Intro to Dissolution Validation

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Overview

- Validation
- What needs to be done prior to Validation?
- Validation of Dissolution Method
- Validation of Analytical Finish
- Other Potential Validation Elements

Why Validate?

- Meet Regulatory Requirements
- To Ensure Method is Suitable for Use
  - Accurate
  - Precise
  - Able to generate meaningful data
  - Reliable
  - Transferrable

Aspects of Validation

Validation must be performed on all aspects of the dissolution method

- Dissolution Method itself
- Analytical Finish
- Automation and other potential changes

USP Requirements for Dissolution Validation

Dissolution is a Category III Test in USP <1225> Validation of Compendial Methods and Requires:

- Accuracy
- Precision
- Specificity
- Detection Limit
- Quanititation Limit
- Linearity
- Range

Pre-Validation Activities

- Dissolution and Analytical Method Development Completed
- 3+ batches of formulation ready for analysis
- SOP written w/ proper level of detail
- Validation Protocol written and approved
- Pre-validation check?

Dissolution and Analytical Method Development

- Dissolution Method has been made for all dosages
- Analytical Finish is defined
- Appropriate filter(s) have been found
- Method has been tested on various formulations good and bad
- Sensitive aspects of method have been identified

Formulation Batches

- At least 3 formulations have been created which can be used for the validation
- Formulation development is often run concurrently with the dissolution method

SOPs

SOPs need to be written so that they properly define the important aspects of a method BUT are not too limited so as to cause future issues. Of particular concern should be:

- Equipment used
- Filters
- HPLC columns
- Sinkers

“Or Equivalent”

The use of the term “or equivalent” is often used to widen the SOP for future changes

Can be problematic as items advertised as equivalent, may not be equivalent

Better term may be “or other validated…”

The Validation Protocol

The Validation Protocol needs to be written and accepted prior to beginning validation. The protocol needs to include:

- All elements to be validated
- Acceptance criteria for each of the elements
- Empty tables to be filled out

Pre-Validation Checks

It may be advisable to perform a mini pre-validation protocol prior to the official Validation

- Sniff out potential issues
- Determine if acceptance criteria may need to be changed
- Less Regulatory Issues

Much like a dress rehearsal…

Validation Parameters

What are the components of validation?

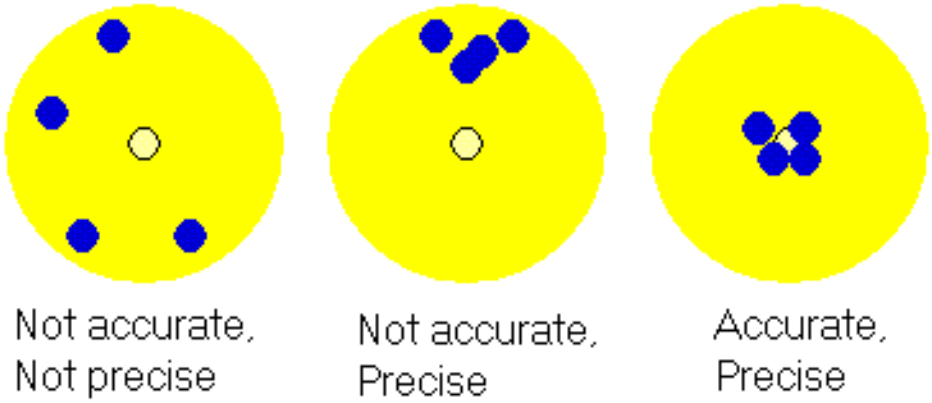
- Accuracy
- Precision
- Specificity
- Detection Limit
- Quanititation Limit
- Linearity
- Range
- Robustness
- Filter Validation
- Standard Stability
- Solution Stability

Accuracy and Precision

Accuracy is the closeness of the results to the actual value

For Dissolution, the sample analysis must give an exact result to gives an accurate value for the amount dissolved at a given time.

Precision is the agreement of individual test results when the method is repeated with multiple samples of the same homogeneous solution



Precision

Precision in an analytical method validation is typically referring to both intermediate precision or repeatability

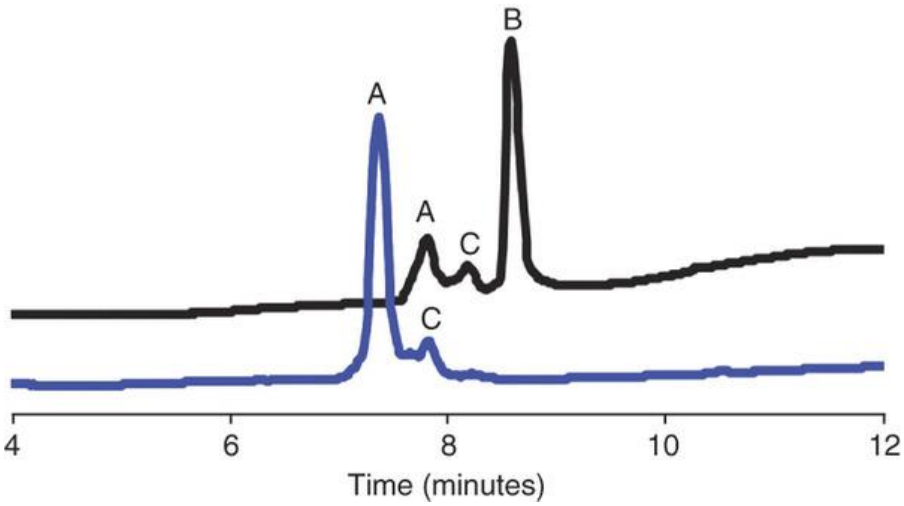
Intermediate precision usually assesses the method with variations to be expected within a lab (different analysts, equipment, etc.)

Repeatability looks at multiple measurements of the same sample by the same user, equipment, etc. over a short period of time.

Specificity

Specificity is the ability to determine the amount of API in the presence of other components that may be expected to be present such as:

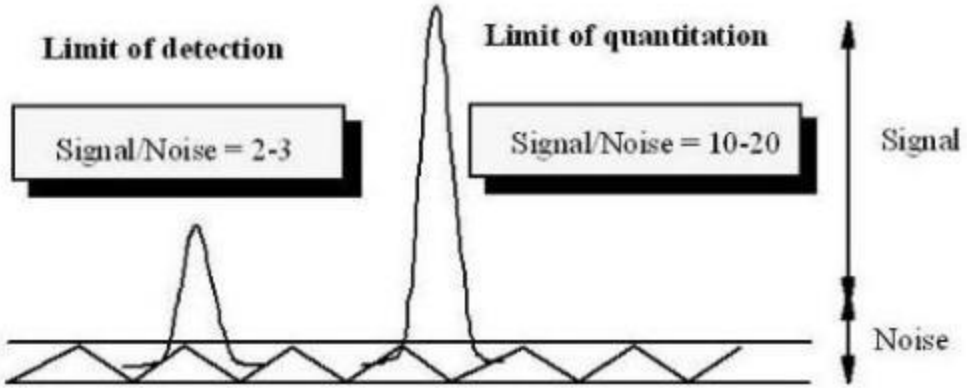
- Impurities
- Degradation products
- Excipients



Detection and Quantitation Limit

Detection Limit (LOD) is the lowest amount of your compound which can be detected, but not necessarily measured. Typically set at 3:1 signal to noise ratio.

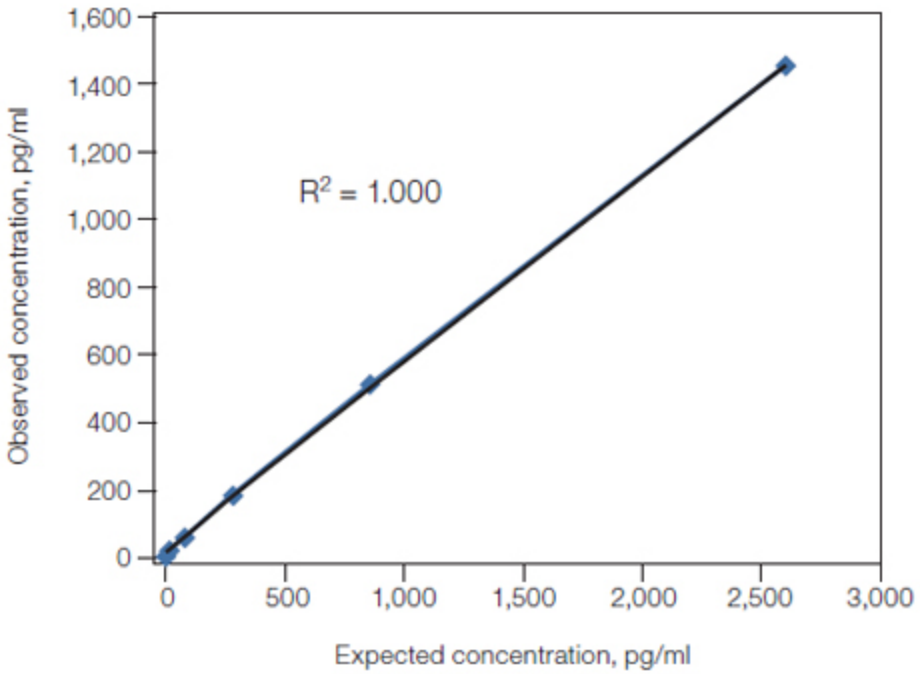
Quanititation Limit (LOQ) is the lowest amount of your compound which can be measured with acceptable prevision and accuracy. Typically 10:1 signal to noise ratio.



Linearity and Range

Linearity is the ability to elicit test results which are directly (or by a well-defined mathematical transformation) proportional to the concentration of analyte within a given range

Range is the interval between the highest and lowest levels on analyte to be determined with appropriate accuracy, precision, and linearity

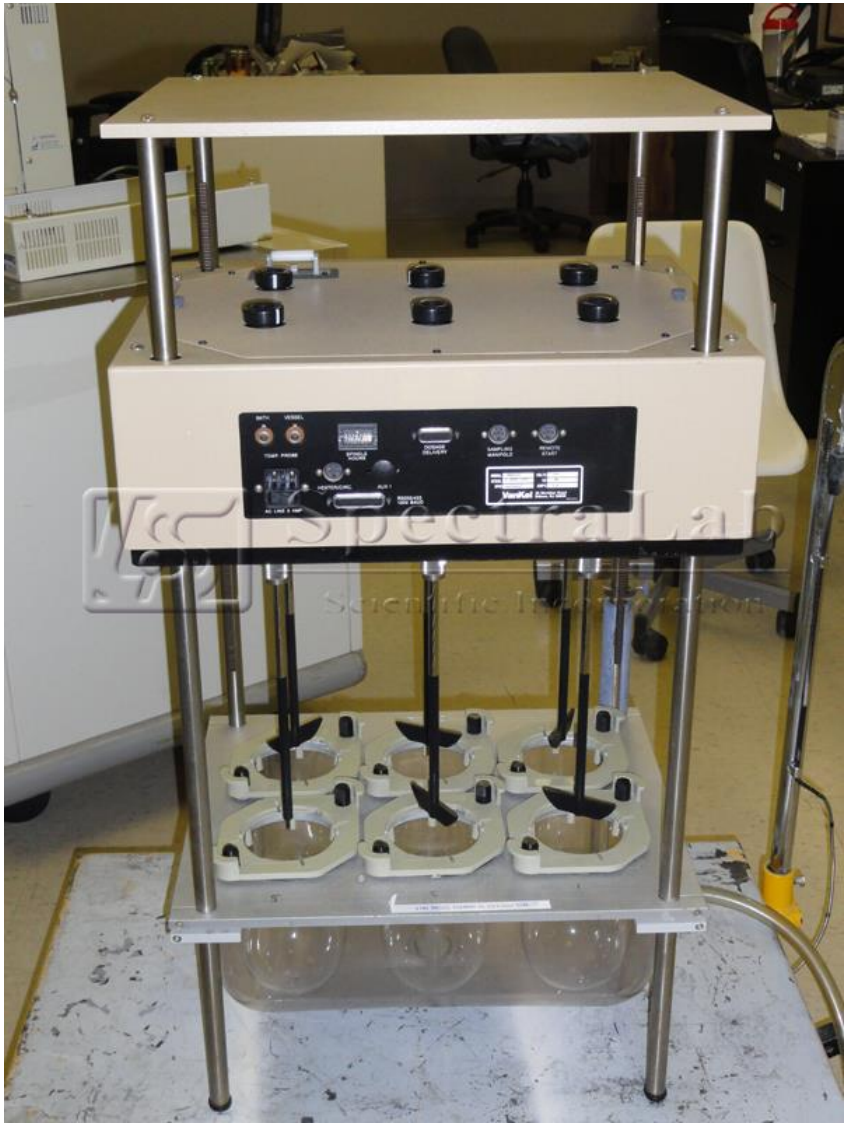


Robustness

Robustness is the ability of a method to be unaffected by small variations in the method parameters

Robustness helps to show the reliability of the method during normal usage

- Buffer concentration
- RPM speed
- Mobile phase preparation
- Dissolution unit?



Stability and Filter Validation

Stability is the determination of the length of time which the samples and standards yield equivalent results as when prepared fresh.

Standards and samples should be validated separately, as matrixes may have an impact

Filter validation ensures that a bias is not created by the filtration method through leaching of chemicals from the filter, inadequately filtering out undissolved API, or adsorbing dissolved drug material.

Performing the Validation

Validation layout

The validation should be planned in a way that:

- Establishes accuracy and precision of analysis first
- Samples can serve more than one function where possible
- Filters are validated before used on dissolution samples

Quarantine of Samples

All samples and standards should be kept until Validation Protocol has been completed and signed off even if:

- Tests passed
- Past expiration date
- Taking up too much room/need glassware

Method Validation is often the strictest cGMP activity in a lab, and proper investigation ability is crucial

Linearity and Range

Linearity should be evaluated to accommodate all dosage strengths

Minimum value: 25% below specification of % Q for lowest dose, first timepoint

Maximum value: 120% or higher of label claim for the highest dose

The minimum and maximum values of linearity become the range of the method

Linearity

- Should have at least 5 linearity levels
- R-squared value of >0.98
- If higher or lower dosage strengths are possible in the future, you may want to extend the range very wide at this point to open the validation for future products

Linearity

To ensure linearity is based on the instrument and not analyst error, prepare solutions from a single stock solution.

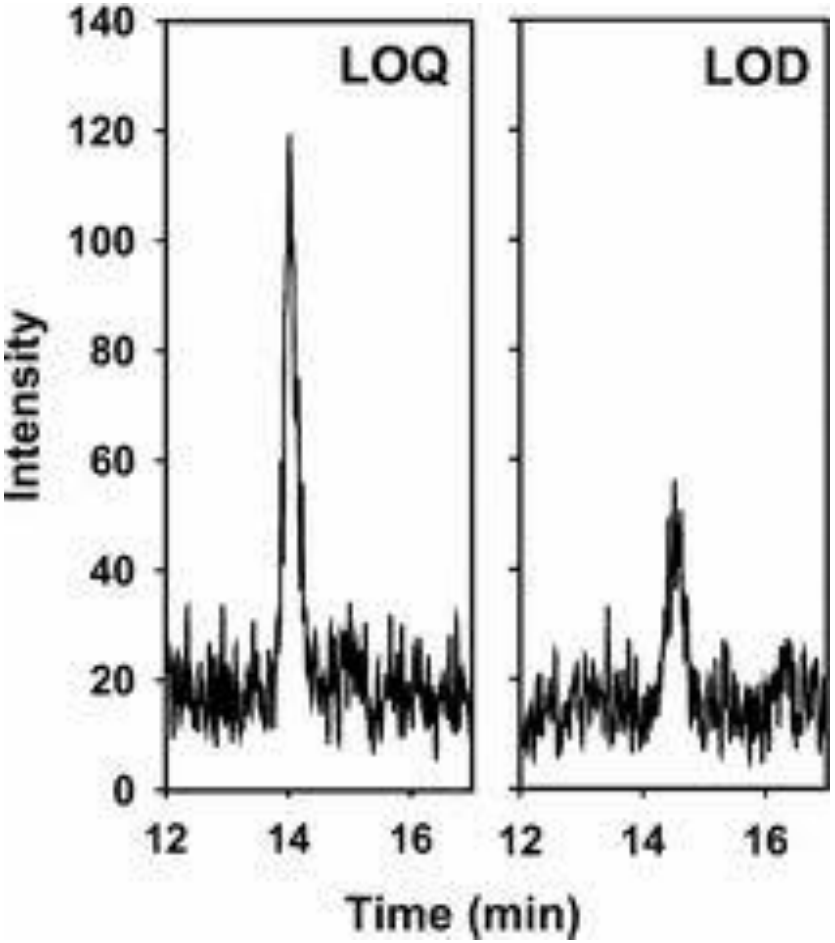
Stock solution may be 10x concentration for example, and create dilutions accordingly.

LOD and LOQ

You can further dilute the same solutions from the linearity study to determine your LOD and LOQ concentrations

LOD typically 3-5 x noise

LOQ typically 10-15 x noise



Accuracy

- Test Accuracy at 3 levels w/ 3 replicates
- Should be done w/ Analyst 1 and 2
- One of the two analysts can use same stock from linearity
- Acceptance Criteria
  - Overall mean 100% +/- 3% >50% Q
  - Overall mean 100% +/- 5% <50% Q

Standard Stability

The 100% LC solution from linearity can also act as the Day 0 Standard Stability. Aliquots of this solution should be split and stored at:

- Lab bench in clear volumetric flask
- Lab bench in amber volumetric flask
- In cold room at ~4C

These aliquots will be compared against fresh standards at various intervals to determine standard stability

Standard Stability

Standards are typically considered stable as long as they have >98% recovery against a fresh standard

Depending on the length of time desired, you may need special storage conditions.

Days	Room	4C	Amber
1	100%	100%	100%
2	99%	100%	99%
3	99%	100%	99%
5	98%	100%	99%
7	98%	99%	98%
12	95%	99%	96%

Plan for retesting/investigational time

Precision – Spiked Placebo

- Precision is generally performed as n=3 by 2 different analysts
- Perform these with spiked placebo samples
- Spiked placebo should contain everything in the formulation matrix
- If performing w/ multiple strengths, a worst-case scenario may save time vs. separate studies for each dose

Precision – Spiked Placebo

- Determine Placebo Mix
  - Order may be important
  - Necessary to record weights, etc.
- Once placebo is created, put mixture into each volumetric flask
- Spike the placebo with known levels of drug as with a standard
  - 120% level
  - 100% level
  - 10-50% level

Precision – Spiked Placebo

You should perform the check with the same filter being used in the dissolution analysis

Make sure filter is validated for use for adsorbance

Acceptance criteria should be <3% RSD

Specificity

The Spiked Placebo samples can also be used to assess specificity of the method

Look for non-parent peaks and potential interferences

Any interference should be <1%

Compare to blanks of media as well

Filter Validation

Filter Validation should challenge all aspect of the filter

- The filter is efficient in removing undissolved active pharmaceutical ingredients (API) from a sample solution
- The filter does not interfere with UV spectra at the wavelength of measurement
- The filter does not adsorb drug product

Filter Validation - Efficiency

Performed to determine if the filter effectively removes undissolved API from the dissolution media and stops the dissolution process of the sample

The Importance of Filter Validation

Example of an efficiency test:

- Prepare a sample solution of ~50% of nominal analytical concentration
- Filter three aliquots through separate filters and dispense each into a separate test tube
- Analyze one sample immediately, one sample after 5 minutes of ultrasonication, and one sample after 10 minutes of sonication
- Filter passes efficiency if sonicated samples show less than a 2% increase in dissolved sample than the non-sonicated sample

Filter Validation

Performed to ensure that the filters used do not contribute to the UV spectra at the wavelength of measurement

Example of a leachability test:

- Prepare a working standard solution at 100% nominal concentration (unfiltered)
- Prepare a dissolution media blank and filter three samples through separate filters
- Analyze three aliquots of unfiltered standard solution and three aliquots of filtered dissolution media
- Leachability test passes if response of each of the filtered blanks is less than or equal to 1% of the mean standard response

The Importance of Filter Validation

Adsorbance Validation:

Performed to ensure that filters do not adsorb dissolved API and artificially lower results. Typically, a validated method states that a specific volume (usually 3-5 mL) be discarded prior to collecting sample for analysis.

The Importance of Filter Validation

Example of an adsorbance test:

- Prepare a working solution at the lowest nominal concentration (% released at 1st timepoint)
- Withdraw 10mL of working solution in triplicate through separate syringes equipped with cannulas
- Place syringe filters at the end of each syringe and dispense in 1mL increments
- Analyze an unfiltered sample, and each 1mL filtered sample
- Determine the % recovery for each filtered sample compared to the unfiltered sample

The Importance of Filter Validation

$$\% \text{ Recovery} = R_{sam}/R_{std} \times 100$$

Rsam = Response of Sample

Rstd = Response of Standard

- Determine the volume needed to flush through the filter so that remaining aliquots will have a recovery 98% -102%
- At a minimum, the final 5mL should have recovery levels between 98% - 102%

Intermediate Precision

Intermediate Precision is the determination of variation within a lab and ensures that the method is precise despite normal changes

Test should be performed with as many differences as possible between Analyst 1 and Analyst 2

What Should Be Different?

- Dissolution Apparatus
- UV/HPLC used (including column)
- Dissolution Media
- Dissolution Standards
- Different Days

Intermediate Precision

- N=6 for each chemist for each strength/lot, for total of n=12
- Should have at least 3 timepoints and infinity point
- Criteria
  - Usually <6% RSD between analysts
  - May need to adjust timepoints and criteria depending on dosage form

Intermediate Precision – Other uses

In addition to assessing the precision of the dissolution method itself, this test can also be used to help assess changes in the method instead of/in addition to f2:

- Automation
- Degassing
- Sinkers

Solution Stability

Solution Stability is performed in the same way as the standard stability

Compare same sample over multiple days in comparison to freshly prepared standards

Robustness

Robustness is generally established through method development

- RPM changes
- Various dosage strengths
- pH differences
- Media differences

Anything where there is a specific sensitivity in the method may need to be validated

Method Validation Completion

- Method Validation report is ideally filled out as you go along
- Upon completion, all tests and the report should be fully reviewed
- SOP for method can be signed off after Validation is fully accepted
- Training on the method should then occur for the appropriate personnel

Future Changes Anticipated?

If there is a change in the product/method which deviates from what was tested in the validation report, some sections may need to be revalidated or investigated

- Different dosage strengths
- Reformulation
- Use of Dissolution Automation
  - Samplers
  - Online UV
  - Fiberoptics

Questions?