Life Cycle Management of Analytical Methods for Biotechnology Products: A Regulatory Perspective

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Presentation Outline

- Life cycle of analytical methods
- Analytical method development and validation through the product life cycle
- Regulatory considerations for changes in analytical methods
- Case Studies: replacement and transfer of analytical methods

Disclaimer

Some of the views expressed during this presentation are my own and may not necessarily reflect the official opinion of FDA
Major Stages in a Method’s Life Cycle

Selection/Development

Qualification

Validation
Selection and Design of Analytical Methods

The selection and design of analytical method should be based on a systematic approach:

• **Intended use of the assay**
  - Purity, impurities, potency, identification, safety
  - Release test, stability test, characterization, in-process testing

• **Identify sources of analytical variation**
  - Method variability, risk from analyst, reagents, and instruments

• **Define reportable results**
  - Independent tests, mean of replicates etc.
Rational Assay Development/Qualification

• Investigate and identify the assay’s critical characteristics and parameters.

• Method robustness studies are performed to evaluate its reliability during conditions of use.

• Detailed assay SOP, with system suitability requirements, that incorporates the assay’s characteristics and parameters into a clear, concise procedural plan.

• Use this information as a basis from which to develop a scientifically sound validation plan that will demonstrate that the assay does what it is intended to do on a routine basis.
Qualification Vs. Validation

- Assay Qualification:
  
  **Determining** whether an assay is suitable for its intended purpose
  
  - Limited pre-determined performance criteria

- Assay Validation:
  
  **Assuring** the assay is suitable for its intended purpose on a routine basis
  
  - Pre-defined assay performance criteria
• Validation trials are run according to an established validation protocol.

• Method performance specifications are pre-established, documented and confirmed during validation trial.

• These specifications must be met by every validation trial.

• A method can fail validation; if it does, assignable cause for the failure must be investigated, resolved, and the assay re-validated.
Why Validate an Assay?

21 CFR 211.165
“The accuracy, sensitivity, specificity, and reproducibility of test methods employed by the firm shall be established and documented in accordance with 21 CFR 211.194(a)(2).”

21 CFR 211.194(a)(2)
“[The firm] shall indicate the location of data that establish that the methods used in the testing of the sample meet proper standards of accuracy and reliability as applied to the product tested ..... The suitability of all testing methods used shall be verified under actual conditions of use.”
Analytical Method Validation

Non-Compendial Method: Non-compendial methods are validated according to the principles described under ICH Q2 (R1) document. Typical validation characteristics are:

- Specificity
- Linearity
- Accuracy
- Precision (repeatability, intermediate precision and reproducibility)
- Range
- Quantitation Limit
- Detection Limit

Compendial Method: Procedures used to evaluate a defined characteristic of the drug substance or drug product that are legally recognized under 21 USC 501(b) (USP/NF). Generally, will need only partial validation i.e., need to be verified under actual conditions of use (USP chapter<1225> and 1226>).
Analytical Method Validation

Analytical methods that need to be validated are:

• Lot release assays
• Stability methods for defining expiration dates/holding times
• Assays for significant process related impurities (e.g., host cell proteins, residual DNA, protein A, etc.)
• Analytical in-process tests
• Excipient and raw material testing (generally compendial)
Expectations For Methods During Product Development

• Ensure safety of the product
• Assay is providing meaningful results
• Assurance that analytical information gained in development can be reliably related to commercial manufacturing
• Determine method performance capabilities including specificity, linearity, accuracy, precision, robustness, and stability
Expectations For Methods During Product Development

- FDA Process Validation Guidance (2011)
  - “Validated analytical methods are not necessarily required during product- and process-development activities or when used in characterization studies.”
  - “…analytical methods should be scientifically sound (e.g., specific, sensitive, and accurate) and provide results that are reliable.”
  - Clinical supply production should follow the CGMPs appropriate for the particular phase of clinical studies.

- Interpretation:
  Review staff focus on the adequacy of non-compendial safety tests for early phase clinical supply material
Lifecycle Management of Analytical Methods

Pre-Clinical
- Selection
- Development
- Optimization

Phase 1
- Safety tests validated,
- Qualified methods
- Set tentative release/stability acceptance criteria

Phase 2
- Optimization/qualification
- Refine lot release criteria
- Set tentative validation acceptance criteria
- Delineate/initiate assay validation parameters

Post-Licensure
- Trend analysis
- Performance review
- Method replacement (supplement)

Phase 3 and BLA
- Full assay validation (strongly recommended for phase 3)
Lifecycle Management of Analytical Methods

Post-licensure activities for the method lifecycle management

1. Revalidation
2. Analytical Method Comparability
   • Change in method (Method Replacement and modification)
   • Analytical Method Transfer

Post marketing changes to analytical procedures must be reported to the FDA in compliance with 21 CFR 314.70 or 21 CFR 601.12.
Lifecycle Management of Analytical Methods Post-Licensure

Revalidation of analytical procedure should be considered:

• When an analytical method can only meet the established system suitability criteria with repeated adjustment to operating conditions stated in the analytical procedure

• When a change is made to an analytical procedure (e.g. change in reagent, equipment, formulation, or manufacturing process)
Lifecycle Management of Analytical Methods

Considerations for Revalidation

- Ensure that the analytical method maintains its critical performance characteristics, e.g., specificity, sensitivity, precision, etc.
- The degree of revalidation depends on the nature of the change
Lifecycle Management of Analytical Methods

Analytical Method Comparability should demonstrate:

- The new method, coupled with any additional control measures, is equal or superior than the original method for the intended purpose.
- The new procedure is not more susceptible to matrix effect than the original procedure.
Considerations for Method Comparability Studies

- To demonstrate that changes in the analytical procedures improve or do not significantly change analytical procedure characteristics that are relevant to the type of analytical procedure, its validation, and its intended use.

- Homogenous samples from the same batches should be included in the studies.

- Statistical analyses should be performed to demonstrate the comparability or equivalency of the new method with the existing method.
Considerations for Analytical Method Comparability

If an assay has stability indicating properties:

• The stability indicating properties of the new assay should be the same or better than the existing assay.

• Appropriate samples from multiple lots should be included that allow the comparison of the new and original method to detect relevant product variants and degradation species.

• If new product related variants or process related impurities are seen with the new assay, information from retained samples should be provided demonstrating that the variants/impurities are not new.
Considerations for Analytical Method Comparability

If an assay has stability indicating properties (contd...):

• Number of batches analyzed for comparison should be statistically relevant

• The statistical analyses performed to compare product testing should be identified. All biases seen with comparative results should be discussed with an explanation
Case Studies

Change in Analytical Methods
Changes in Analytical Methods For Purity and Charge Assays

• New methods to detect purity and charge (cSDS, cIEF, CEX, etc.) are used to replace old methods such as SDS-PAGE or IEF method.

• The new assays have higher sensitivity detecting product-related impurities, at release or during stability studies, that were not detected by the original method.

• Use of these new methods raises the question whether the impurities are new or the new assay is better than the old assay at detecting product degradants.
Case Study #1: Change from IEF to cIEF

- Sponsor seeks to replace a qualitative (IEF) assay with a quantitative (cIEF) assay.

- The IEF assay had acceptance criteria of “compares to reference” while the cIEF assay had quantitative acceptance criteria for the major peaks.

- Using these acceptance criteria, stability samples were failing by the IEF assay at earlier time points than by the cIEF assay.

- It appeared that the new assay was not as stability indicating as the old assay.

- The stability failures by IEF appear to be due to the appearance of a particular isoform compared to the reference standard. In case of cIEF same isoform is consistently present at very low levels and is detected by the cIEF, even in the reference standard.
Case Study #1: Replacing IEF to cIEF

Recommendation
The cIEF was allowed for use in stability testing based on the data and risk assessment provided by the sponsor that showed:

• Detection by the IEF was the result of a less than 0.5% change in its level. Because this was considered a ‘change’ from reference, it was considered a stability failure by IEF, but did not actually represent a significant change in product quality.

• Impurity was identified and justified not to impact product efficacy or safety.
Case Study #2: Replacing IEF with cIEF

At the time of licensure, a sponsor seeks to replace an IEF assay with a cIEF assay.

• The new cIEF method was shown to have comparable or better performance compared to the old IEF method.

• There was, however, limited data from drug product lots using the new cIEF method upon which to base quantitative release and/or stability acceptance criteria.

Recommendation
New method run concurrently with old method until sufficient data accumulated with which to establish relevant acceptance criteria.
Case Study#3 : Host cell protein (HCP) assays

HCP assay was updated from a third-party anti-HCP assay to a product specific assay.

• Third party assay: The anti-HCP antiserum is raised against a ‘generic’ cell line (e.g., CHO, NS0, E. coli).

• Specific assay: The anti-HCP antiserum is raised against a cell line that is the same as that used for transfection. Generally a vector transfected cell line.
Commercial versus Specific HCP Assays

Third party HCP Assay Antibody

Specific HCP Assay Antibody

Silver stain of HCP assay standards

Western blots
**Non-Specific Vs. Specific HCP Assays**

<table>
<thead>
<tr>
<th>Specific Assay</th>
<th>Historical process Results</th>
<th>Current process Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Range</td>
<td>421 – 1256</td>
<td>39 – 123</td>
</tr>
<tr>
<td></td>
<td>290 – 644</td>
<td>47 – 76</td>
</tr>
<tr>
<td></td>
<td>154 – 303</td>
<td></td>
</tr>
<tr>
<td></td>
<td>83 – 146</td>
<td></td>
</tr>
<tr>
<td>Avg. ± Std. Dev.</td>
<td>933 ± 271</td>
<td>78 ± 21</td>
</tr>
<tr>
<td></td>
<td>452 ± 112</td>
<td>64 ± 10</td>
</tr>
<tr>
<td></td>
<td>207 ± 36</td>
<td></td>
</tr>
<tr>
<td></td>
<td>118 ± 18</td>
<td></td>
</tr>
<tr>
<td>Third Party Assay</td>
<td>All ≤ LOQ⁴</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td>Range</td>
<td>4 – 106</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td></td>
<td>20 – 74</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td></td>
<td>5 – 13</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td></td>
<td>All ≤ LOQ⁴</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td>Avg. ± Std. Dev.</td>
<td>29 ± 25</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td></td>
<td>49 ± 19</td>
<td>All &lt; LOQ</td>
</tr>
<tr>
<td></td>
<td>8 ± 2</td>
<td>All &lt; LOQ</td>
</tr>
</tbody>
</table>

**Recommendation:** Implement specific assay early in development
Analytical Method Transfer
Analytical Method Transfer

- Method Transfer is managed under an internal transfer protocol that details the parameters to be evaluated in addition to the predetermined acceptance criteria that will be applied to the results.

- A statistically relevant number of test articles are used by the originating and receiving laboratories (e.g., same lots of DS and/or DP lots).

- In cases where the transferred analytical method is also stability indicating; forced degradation samples or samples containing pertinent product-related impurities should be analyzed.
Analytical Method Transfer

The comparative analytical method transfer studies are performed to evaluate:

- Accuracy
- Precision (repeatability and Intermediate precision, e.g., equipment, operators, different days)
Case Studies: Analytical Method Transfer
Case Study #4: Analytical Method Transfer

Following a change in the drug substance (DS) manufacturing process and site change, the SDS-PAGE method for purity of the DS was transferred to new site.

• At the new site a different densitometer was used to quantify protein gels.

• Result on DS lots from the new site with the new densitometer showed a decrease in product purity for the non-reduced SDS-PAGE assay.
Case Study #4: Analytical Method Transfer

• To demonstrate that the decrease in product purity was not due to the process change the sponsor performed side-by-side testing as follow:
  - DS batches produced by new manufacturing process were tested at the previous analytical laboratory site and at the new analytical site.
  - DS lots from original and new manufacturing process were tested at the new analytical site.

• Results from these side-by-side comparisons showed that the apparent decrease in purity was due to the use of the new densitometer and not due to change in product quality.
Case Study #5: Analytical Method Transfer

Sponsor submitted a PAS for the transfer of multiple analytical methods for drug product (DP) testing. The following deficiencies were noted in the method transfer study:

- It did not include comparison on percent monomer - a lot release and stability specification.
- The method transfer included only one analyst at the recipient site.
- The results of the DP lots used to support the transfer of the method used to detect impurities were predominantly reported as $< \text{LOQ}$. No data was provided to demonstrate that LOQ of the transferred method at the recipient site is comparable to the original site.
Case Study #5: Analytical Method Transfer

Recommendations:
- To compare monomer results from both facilities
- Method transfer should include two operators at both sites or provide data from additional testing sites to demonstrate that assay is not subject to operator bias.
- Include analysis of additional lots with detectable impurities. The analysis of spiked samples with known amount of impurities could also be used.
Summary

1. The Lifecycle Management of Analytics involves regular performance trending and evaluation of the capabilities of the method; which may trigger the need for analytical updates and revalidation.

2. If making a major change in how a quality attribute is measured:
   - Assess consistency and comparability across multiple lots.
   - Release and stability data from multiple lots will be required to set commercial specifications.
Summary

3. Comparative method transfer studies should include:
   ▪ assessments of accuracy and precision,
   ▪ forced degradation samples or samples containing relevant product related impurities (for stability indicating assays)

4. A statistically relevant number of DS and/or DP samples should be analyzed in analytical method comparability study.

5. In anticipation of life cycle changes in analytics appropriate number of DS and DP samples representative of pivotal and marketed product should be archived to allow for comparative studies.
References on Analytical Methods Validation

1. ICH Guidelines Q2(R1) [2005]
2. FDA draft Guidance on Analytical Procedures and Methods Validation [2000]
4. FDA -GFI: Changes to an Approved Application for Specified Biotechnology and Specified Synthetic Biological Products [1997]
5. FDA guidance on Content and format of IND for Phase 1 studies of Drug, including Well-Characterized, Therapeutic, Biotechnology products [2000]
6. FDA-GFI: INDs for phase 2 and phase 3 studies, CMC Information [2003]