Supplementary Training Modules on Good Manufacturing Practice

Validation

WHO Technical Report Series, No. 937, 2006. Annex 4.

Validation

- Part 1. General overview on qualification and validation
- Part 2. Qualification of HVAC and water systems
- Part 3. Cleaning validation
- **Part 4. Analytical method validation**
- Part 5. Computerized system validation
- Part 6. Qualification of systems and equipment
- Part 7. Non sterile product process validation

Supplementary Training Modules on Good Manufacturing Practice

Analytical Method Validation

Part 4

WHO Technical Report Series, No. 937, 2006. Annex 4 Appendix 4

Validation

Objectives

To discuss various aspects of analytical method validation including:

- Principles of analytical method validation
- Pharmacopoeia methods
- Non-pharmacopoeia methods
- Approaches to analytical method validation
- Characteristics of analytical procedures

Validation

Principle

- Guideline presents information on the characteristics to be considered (other approaches may be followed)
- Manufacturers to demonstrate - analytical procedure is suitable for its intended purpose (validation)
- Validate analytical methods - whether they indicate stability or not
- Validated by R&D before being transferred to the quality control unit when appropriate

1.1 – 1.4

Validation

General

- Specifications for materials and products, with standard test methods
- Manufacturer to use “pharmacopoeial specifications and methods”, or suitably developed “non-pharmacopoeial specifications and methods” as approved by the national drug regulatory authority
- Use well-characterized reference materials, with documented purity, in the validation study

2.1 – 2.3

Validation

General (2)

- Tests include:
  - *identification tests*
  - *assay of drug substances and pharmaceutical products*
  - *content of impurities and limit tests for impurities*
  - *dissolution testing and determination of particle size*
- Results should be reliable, accurate and reproducible

2.4 – 2.5

Validation

General (3)

- When should verification or revalidation be done?
  - *changes in the process for synthesis of the drug substance*
  - *changes in the composition of the finished product*
  - *changes in the analytical procedure*
  - *transfer of methods from one laboratory to another*
  - *changes in major pieces of equipment instruments*
- Extent depends on the nature of the change(s)
- Evidence of “analyst proficiency”

2.6 – 2.8

Validation

Pharmacopoeial/Non-pharmacopoeial methods

- **Pharmacopoeial methods:**
  - *prove that the methods are suitable for routine use in the laboratory (verification)*
  - *for determination of content or impurities in products, demonstrate that method is specific for the substance under consideration (no placebo interference)*
- **Non-pharmacopoeial methods:**
  - *Should be appropriately validated*

3. – 4.

Validation

Method validation

- Protocol: includes procedures and acceptance criteria
- Report: documented results
- Justification needed when non-pharmacopoeial methods are used (if pharmacopoeial methods are available). Justification to include data, e.g. comparisons with the pharmacopoeial or other methods
- Detailed standard test methods include:
  - *chromatographic conditions, reagents and others*

5.1 – 5.3

Validation

Characteristics that should be considered during validation of analytical methods include:

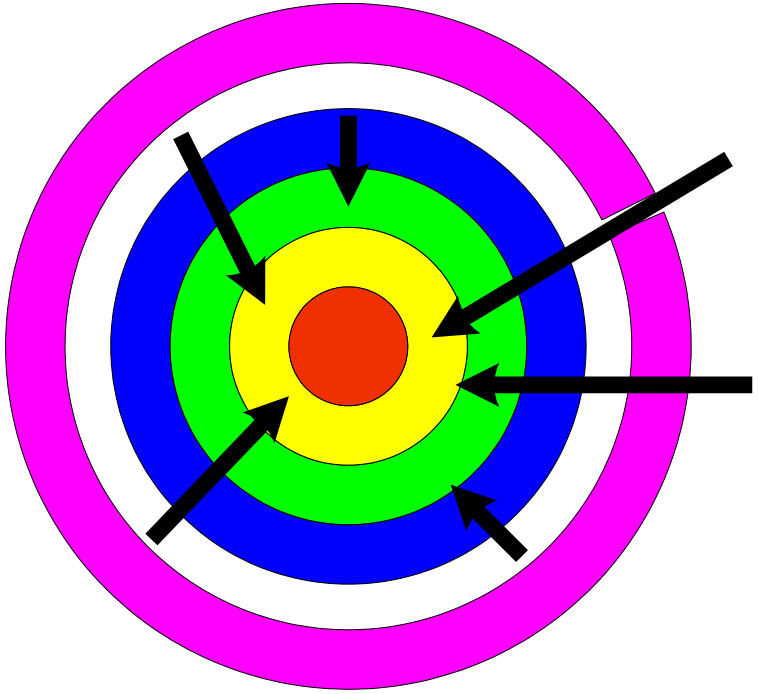
- *specificity*
- *linearity*
- *range*
- *accuracy*
- *precision*
- *detection limit*
- *quantitation limit*
- *robustness*

6.1

Validation

Accuracy: is the degree of agreement of test results with the true value, or the closeness of the results obtained by the procedure to the true value. It is normally established on samples of the material to be examined that have been prepared to quantitative accuracy. Accuracy should be established across the specified range of the analytical procedure.

Note: it is acceptable to use a "spiked" placebo where a known quantity or concentration of a reference material is used.



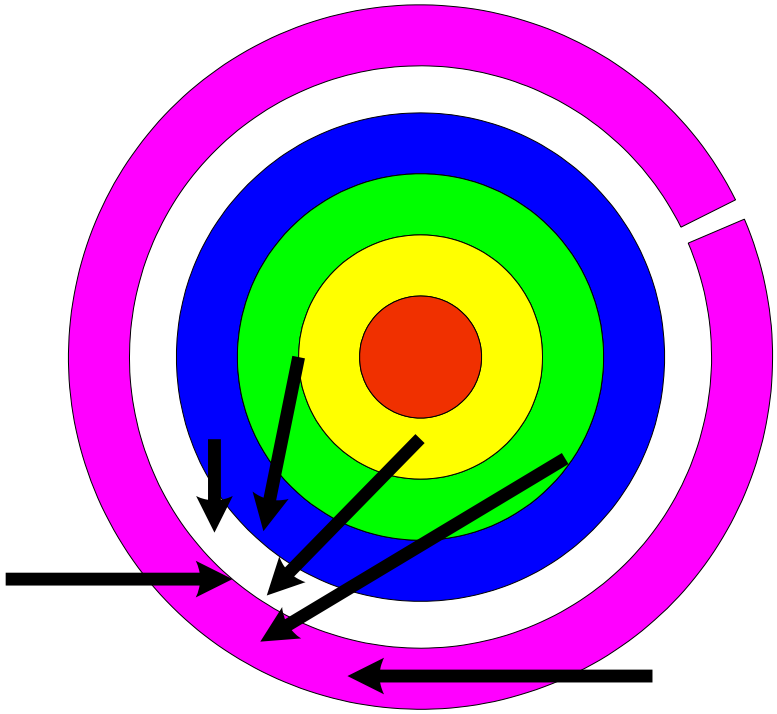
Accurate but imprecise

6.1.1

Validation

Precision: is the **degree of agreement among individual results**. The complete procedure should be applied repeatedly to separate, identical samples drawn from the same homogeneous batch of material. It should be measured by the scatter of individual results from the mean (good grouping) and expressed as the relative standard deviation (RSD).

6.1.2



Inaccurate but precise

Validation

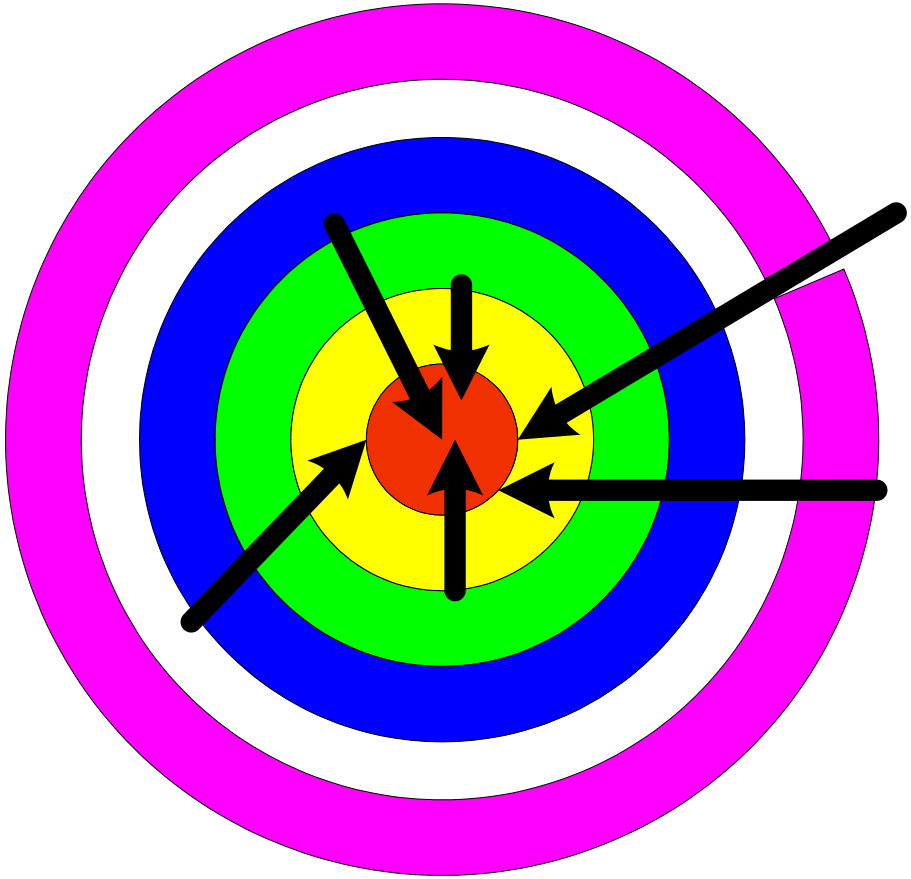
Precision:

- *Repeatability:* A minimum of nine determinations covering the specified range for the procedure, e.g.
  - *three concentrations/three replicates each, or a minimum of six determinations at 100% of the test concentration*
- *Intermediate precision:* Within-laboratory variations
  - *usually on different days, different analysts and different equipment. (If reproducibility is assessed, a measure of intermediate precision is not required.)*
- *Reproducibility: Precision* between laboratories

6.1.2.1 – 6.1.2.3

Validation

Accurate and Precise



Validation

***Robustness* (or *ruggedness*):**

is the ability of the procedure to provide analytical results of acceptable accuracy and precision under a variety of conditions. The results from separate samples are influenced by changes in the operational or environmental conditions. Robustness should be considered during the development phase, and should show the reliability of an analysis when deliberate variations are made in method parameters.

6.1.3

Validation

Factors that can have an effect on robustness when performing chromatographic analysis include:

- *stability of test and standard samples and solutions*
- *reagents (e.g. different suppliers)*
- *different columns (e.g. different lots and/or suppliers)*
- *extraction time*
- *variations of pH of a mobile phase*
- *variations in mobile phase composition*
- *temperature*
- *flow rate*

6.1.3.1

Validation

Linearity:

indicates the ability to produce results that are directly proportional to the concentration of the analyte in samples

- *A series of samples should be prepared in which the analyte concentrations span the claimed range of the procedure. If there is a linear relationship, test results should be evaluated by appropriate statistical methods*
- *A minimum of five concentrations should be used*

6.1.4

Validation

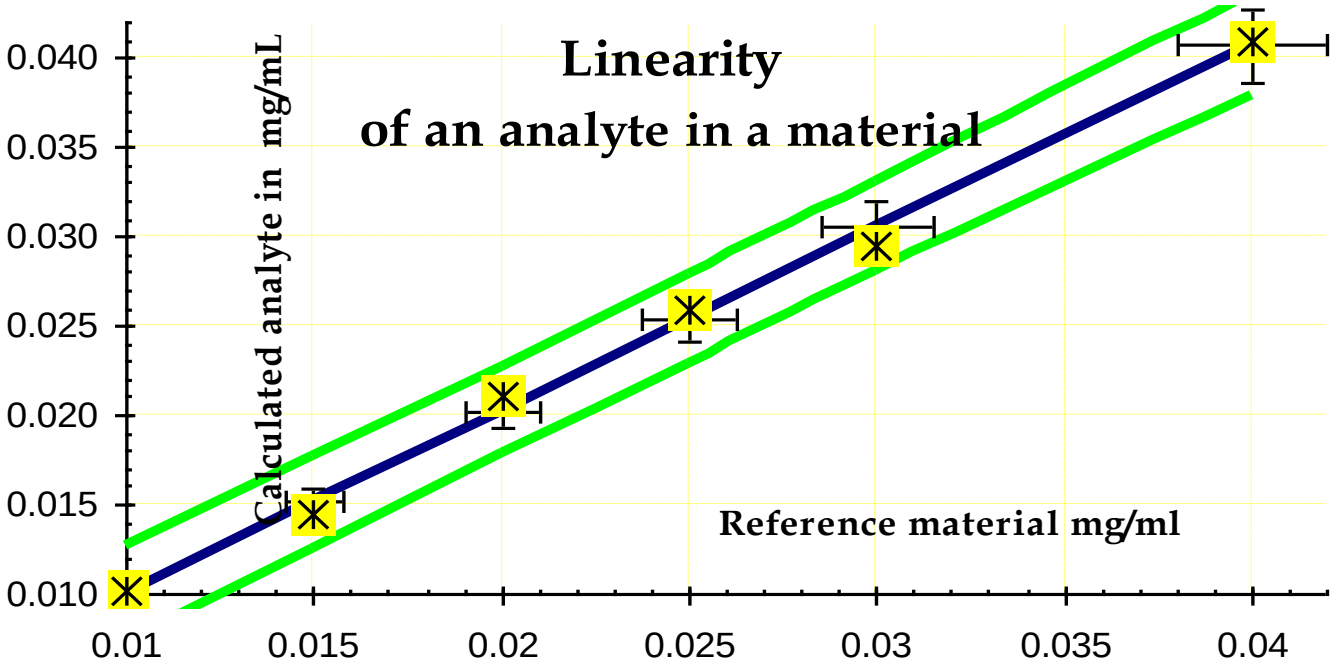


Table of values (x,y)

#	x Reference material mg/ml	y Calculated mg/ml
1	0.0100	0.0101
2	0.0150	0.0145
3	0.0200	0.0210
4	0.0250	0.0260
5	0.0300	0.0294
6	0.0400	0.0410

Validation

Range:

is an expression of the lowest and highest levels of analyte that have been demonstrated to be determinable for the product

The specified range is normally derived from linearity studies

6.1.5

Validation

***Specificity* (*selectivity*):**

is the ability to measure unequivocally the desired analyte in the presence of components such as excipients and impurities that may also be expected to be present.

An investigation of specificity should be conducted during the validation of identification tests, the determination of impurities and assay

6.1.6

Validation

***Detection limit* (*limit of detection*):**

is the smallest quantity of an analyte that can be detected, and not necessarily determined, in a quantitative fashion

- Approaches (instrumental or non-instrumental):
  - *visual evaluation*
  - *signal to noise ratio*
  - *standard deviation of the response and the slope*
  - *standard deviation of the blank*
  - *calibration curve*

6.1.7

Validation

***Quantitation limit* (*limit of quantitation*):**

is the lowest concentration of an analyte in a sample that may be determined with acceptable accuracy and precision

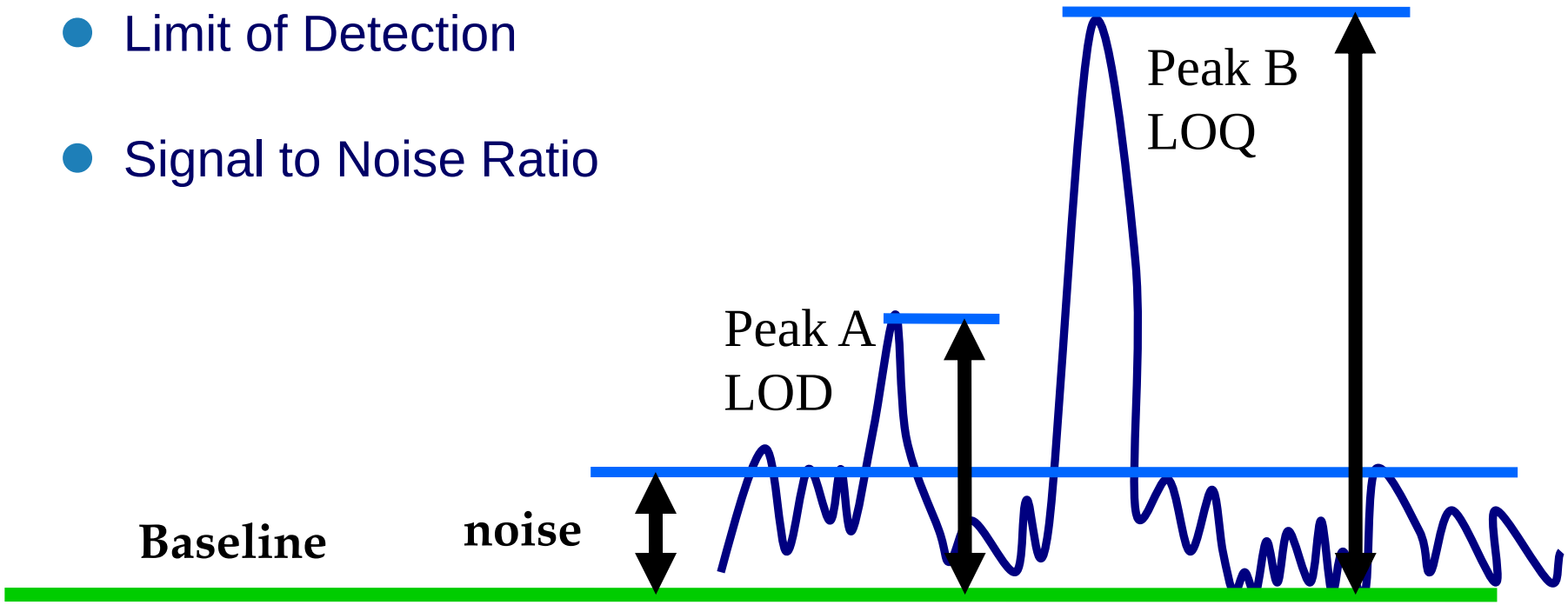
- Approaches (instrumental or non-instrumental):
  - *visual evaluation*
  - *signal to noise ratio*
  - *standard deviation of the response and the slope*
  - *standard deviation of the blank*
  - *calibration curve*

6.1.8

Validation

LOQ, LOD and SNR

- Limit of Quantitation
- Limit of Detection
- Signal to Noise Ratio



World Health Organization

Validation

Table 1. Characteristics to consider during analytical validation

6.2

Validation

System suitability testing

- Integral part of many analytical procedures
- Based on the concept that:
  - *the equipment, electronics, analytical operations and samples to be analysed constitute an integral system that can be evaluated as such*
- Depends on the type of procedure being evaluated
  - *e.g. resolution test for an HPLC procedure*

6.3

Validation

- Group session