Testing Strategies for Ex-vivo Gene Therapies

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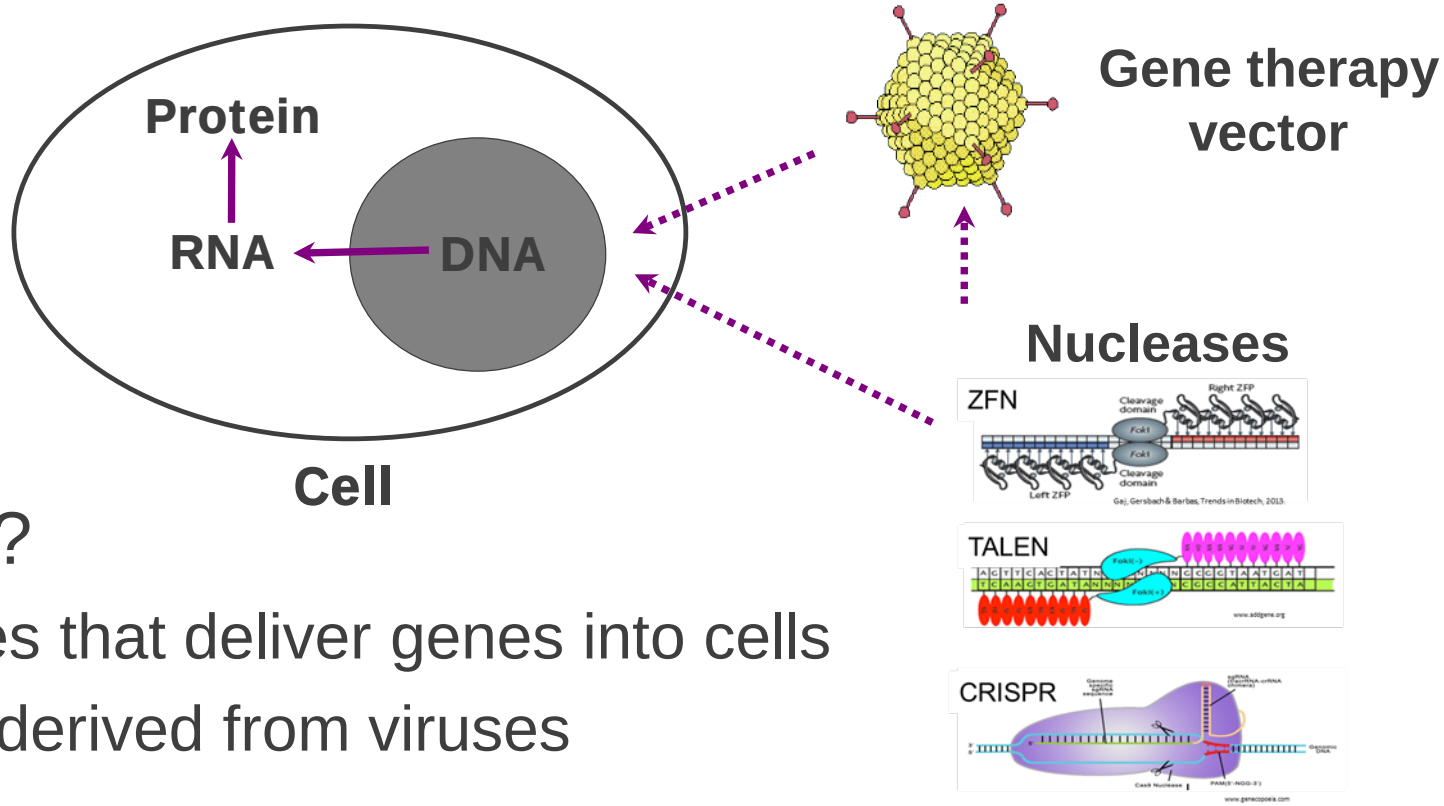
What is Gene Therapy?

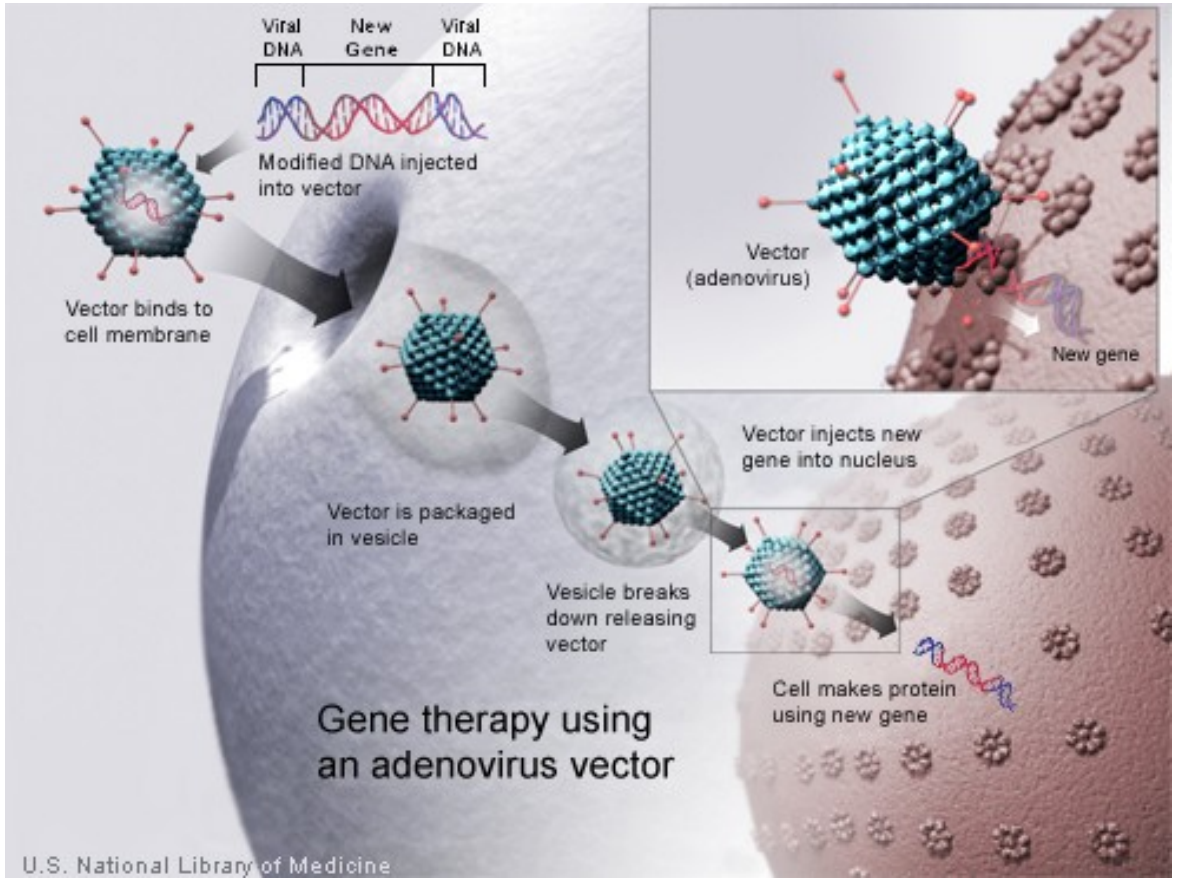
- Gene therapy vectors modify the genetic instructions of cells



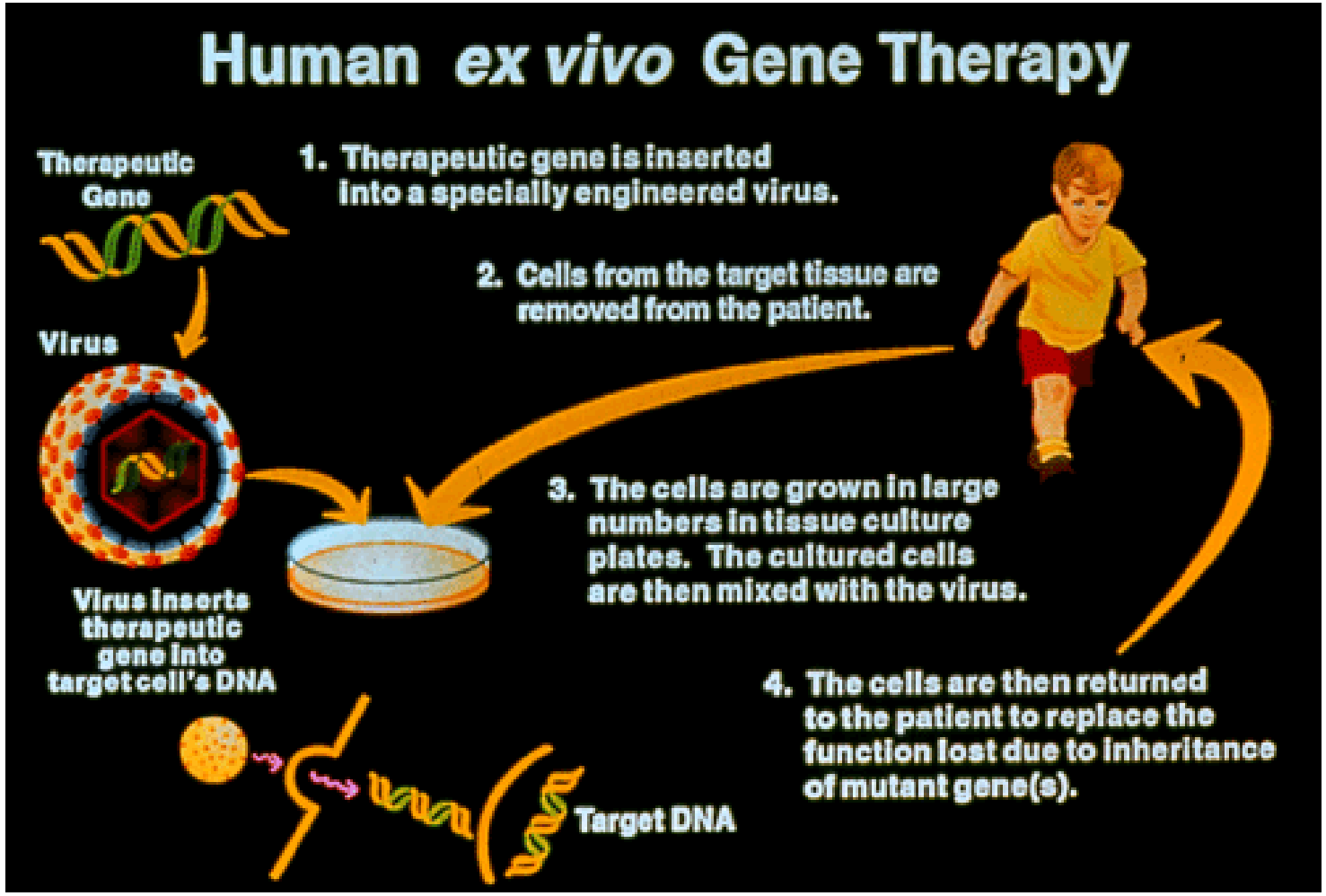
- What are vectors?
  - Miniature machines that deliver genes into cells
  - Many vectors are derived from viruses

Viral-based Gene Therapy Vectors

- Viruses are efficient gene delivery machines because they naturally infect cells



Ex vivo Gene Therapy



Recent Progress in Gene Therapy Clinical Trials

- Improving vision
  - Disease: Leber's congenital amaurosis
  - Target: The retina
- Correcting blood clotting defects
  - Disease: Hemophilia
  - Target: The liver
- Retargeting the immune system to attack cancer
  - Disease: Cancer
  - Target: Tumors

FDA OTAT Draft Guidances

1. Chemistry, Manufacturing, and Control (CMC) Information for Human Gene Therapy Investigational New Drug Applications (INDs)
2. Testing of Retroviral Vector-Based Gene Therapy Products for Replication Competent Retrovirus (RCR) during Product Manufacture and Patient Follow-up
3. Long Term Follow-up After Administration of Human Gene Therapy Products
4. Human Gene Therapy for Hemophilia, on gene therapy products intended for treatment of hemophilia
5. Human Gene Therapy for Retinal Disorders
6. Human Gene Therapy for Rare Diseases

History of CMC Gene Therapy Guidances

- 1991 Points to Consider (PTC) in Human Somatic Cell and Gene Therapy
- 1998 Guidance for Industry: Guidance for Human Somatic Cell and Gene Therapy
- 2008 Guidance for Reviewers and Sponsors: Content and Review of Chemistry, Manufacturing, and Control (CMC) Information for Human Gene Therapy Investigational New Drug Applications (INDs)
- 2018 Draft Guidance for Industry: Chemistry, Manufacturing, and Control (CMC) Information for Human Gene Therapy Investigational New Drug Applications (INDs)

Common Themes on Testing Outlined in CMC Guidances

- General Biological Product Standards (21 CFR 610s)
  - Safety testing
  - Characterization testing
- Timing
  - When to develop tests
  - When to take samples during production
  - Release Specifications and Redundant Testing
- Reference standards

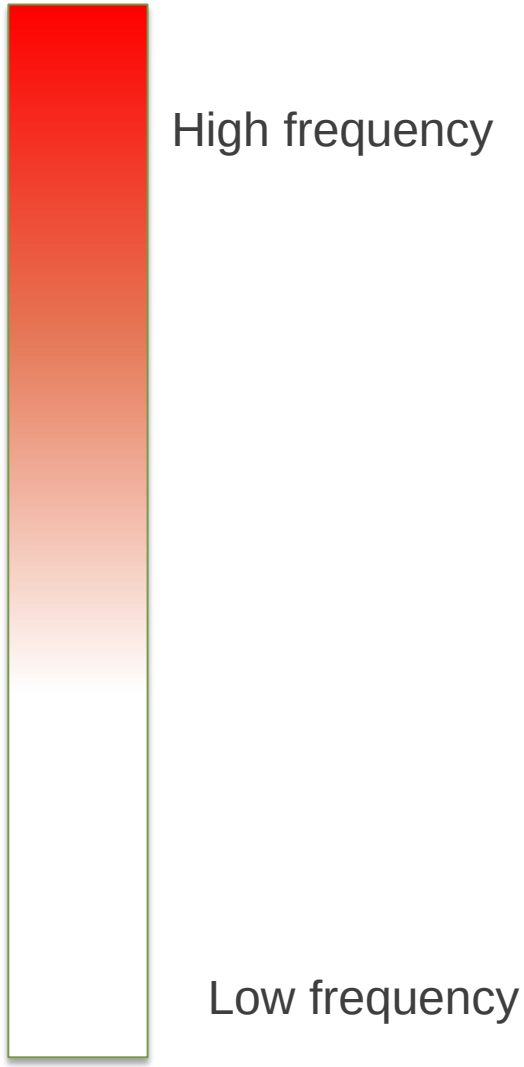
- Caveat and Exclusions
  - Critical Quality Attributes (CQAs) may be unknown or incompletely understood
  - Exclude donor eligibility testing (for manufacture)
  - Exclude testing subjects for eligibility (for investigational treatment)

General Biological Product Standards

- Required prior to release of each lot
- 21 CFR 610 Subpart B – General Provisions
  - 610.10 Potency
  - 610.12 Sterility
  - 610.13 Purity
    - Endotoxin
    - Impurity profile
  - 610.14 Identity
  - 610.30 Mycoplasma

Testing Toolkit for Ex-vivo Modified Cell Manufacturing Includes…

- Viable cell Count
- Cell phenotype
- Appearance / culture conditions
- Sterility / Bioburden
- Mycoplasma
- Replication Competent Retrovirus
- Vector copy number
- Dose (number of gene modified cells)
- Residuals
- Potency / Functional activity



Testing Toolkit for Ex-vivo Modified Cell Manufacturing Includes…

- Viable cell Count
- Cell phenotype
- Appearance / culture conditions
- Sterility / Bioburden — CFR 610.12
- Mycoplasma — CFR 610.30
- Replication Competent Retrovirus
- Vector copy number

Safety

- Dose (number of gene modified cells)
- Residuals
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Testing Toolkit for Ex-vivo Modified Cell Manufacturing Includes…

- Viable cell Count
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- Replication Competent Retrovirus
- Vector copy number
- Dose (number of gene modified cells)
- Residuals — CFR 610.13
- Potency / Functional activity — CFR 610.10

} Product specific characterization

Test Timing

- Test Development
  - Safety tests (compendial or platforms specific tests) for first in human
  - Dose assay qualified for first in human study
  - Product specific tests (potency and impurity profile) developed during clinical studies
- Sampling during production
  - maximize the sensitivity of safety testing, perform test at the stage of production at which contamination is most likely to be detected
  - For impurities below assay limits, consider sampling “upstream” for a more informative readout

Test Timing (cont)

- Release Specifications and Redundant Testing
  - Common Technical Document (CTD) has separate specification for DS and DP
  - Usually separate release specification for vector and final cell product

Release Specifications and Redundant Testing (cont.)

- Safety testing (sterility, endotoxin, mycoplasma, RCR/RCL)
- Identity
- Impurity profile
- Potency
- 2018 Draft guidance text on redundant testing
  - "Not all testing listed in the guidance is required for release of both the DS and DP. In some cases, repeat testing may be good practice; however, redundant testing may not always be feasible or practical. In this case, we recommend that you provide a rationale to support the selection of testing performed for release of either DS or DP."

Try to maximize the effectiveness of testing and minimize redundancies

Reference Standard Materials (RSM)

- Use RSM is to improve accuracy
  - Control for assay variability between labs
- RSM Platforms
  - Ad5 (AATC)
  - AAV2/AAV8 (AATC)
  - LVV (RSM initiative under development, ISBioTech)
  - Genome Editing Consortium (no RSM, NIST)
- Use RSM to qualify in-house reference standard
  - Virus particles (VP)
  - Infectious titer (IU), and/or
  - Genome copies (GC)

Summary

- Gene Therapies are innovative approaches showing promise for treatment of severe, life-threatening diseases
- Testing for Gene Therapy products can be generalized as safety tests or product-specific characterization tests (potency, impurity profile)
- Consider ways to maximize the effectiveness of release tests (DS and DP) and minimizing redundancies.
- Reference standards (existing or those under development) can improve test accuracy

Thank You

- Acknowledgements

  Leslie Wilder

  Jessie Hanrahan

  Anne-Virginie Eggimann

Useful References

- OTAT Learn webinars

https://www.fda.gov/BiologicsBloodVaccines/NewsEvents/ucm232821.htm

- FDA Cell and Gene Therapy Guidance

https://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/CellularandGeneTherapy/default.htm