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Demonstrating Comparability of AAV Gene Therapy Products: Application of Lessons Learned

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Biogen

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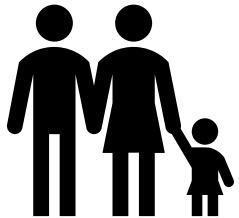
Overview

- Why is Comparability Necessary?
- Comparability Challenges during Development
- Maximizing Success
- Outcomes
- Regulatory Precedents and Support

Why is Comparability Necessary?

- Regulatory requirement
  - Must be performed for any manufacturing change to DS and / or DP before, during and after clinical development
  - In a phase-appropriate manner
- Aims for demonstrating comparability:
  - To ensure the changes do not adversely impact on product quality, safety and efficacy
  - Patient safety



Regulatory Landscape

- The guidance available to gene therapy product developers for comparability assessments has expanded:
  - ICH Q5E remains the basis for designing comparability assessments
  - ICH Q5E can be used in combination with the EMA Q&A on comparability considerations for ATMPS when planning comparability assessments
  - Other Agencies have also provided guidance on comparability assessment
    - The regulatory requirements comparability appear consistent across the different regions and focus on developing an understanding of whether changes have an impact on the product quality and if this could have an impact on clinical safety and efficacy.
    - However, how the guidances are implemented may be inconsistent across regions.
    - There may also be an issue with less experienced agencies accepting alternative approaches, leading to challenges in global approval.
- Overall, the general principles of ICH Q5E can be applied to AAV-based gene therapy products and a step-wise approach for the comparability assessment is recommended.

Challenges

One small change can expand to a large effect

Many small changes can lead to a very complicated effect

Challenge the Perception

- Process development for gene therapy products may be perceived to represent an additional challenge

Product

Process

- Improving the understanding of the product could manage this link between the product and the process

Learnings from Monoclonal Antibody Experience

	mAb Experience	AAV Experience
Risk-based approach	✓	✓
Well characterized	✓	✗
Understood CQAs	✓	?
Known structure-activity relationships	✓	✗
Well developed analytical toolkit	✓	?
Well developed potency assays	✓	?
Stability profile	✓	✓
Forced degradation profile	✓	✓
Additional *in vitro* toolkit	✓	?
Bridging nonclinical study requirements	✗	✓
Clinical experience	✗	✓

Components of a Robust Comparability Strategy

Sufficient knowledge / capability to understand the impact of any process change to the quality attributes on product safety or efficacy

Well defined and understood process / CQAs

Extensive panel of well-controlled, robust and sensitive methods and orthogonal characterisation assays

Methods capable of detecting small changes in CQAs, including potency matrix testing

Material from multiple pre & post change batches (including clinical batches)

Acceptance criteria defined from statistical analysis of multiple pre change batches, including clinical lots

Integrate and evaluate data from different sources (DS, DP, intermediates, in-process controls, PV data, stability)

Changes later in development will require a more comprehensive comparability strategy

Robust comparability protocol

Comparability Protocols

- **Design a comparability strategy capable of detecting changes in product quality**

- Evaluation of the process changes to determine how risks can be mitigated
- Analytical considerations
- Testing strategy
- Acceptance criteria

Evaluation of Manufacturing Changes

Identify and describe	• Identify and describe all manufacturing changes
Evaluate	• Evaluate each change for the potential impact on product properties
RBA	• Utilize risk-based approach
Design	• Define the CQAs and design the development strategy

Analytical Considerations

- Is the method well-controlled?
- What is the qualification / validation status of the method?
- What is the variability of the method?
- Can the method detect small changes in the product quality attributes?
- Can appropriate comparability acceptance criteria be assigned?
- Has the method changed during product development?
- Has appropriate method bridging been conducted where there have been changes?
- Are retain samples available for comparability assessments?

Testing Strategy

Assess process steps most appropriate to detect a change

Compare process and product

In-process, batch release, extended characterisation, stability

Biological activity is an important component of the testing strategy

Define an appropriate number of representative pre- and post-change batches

Side-by-side testing is the preferred strategy

Acceptance Criteria

Post change product to be highly similar, not identical

Determine how to define ‘highly similar’ and defining the “similarity condition”?

Clinical batch release specifications are generally not considered adequate

Acceptance criteria set using multiple batches and appropriate statistical analysis

Assess any trends within the acceptance criteria

Assessing Stability for Comparability

Stability may be required (DS and DP)

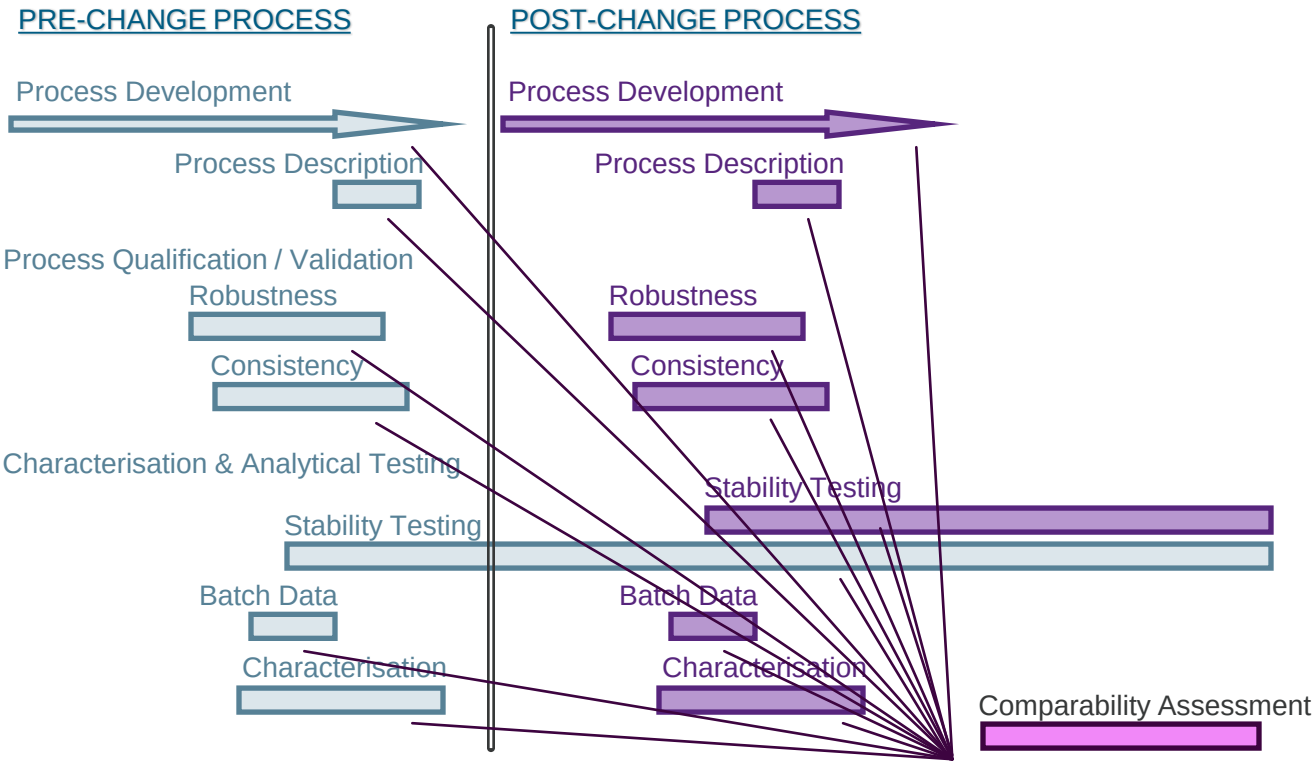
Stability at intended storage may not be sufficient / practical

- AAVs are remarkably stable
- Time to perform study

Stress stability can be beneficial

- Quicker and may be more sensitive to detect product change
- Requires prior identification of degradation pathways
- Methods to be stability indicating, sensitive and quantitative
- Study conditions to be defined

Consider Everything, Ignore Nothing



Challenges for a Successful Comparability Study

Process and analytical understanding improving but still limited	Suitable panel of qualified/validated, sensitive analytical methods may not be available	CQAs may not be identified early in clinical development	Potency assays may not be available to assess all parts of the biological activity
Testing methods may have changed – changes should be well-controlled and suitably bridged	Limited number of batches used in development and in the clinic	Sufficient batches to set acceptance criteria using appropriate statistical tools	Limited material to allow sufficient reserve samples to support comparability
Stability of reserve samples	Need for DS and DP stability data to support changes	Changes may be introduced after pivotal dosing completed	Existing knowledge may not be sufficiently predictive to ensure that differences in quality attributes have no adverse impact upon safety or efficacy

Outcome of Comparability Assessment

Requirement for Nonclinical / Clinical Studies

- ‘If a manufacturer can provide assurance of comparability through analytical studies alone, nonclinical or clinical studies with the post change product are not warranted’ (ICH Q5E)
- Assess the use of *in vitro* and *in vivo* nonclinical studies to augment the analytical assessment
- Due to the complex nature of GT products, diverse MOA, limited process understanding, material availability and analytical limitations, it is possible clinical studies will be required
- Regulators are requesting clinical experience or inclusion of post change material in pivotal study for changes made late in development
- Additional / unexpected requirement for nonclinical and / or clinical studies can have a big impact on program timelines / budgets / logistics
- Important consideration when planning manufacturing changes

Regulatory Precedents

Reviews of approved ATMPs highlight comparability questions were raised during assessment for the majority of approved products

Use regulatory intelligence and precedents to help develop strategy

Gain prospective acceptance of the comparability strategy by seeking advice from the regulators

Luxturna Review

EMA:

Comparability Exercise for Active Substance

A comparability evaluation was performed to demonstrate that the active substance produced at Spark is comparable to the material used in the Phase III pivotal clinical study and that the change of manufacturing facility had no impact on the quality attributes of the active substance.

- EMA review findings:
  - Comparability of the proposed commercial FP with the clinically qualified material was of concern and underpinned a large number of the issues raised.
  - However, the comparability evaluation did demonstrate that the active substance and drug product produced at Spark is comparable to the material used in the Phase III pivotal clinical study in a 1:1 comparison. The evaluation consisted of a combination of analytical release testing and side-by-side testing.

clinically qualified material was of concern, and underpinned a large number of the issues raised. Tightening of acceptance criteria for critical process controls and release specifications in line with clinically qualified material was considered necessary unless additional validation data could justify the wider ranges claimed. The control strategy has now been tightened in several areas, as requested, and additional validation datasets have been provided. Assays which were insufficiently described have now been more thoroughly detailed and important issues regarding the validation of several critical analytical methods are now largely resolved (see also "Recommendations for future quality development").

FDA:

3.2.P.2.3.3 Comparability

A comparability evaluation was conducted by Spark to demonstrate that the voretigene neparvovec-rzyl Drug Product produced at (b) (4) is comparable to the CHOP material used in the Phase 3 pivotal clinical study and that the change in manufacturing facility and container closure has had no impact on product quality. Following are data of analytical results for side-by-side testing from Drug Product comparability study:

- FDA review findings:
  - DS comparability under a prospective protocol.
  - Comparability is 1 clinical + 1 PPQ. 1 Engineering run used to set/confirm criteria.
  - Acceptance criteria for potency are wide, but results showed that the two products are comparable using lot release data, and the results from the side-by-side comparability assessment.

manufactured at CHOP.

EMA: Luxturna, INN-voretigene neparvovec (europa.eu)
FDA: https://www.fda.gov/vaccines-blood-biologics/cellular-gene-therapy-products/luxturna

UPSTAZA Review

EMA:

- Many significant and substantial changes were introduced as part of the Process C commercial manufacturing process:
- A major objection was raised at Day 120 on the deficiencies in the comparability exercise as data from only one Process A clinical batch and one Process B clinical batch were available.
- The analytical comparability exercise included tests for potency, identity product-related purity, process-related impurities and safety and the panel of tests was considered reasonably comprehensive for demonstrating comparability of an AAV product.
- However, there were uncertainties whether the commercial batches have comparable biological activity and comparable levels of empty capsids compared to the clinical batches.
- As there are no more samples from Process A and B batches available for further side-by-side testing, no further information which could aid in regulatory decision making can be obtained from these batches. Therefore, no further questions are raised from a quality perspective.
- The remaining uncertainty on comparability was considered acceptable in the context of the overall benefit/risk decision.
- However, the clinical data from Process A material was considered only supportive and only clinical data from Process B material was pivotal.
- In addition, further process data had to be submitted post-approval to confirm consistency.

2.4.4.

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The MAH

Descrip
In order applican product finished product batch. This data should be provided by March 2023.

3.3. *Uncertainties and limitations about favourable effects*

There is limited safety or efficacy data from patients treated with the commercial product. The comparability exercise conducted between product manufactured using process A and process B, was not conclusive. Therefore, the clinical data from patients treated with process B product is considered

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quality of life, the limited data relating to the efficacy and safety of the commercial product, the lack of demonstration of comparability between process A and process B product and the clinical relevance, if any, of the high percentage of empty capsids in the commercial product.

Zolgensma Review

FDA:

3.2.P.2.3 Manufacturing Process Development

Reviewer comment: Evaluation of comparability is difficult due to changes in assays, variable assays, manufacturing problems with early AveXis lots conclusions can be reached, however:

- *The protein composition is equivalent between*
- *The formulation is different (b) (4) for most AveXis lots), but there is no evidence would alter the product CQAs.*
- *In terms of purity, the post-PPQ AveXis lots ar AAV9SMN0613.*
- *The in (b) (4) assay indicates that potenc*

- FDA review findings:
  - The commercial manufacturing process produced drug product with CQAs comparable to those of the initial clinical lot.
  - Although the concentration of drug product declined over time during storage, the ratio of potency to content (vg) was comparable when pre- and post-change lots were compared.
  - FDA conducted an extensive reassessment of the in vivo data and concluded that the results supported comparability of the pre- and post-change material.

EMA:

2.2. Quality aspects

2.2.2. Active Substance

Overall the changes between upstream process A and process B are suffic
the change of production sites, upscaling and introduction of bioreactor,
as process optimisation and improvement of robustness and consis
downstream Process B were extensive and Process A a
addition a MCB/WCB of HEK293 cells was introduced fo
were manufactured by a commercial supplier. Due to ch

titre and infectivity.

The response of the applicant showed that only a Phase
measured titre due to a change in the analytical method. All Phase 3 clinica
batches were tested using the revised test method and show consistent

After the Phase 1 clinical trial using the initial clinical lo
changed considerably. The current manufacturing proc
quality attributes that are comparable to those of the in
concentration of drug product declines over time durin
genomes is comparable when lots from the current ma
directly to the initial clinical lot, including comparable ability to enhance survival in a mouse model of SMA. Drug product manufactured using the current manufacturing process has better purity (b) (4)

2.2.2. Discussion on chemical, pharmaceutical and biological aspects

Three manufacturing processes are described during development: Process A, Process B-initial, and Process B-commercial. Major changes are made between Process A and Process B. Comparability of batches manufactured according to Process A and batches manufactured according to Process B has not been demonstrated. However, as the single Process A batch is > 4 years old and bridging of the test results is not possible this issue (D120 Major Objection) was not further pursued. As comparability of the Process A batch and Process B batches cannot be demonstrated, the assessment of the benefit risk of Zolgensma will be based on clinical data obtained with Process B batches.

2.3. Non-clinical aspects

The pharmacology nonclinical data of Zolgensma included in the dossier have been conducted with research viral preparations. During further development of the manufacturing process of Zolgensma, manufacturing Process A has been established at the Nationwide Children's Hospital (NCH) for manufacturing of the test material used in the Phase I clinical trial. Finally manufacturing Process B has been established at AveXis, which represents the current proposed and validated commercial process. Process A material has not been tested in non-clinical studies. Process B material has been used in

explain the variation seen in the clinical studies (CL-302, CL-303, and CL-304) the applicant committed to further analyse whether tightening of the acceptance criteria for quality parameters is needed to ensure optimal clinical outcome.

- EMA review findings:
  - EU reviewers took a much stronger stance on comparability.
  - Additional data, including retesting of Process A material with Process B-initial and Process B material could not be conducted due to the age of the material.
  - EMA considered that comparability of the Process A batch (Ph1) and the Process B batches (Ph3, other clinical studies and commercial) could not be demonstrated.
  - Therefore, the clinical data from Process A material was considered only supportive and only clinical data from Process B material was pivotal and the assessment of the benefit risk of Zolgensma was based on the clinical data obtained with Process B batches.

EMA: Zolgensma, INN-Onasemnogene abeparvovec (europa.eu)
FDA: https://www.fda.gov/vaccines-blood-biologics/zolgensma

AAV Review Precedents

Glybera

Manufacturing process development

The original manufacturing process (AMT-010) used a plasmid based system. The plasmids were then transfected into HEK293 in order to rescue the recombinant AAV. Scientific advice had been sought in relation to the comparability assessment of the DS derived from this process and the baculovirus production system (AMT-011) introduced for commercial production. On the whole the applicant has complied with the advice given. Some other concerns w
addressed by the applicant's response to the LoQ.

During the development of the AMT-011 process a num

A comparability assessment of product from these proc
importance is the comparability between the process u
process. The results indicate that the product purity ha
manufacturing process. In most analyses the commerc
the clinically used process, except for significantly high

Overall the consistency in product quality throughout d

The applicant did not consider the evaluation of compa
necessary as AMT011 has been qualified independently of AMT-010 on the basis of non-clinical studies. This is considered acceptable as toxicology and pharmacology studies were repeated with AMT-011.

- A few good examples:
  - Glybera is an old approval and shows how nonclinical studies were used to fill some gaps.
  - The Imlygic example shows that EMA does have expectations for providing all comparability data for all stages of process development, and the information was available and was provided during review.

ng process development three primary manufacturing
ogene laherparepvec during product development.
lts of comparability assessment were submitted at the
ere not included in the original MA submission. These
n request. The analytical comparability between Process B and Process C, as well as between Process C pre- and post-facility and equipment modifications are adequately addressed and comparability between batches manufactured using the different manufacturing processes has been demonstrated.

Comparability exercise for finished medicinal drug product

Active substance and drug product process, analytical, non-clinical and clinical comparability data has been submitted. An assessment of drug product, including stability evaluations was performed for each comparability exercise and comparability was shown, also between the commercial and clinical trial formulations.

Glybera: Glybera, INN-alipogene tiparvovec (europa.eu)
Imlygic: Imlygic; INN-TALIMOGENE LAHERPAREPVEC (europa.eu)

Use of PACMPs

Hemgenix: Hemgenix; INN-etranacogene dezaparvovec (europa.eu)
Roctavian: Roctavian; INN-valoctocogene roxaparvovec (europa.eu)
Imlygic: Imlygic; INN-TALIMOGENE LAHERPAREPVEC (europa.eu)
Zolgensma: Zolgensma, INN-Onasemnogene abeparvovec (europa.eu)

Hemgenix

2.4.3.5. Post-approval change management protocol(s)

A post-approval change management protocol (PACMP) was submitted to introduce a process change in the active substance manufacturing process. The general approach proposed by the applicant was considered acceptable in general, additional recommendations were provided to the applicant during the procedure. However, the applicant decided to withdraw the PACMP.

Roctavian

2.4.3.5. Post-approval change management protocol

A PACMP is submitted to introduce a process change in the finished product manufacturing process. The proposed PACMP is acceptable.

II/0004/G	This was an application for a group of variations. B.I.e.5.c - Implementation of changes foreseen in an approved change management protocol - For a biological/immunological medicinal product B.I.a.1.j - Change in the manufacturer of AS or of a starting material/reagent/intermediate for AS - Replacement or addition of a site where batch control/testing takes place and any of the test method at the site is a biol/immunol method	26/04/2023	n/a

Imlygic

Post approval change management protocol

As part of the MA application for Imlygic, Amgen has submitted a post-approval change management protocol (PACMP). The purpose of the PACMP is to scale up the cell build process for talimogene laherparepvec. Following approval of the marketing authorisation and the PACMP, the changes described in the protocol will be introduced through a type IB variation application procedure.

The proposed changes have been adequately presented and discussed. The minor revisions and/or clarifications requested have been agreed by the Applicant, the final updated PACMP will be submitted with the eCTD closing sequence.

Zolgensma

II/0020/G	This was an application for a group of variations. B.II.g.2 - Introduction of a post approval change management protocol related to the finished product B.II.b.1.d - Replacement or addition of a manufacturing site for the FP - Site which requires an initial or product specific inspection B.I.a.1.e - Change in the manufacturer of AS or of a starting material/reagent/intermediate for AS - The change relates to a biological AS or a starting material [-] used in the manufacture of a biological/immunological product	19/05/2022	25/08/2022

AAV Review Precedents - Seeking Regulator Advice

Hemgenix

The protocol assistance pertained to the following quality, non-clinical, and clinical development aspects:

- The comparability strategy to address changes in the manufacturing process for AMT-060 (predecessor of AMT-061) to be employed for clinical phase III and commercial vector production. Proposal to adjust the manufacturing process in order to improve process and product consistency.

  The comparability strategy to address changes in the manufacturing process to be employed for clinical phase III and com
  characterisation.
  Sufficiency of the nonclin
  with AMT-061 and AMT-0

Manufacturing Process

Production of the active su
described. The comparabili
of process performance, ac
characterisation.

2.5. Non-clinical as

2.5.1. Introduction

clinical route of administration. The etranacogene dezaparvovec batches used in the non-clinical safety testing were representative of the final product used in clinical phase 2b and 3 studies. AMT-060 and etranacogene dezaparvovec were similar in terms of transduction efficacy, hFIX transcription and translation efficacy, biodistribution pattern and safety. Up to 4 to 6-fold higher FIX clotting activity was noted with etranacogene dezaparvovec administration in comparison to equal doses of AMT-060 in mice and monkeys.

2.6.6. Discussion on clinical efficacy

The applicant has not undertaken a proper dose finding study for etranacogene dezaparvovec. Only 3 patients were recruited into this first clinical study, based on the result derived from the predecessor product, AMT-060. However, these early results are encouraging and the efficacy results of the pivotal study are in-line with this dose finding study. Consequently, the issue of not performing a proper dose finding study was not further pursued.

Roctavian

A kick-off meeting was held on 10 May 2017. The objective of the meeting was to discuss the development programme and regulatory strategy for the product. The applicant was recommended to address the following key issues through relevant regulatory procedures:

- Strategy for comparability of drug product material from manufacturing processes C, D and D' and qualification of the proposed assay for product strength;

The Protocol assistance pertained to the following *quality, non-clinical, and clinical* aspects:

processes was previously

ed during clinical trials was
according to the commercial

ble.

process and the processes
comparable. Characterisation
roduct.

An inter-process comparability was conducted in Study BMN 270-16-049; this study had the objective to compare two manufacturing processes: process C (used for phase I/II studies) and process D (used for phase III studies). BMN 270 was administered as a single IV injection at 6.0E12 or 6.0E13 vg/kg to Rag2-/- mice with a 36-day observation period. It appeared that, at the same dose level, the average hFVIII-SQ protein and FVIII activity levels were higher for Phase I/II material compared with Phase III material.

reported. No trends were observed. The safety profile of patients treated at the therapeutic dose is generally comparable regardless of the clinical batch/manufacturing process used.

Seeking Regulator Advice:

- Seeking advice from Agencies prior to submission is highly beneficial, especially where Module 3 package is limited.
- If you ask for advice, follow it, unless a good scientific justification can be provided.

d Proprietary

Hemgenix: Hemgenix; INN-etranacogene dezaparvovec (europa.eu)
Roctavian: Roctavian; INN-valoctocogene roxaparvovec (europa.eu)

Agency Engagement Opportunities

US

- INTERACT meeting
- CATT meeting
- Pre-IND meeting
- Type B and C meetings
- EOPII meetings
- Pre-BLA meetings

EMA

- Innovation Task Force
- EMA Scientific Advice / Protocol Assistance
- Pre-MAA meetings
- PRIME meetings
- ATMP Certification (SMEs and Academia)

Germany

- Pre-advice
- National Scientific Advice
- Simultaneous National Scientific Advice
- Portfolio meeting

UK

- MHRA Innovation Office
- Scientific Advice
- Pre-MAA meetings

PMDA

- Regulatory Science consultation:
  - Informal meeting
  - Formal PMDA consultation

Key Success Factors

Well documented process and development history	Know your product; know your process Exploit experience from small scale models
Comprehensive product characterization	Understand the capabilities and limitations of analytical methods
Develop a well thought out strategy for development and commercialization	Try not to complicate matters; avoid major changes between phase III process and commercial process
Be creative: In vivo and *in vitro* nonclinical studies could be helpful to complement analytical characterization	
Clinical studies may be necessary if analytical comparability is not established, and nonclinical results are uncertain or irrelevant	
Even with comparability, providing clinical experience may be required by the regulators	

Summary

Changes to manufacturing processes inevitable
Comparability is a challenge for gene therapy products
Plan changes early where possible
Implement a risk-based approach
Define methods and acceptance criteria capable of detecting changes to CQA
Evaluate final product, intermediates, in-process controls, stability
Well controlled method changes
Ensure sufficient retains from all pre and post change lots
Assess if nonclinical studies could supplement analytical comparability
Be prepared for the requirement for clinical exposure
More comprehensive strategy required for late-stage changes
Engage early with regulatory agencies