Validation of Thin Layer Chromatographic Procedures

International Symposium for High-Performance Thin-Layer Chromatography

HPTLC 2011

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Topics

- ICH Q2(R1) and nothing else?
- Other guidance documents describing analytical validation
- How to select the appropriate validation approach
- Common pitfalls when adopting Q2(R1)
- Measurement Uncertainty

ICH Q2(R1) – Analytical Validation

- ICH Q2(R1), Validation of Analytical Procedures: Text and Methodology, ICH, Geneva, 2005
- Originally issued as Q2A “Validation of Analytical Procedures: Definitions and Terminology” (adopted October 1994) and Q2B “Validation of Analytical Procedures: Methodology” (adopted November 1996)
- Initially issued as
  - “... a ***discussio***n of the characteristics for consideration during the validation of the analytical procedures included as part of registration...”
- Now emerged to be a questionable “Gold Standard”?

ICH Q2(R1) – Analytical Validation

- ICH = International Conference on Harmonisation
- Launched in 1990, bringing together the drug regulatory authorities and pharmaceutical industry associations of Europe, Japan and the United States
- Mission: Harmonisation of the requirements for pharmaceutical product registration
- ICH has issued various Guidelines on
  - Quality
  - Safety
  - Efficacy
- These guidelines are consensus documents that ***leave room for individual considerations and approaches***

ICH Q2(R1) – Analytical Validation

- In case of ICH Q2(R1) ***better*** Guidelines on analytical validation of various chemical analytical organisations (ISO/IUPAC/AOAC; EURACHEM) have unfortunately ***not been considered*** in the ICH process
- Although initially developed to cover (synthetic) APIs and finished products with known, well characterised matrix and tight expectations of assay and content of potential impurities...
- ...ICH Q2(R1) (or the very similar USP Chapter <1225>) approach is ***blindly*** used whenever a method validation is required
  - "Cooking receipt approach"
  - No more critical reflection on a method's purpose and the required performance characteristics → simply follow ICH Q2(R1) or USP Chapter <1225>.

Why validated a procedure?

- "The objective of validation of an analytical procedure is to demonstrate that it is suitable for its intended use“

  (ICH Guideline Q2(R1): “Validation of Analytical Procedures: Text and Methodology”)

- ***No to show how good your laboratory works!***

- ***The intended use of a procedure decides on the approach to be taken and the acceptance parameters***

Why validated a procedure?

The ***classical inadequate*** description of the objective...

- "The method has been developed to determine XXX in bulk drug and in pharmaceutical dosage form"

The ***theoretical correct*** description of the objective...

- "The method has been developed to determine XXX in a sustained release tablet formulation containing ... as excipients, 40 mg XXX, with a manufacturing capability of assuring ± 1.5% accuracy of potency. This requires an assay procedure with a long term uncertainty of nmt 1.5% to allow control of the specification limits for assay of ± 5% as expected by the European regulatory authorrities"

Other appropriate Validation Guidelines

Bioassays:

- Analytical Methods Validation: Bioavailability, Bioequivalence and Pharmacokinetic Studies
  - Journal of Pharmaceutical Sciences, Vol 81, No. 3, March 1992
- FDA Guidance for Industry: Bioanalytical Method Validation, 2001

Herbals and dietary supplements

- Validation of Standardized High-Performance Thin-Layer Chromatographic Methods for Quality Control and Stability Testing of Herbals
  - Kathrin Koll, Eike Reich, Anne Blatter, Markus Veit

    Journal of AOAC International, Vol. 86, No. 5, 2003, 909

Other appropriate Validation Guidelines

General Guidance:

- Guidelines for collaborative study procedure to validate characteristics of a method of analysis, J. Assoc. Off. Anal.Chem. 72 (1989) 694
- International Union of Pure and Applied Chemistry: Harmonized Guidelines for Single Laboratory Validation of Methods of Analysis (IUPAC Technical Report)

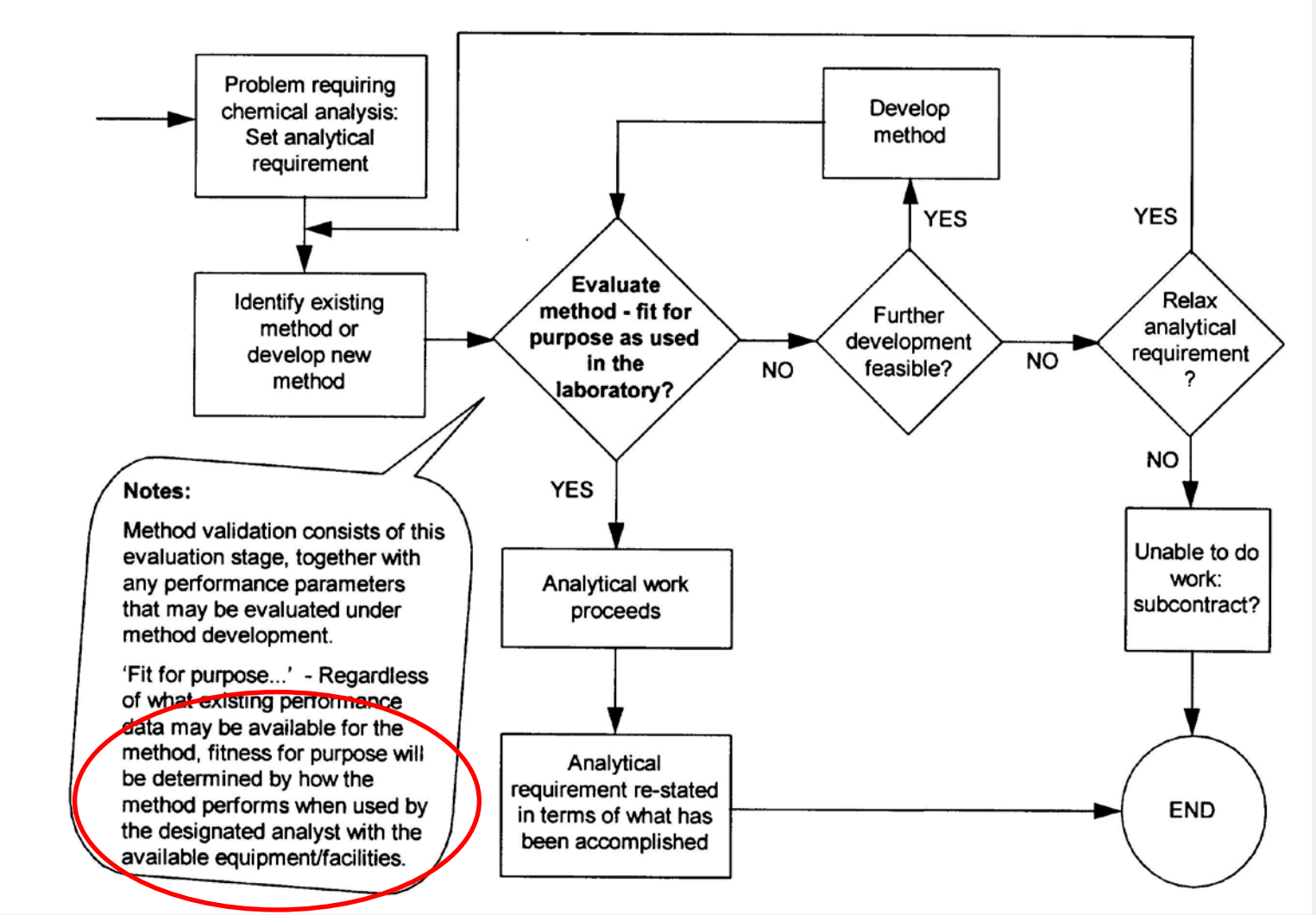
The most comprehensive literature

- W. Funk, V. Damman, G. Donnevert, Quality Assurance in Analytical Chemistry, ECH, Weinheim, Germany, 1995.

How to validated a procedure?

- Clearly define the intention of your method/procedure and select appropriate Guidance to follow
- Define performance characteristics important to quality and expected performance parameters to be fulfilled
- Development of a procedure and its validation is an iterative process!
- The procedure‘s suitability must be studied in initial validation experiments.
- There is no way to first develop a method and later on validate it as indicated in ICH Q2(R1)
- If these preliminary validation data are inappropriate, either the procedure and the basic technique itself, the equipment or the acceptance criteria have to be changed!
- Robustness tests are part of this development phase.

IUPAC Development - Validation Cycle

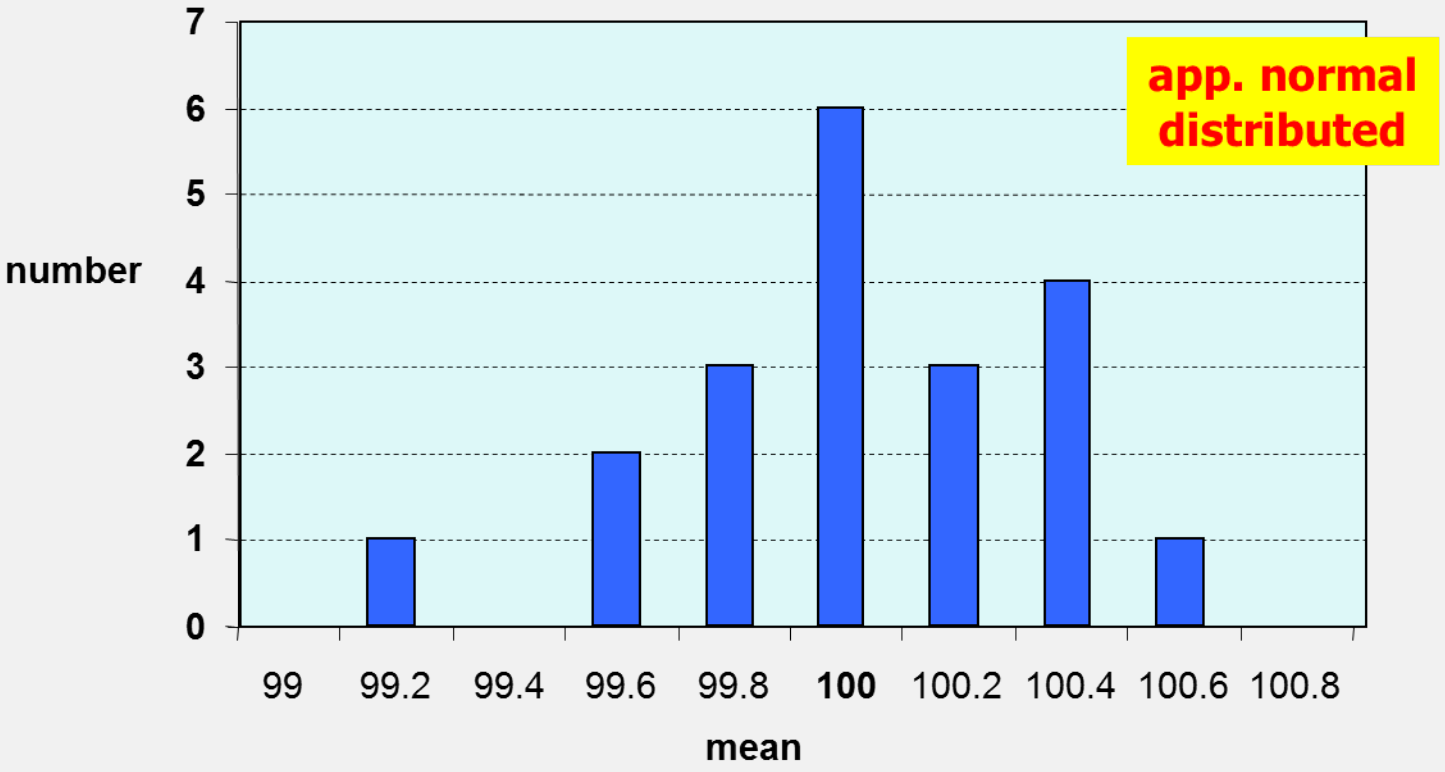


Disadvantages of ICH Q2(R1)

- Overemphasising linearity of calibration model – most analytical procedures show non-linear calibration models
- As a consequence, HPTLC procedures are mainly developed using one point calibration instead three point calibration as required by European Pharmacopoeia
- Isolated consideration and determination of a procedure's precision (repeatability/intermediate precision) and its accuracy
- Does not at all reflect the IUPAC approach of assessing a procedure's overall uncertainty to be expected on a long range
- May lead to a false sense of "good" method precision and therefore unjustified adoption of HPTLC or TLC procedures for tasks they are not suited for.
- Robustness tests are mentioned, but not included into the tables

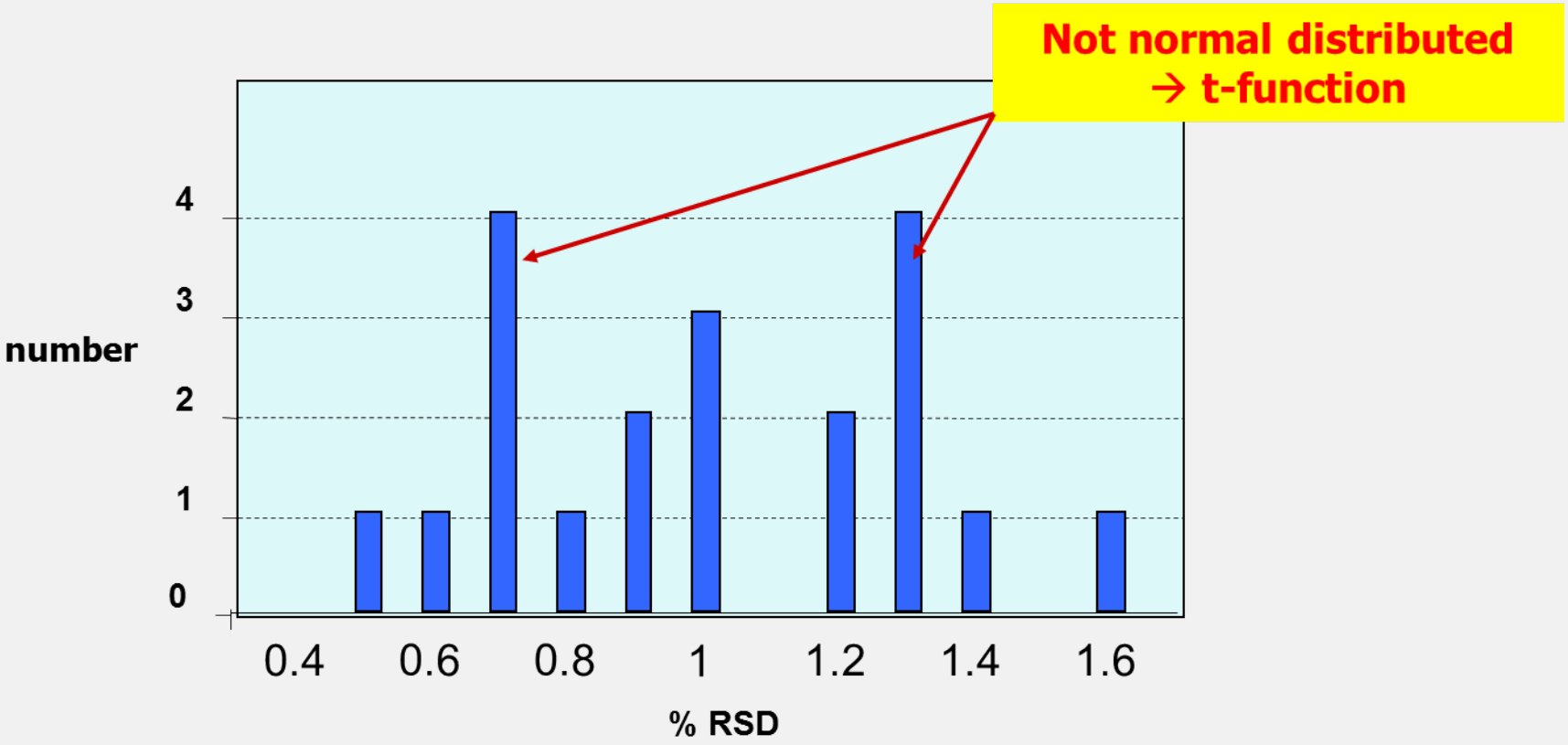
Validation – Precision – Mean

- Simulated data: 20 x mean, calculated each time from 6 randomly generated replicate “measurements”, mean = 100, RSD = 1.0 %



Validation – Precision – RSD

- Simulated data: the 20 corresponding RSDs of the 20 x 6 replicates

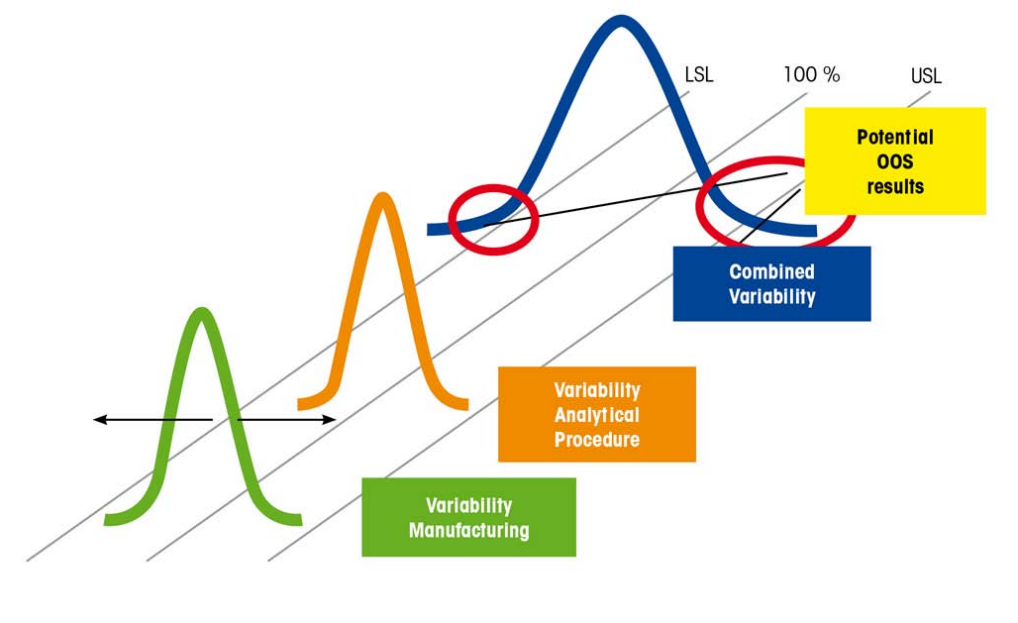


B. Renger 2001

The Concept of Measurement Uncertainty

- Measurement uncertainty is a ***statistical parameter*** describing the possible fluctuations of the result of a measurement ***over time***.
- Measurement uncertainty can be determined by the addition of the ***variances of the individual steps of a given analytical method*** or by an approach which starts with a method's intermediate precision.
- Adapted to ***chemical analytical measurements***.
  - Eurachem/CITAC Guide: Quantifying uncertainty in analytical measurement, 2nd edition, S.L.R. Ellison, M. Rösslein and A. Williams, (Eds.), 2000,
  - http://www.eurachem.org/guides/pdf/QUAM2000-1.pdf
- Reporting a result's analytical uncertainty is mandatory according to Paragraph 5.4.6 of ISO norm 17025 "General requirements for the competence of testing and calibration laboratories"
- Takes into consideration not only the random errors, but also sources of systematic errors (bias)

Process Capability and Variability



- Overall variability of a ***given analytical system*** is combination of (manufacturing) process capability and analytical variability/uncertainty
- Low concentrations of the analyte in the product will increase overall variability
- As a consequence, more OOS results originating from mere statistical reasons have to be expected

Typical Analytical Uncertainty

Analytical Technique	SST (n = 6) RSD [%]	Intermediate Precision (n = 6) RSD [%]	Long-term Uncertainty RSD [%]
HPLC, automat.	0.4 – 0.5	0.6 – 0.8	0.9 – 1.1
HPLC, manual	0.7 – 1.0	1.1 – 1.5	1.6 – 2.2
GC, direct injection	~ 1.0	1.5	2.2
GC, headspace	~ 1.6	2.3	3.5
CE	~ 1.0	1.5	2.2
HPTLC	1.4 – 1.9	2.1 – 2.9	3.2 – 4.3

Meyer, Küppers, Renger, LCGC 2000; Renger J. Chrom. B, 2000; Layloff AG/PT, 2002;
Wätzig, Ermer, PZ Prisma 2003, Wätzig Chromatographia, 2005

Summary

- Validation of procedures should be based on the intended use and the guidance to follow selected appropriately
- ICH Q2(R1) or USP Chapter <1225> have been developed for a very narrow range of applications in pharmaceutical industry and must not be considered a mandatory standard
- If validation is performed according to ICH Q2A(R1) or USP Chapter <1225> it should be performed to report a procedure's true long term variability, not to show "how good the laboratory works".
- Adoption of some elements of the concept of measurement uncertainty helps to understand the true analytical variability/uncertainty of pharmaceutical analytical methods.

Summary

- Recurrent failures in manuscripts describing validation of quantitative TLC/HPTLC procedures in pharmaceuticals have been addressed, but results up to date are not very encouraging
  - Katalin Ferenczi-Fodor, Bernd Renger*, and Zoltán Végh, Journal of Planar Chromatography 23 (2010) 3, 173–179
- Validation data in most cases not determined correctly
- Reported validation data often intend to prove “how good the lab is”, not what variability has to be expected during routine use of the proposed method
- Proposed method’s capability to control tight specification limits often not supported by reported validation data
- To support further acceptance and application of TLC/HPTLC as a real quantitative analytical technique, more stringent quality standards have to be applied - by authors *and* journals

References, Disclaimer & Thanks

- The concepts, conclusions, assessments and statements presented have been developed over a long time with the input and help of various colleagues.
- Without neglecting all the others that contributed, I want to express my special thanks to
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  - Zoltán Végh
  - Marco Zeller
  - Harald Jehle
  - Werner Funk