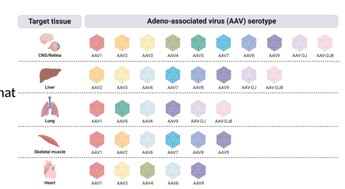
Adeno-associated Viral Vectors (AAVs) are used to carry functional DNA to patients and have proven to be a safe and effective treatment option. Gene therapy currently costs up to ***$2M per dose***. AAVantage aims to meet the growing demand while addressing the high cost of production and complex process designs. Canada's limited capability in biologics manufacturing highlights the pressing need for a robust domestic life science sector.



AAV Gene Therapy Application

Used to target various tissues:
- CNS and Retina
- Liver
- Lungs
- Skeletal Muscle
- Heart

Common genetic diseases/conditions that can be treated:
- ***Alzheimer's Disease***
- ***Hemophilia***
- ***Ocular Dystrophy***
- ***Spinal Muscular Atrophy***
- ***Cancer***



SAFETY & ENVIRONMENT

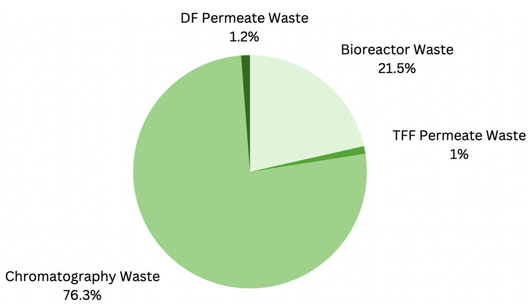
Safety Risks

- Equipment Malfunction
- Spills
- Contamination
- Personnel Exposure to Biomaterials
- Human Error
- Process Disturbances

Mitigations

- Operator Training & PPE
- HEPA Ventilation
- cGMP Protocols
- Sensors, Alarms, Signage
- Equipment Maintenance
- Wash Stations & Cleaning

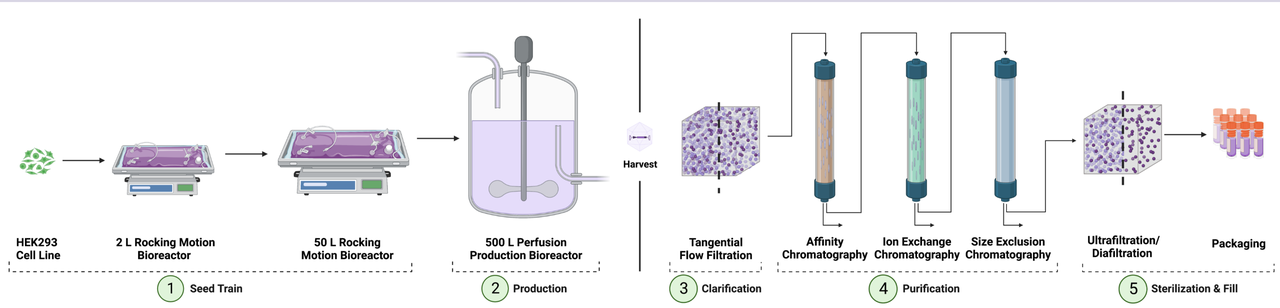
Annual Waste Production



OUR PROCESS

UPSTREAM

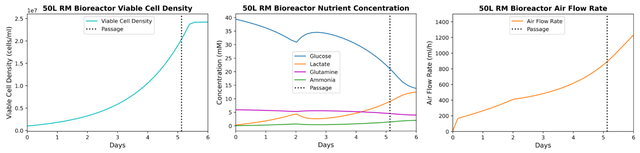
DOWNSTREAM



1 Seed Train

Two ***Rocking Motion Perfusion Bioreactors*** of 2L and 50L are operated in series where HEK293 cells are expanded to a sufficient cell density for inoculation into the production bioreactor. Three key parts of the bioreactor are:

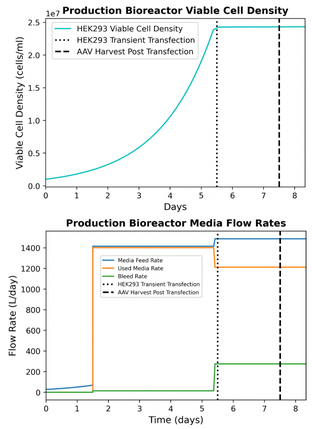
- Perfusion allows fresh media to be fed to the bioreactor ensuring enough nutrients are available for cell growth
- A cell retention device dispenses used media while retaining the cells
- A cell bleed is used to remove media and cells maintaining cell density

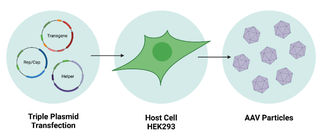


2 Production Bioreactor

The ***500L Perfusion Stirred Tank Production Bioreactor*** is the final step of cell growth. Operation of the final bioreactor includes:

- Cells reaching high cell density and viability
- Operating in two phases, intensified cell growth and viable cell maintenance
- A sub-surface sparger system delivers a combination of gases to maintain the desired dissolved oxygen concentration
- Cells are transfected at their peak viable cell density



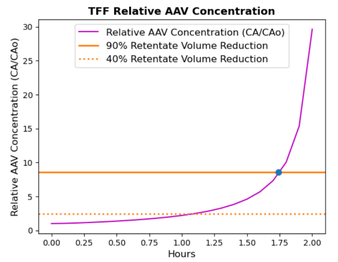


Transfection

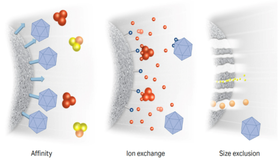
consists of three DNA plasmids mixed with a transfection reagent to be inserted into the HEK293 cells. The plasmid will encode for AAVs production and cell lysate is used to break down HEK293 cell membrane to extract AAVs.

3 Tangential Flow Filtration

A highly efficient technique that can provide yields of up to 95%. The ***hollow fibre filter*** allows for AAV retention in retentate and purification of cellular debris and large impurities. TFF operates by crossflow filtration, in which the feed tank is pumped parallel to the membrane surface.

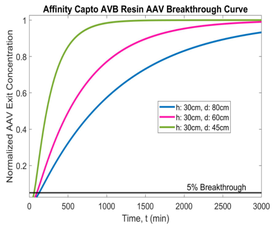


4 Chromatography



Affinity Chromatography

operates using the bind-and-elute method. AAVs bind to specific ligands on the resin surface. At the same time, unbound impurities such as DNA and host cell proteins flow to a waste collection tank. An elution buffer is then used to release the AAVs from the resin.



Ion Exchange Chromatography

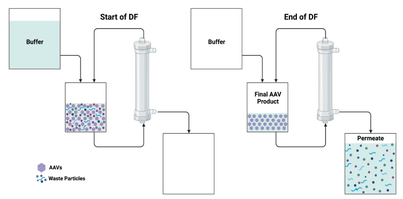
exploits the difference in the net surface charge of the impurities and AAV product to separate the particles.

Size Exclusion Chromatography

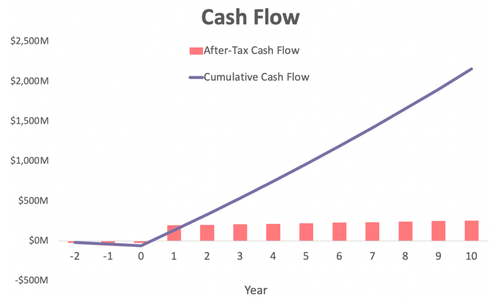
separates the impurities and product by their molecule sizes. Smaller impurities are captured in the resin pores while the AAVs flow through.

5 Diafiltration

Diafiltration allows for quick removal of salt and buffer replacement in a manner that minimizes risk of sample loss and contamination.

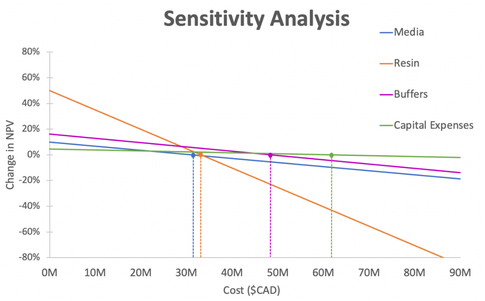


ECONOMICS



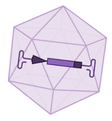
Total Capital Investment	Annual Operating Cost
$61.3M	**$145.1M**

Net Present Value	Discounted Payback Period	Internal Rate of Return
$1.3B	**3.4 years**	**120%**



PRODUCT

AAVantage Therapeutics is able to produce ***35 batches per year*** while operating 90% of the year. Over ***4000 doses per year*** are produced, impacting a large population of individuals with rare genetic diseases and advancing gene therapy research in Canada.



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Lonza
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