# Method validation and verification

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# Method Validation and verification

- *What is it?*
- When is it required?
- Why is it necessary?
- What are required?
- How much is adequate?
- How should it be done?
- Any questions on the questions?

# Method Validation – what is it

**Definition** –

- validation is the confirmation by examination and the provision of objective evidence that the particular requirements for a specific intended use are fulfilled *(ISO/IEC 17025:2005 cl. 5.4.5.1)*
- verification, where the specified requirements are adequate for an intended use (ISO/IEC Guide 99:2007)

# Method Verification – what it is

- Verification – provision of objective evidence that a given item fulfils specified requirements (ISO/IEC Guide 99:2007)

# Selection of methods (ISO/IEC 17025:2005 cl. 5.4.2)

- Methods published in international, regional or national standard shall preferably be used.
- Laboratory developed methods or methods adopted by the laboratory may also be used if they are appropriate for the intended use and if they are validated (cl. 5.4.2).

# Non-standard methods (ISO/IEC 17025:2005 cl. 5.4.4)

- Verification is applicable only for standard methods which have been validated.
- (Non-standard methods) The method developed shall have been validated appropriate before use (cl. 5.4.4).

# Laboratory-developed methods (ISO/IEC 17025:2005 cl 5.4.3)

- …shall be a planned activity and shall be assigned to qualified personnel equipped with adequate resources.
- Plans shall be updated as development proceeds ….

# Validation of methods (ISO/IEC 17025: 2005 cl. 5.4.5)

***cl. 5.4.5.2***

- The laboratory shall validate

  *non-standard methods,*

  *laboratory designed/developed methods,*

  *standard methods used outside their intended scope, and*

  *amplifications of standard methods*

  to confirm that the methods are fit for the intended use.

# Laboratory internal validation and verification

| Existing information | Laboratory requirement |
|---|---|
| Fully validated standard methods (have been studied in a collaborative trail) | Verification (Secondary validation) |
| Standard methods – amplifications and modifications e.g. new instrument | Validation |
| Standard methods – outside their intended scope | Validation |
| Laboratory – developed and non-standard methods | Validation (Primary validation) |

# Validation of methods (ISO/IEC 17025: 2005 cl. 5.4.5)

***cl. 5.4.5.2***

- The validation shall be as extensive as is necessary to meet the need of the given application or field of application.

# Validation of methods
# (ISO/IEC 17025: 2005 cl. 5.4.5)

***cl. 5.4.5.2***

- The laboratory shall record the results obtained, the procedure used for the validation, and a statement as to whether the method is fit for the intended use.

# Method Validation – what are required

Validation of methods (cl. 5.4.5.2 Note 1)

- May include procedure for sampling, handling and transportation

# Method Validation – what are required

Validation of methods (cl. 5.4.5.2 Note 2)

- techniques for method performance determination include
  - Calibration using reference standards and Reference Materials
  - Comparison of results achieved with other methods
  - Interlaboratory comparisons
  - Systematic assessment of the factors influencing the result
  - Assessment of uncertainty of results based on scientific understanding of the theoretical principles of the method and practical experience

# Method Validation – what are required

## Validation of methods (cl. 5.4.5.2 Note 3)

- When changes are made in the validated non-standard methods, the influence of such changes should be documented and, if appropriate, a new validation should be carried out.

# Method Validation – what are required

Validation of methods (cl. 5.4.5.3 )

Examples:

- Uncertainty of results
- Detection limit
- Selectivity
- Linearity
- Repeatability
- Reproducibility
- Robustness/cross-sensitivity

# Method Validation – what are required

Validation of methods (cl. 5.4.5.3 Note 1 )

- Validation includes
  - Specification of the requirements
  - Determination of characteristics of method
  - Check requirement fulfilled by method
  - Statement on validity

## Method Validation – what are required

Validation of methods (cl. 5.4.5.3 Note 2)

- Regular review as method development proceeds to verify customer needs fulfilled
- Modifications to development plan due to change in requirements should be approved and authorised

## Method Validation – what are required

Validation of methods (cl. 5.4.5.3 Note 3)

- Validation is a balance between costs, risks and technical possibilities

## Method Validation – what are required

Validation of methods

(5.4 H HOKLAS Policy (c))

- Requirements differ significantly from one technical discipline to another
- Should commensurate with intended use
- Confirm fit for intended use

## Method Validation – what are required

Validation of methods

(5.4 H HOKLAS Policy (d))

- HOKLAS classifies test methods into 3
  - standard method
  - standard method with modifications
  - in-house method

## Method Validation – what are required

Validation of methods

(5.4 H HOKLAS Policy (d))

- standard method
  - –Conforms exactly to the standard

# Method Validation – what are required

Validation of methods

(5.4 H HOKLAS Policy (d))

- standard method with modifications
  - Differs from standard but deviations unlikely to affect test results
  - Need supporting evidence, i.e. validation required
  - Modifications stated in scope of accreditation and test reports

## Method Validation – what are required

Validation of methods

(5.4 H HOKLAS Policy (d))

- In-house method
  - Methods other than the above two
  - Validation required
  - Cannot make reference to other standard method, i.e. cannot claim "based on" a standard method

## Method Validation – what are required

# Validation of methods

# (5.4 H HOKLAS Policy (f))

- Non-standard methods shall be fully documented and validated

# Method Validation – what are required

## Validation of methods

## (5.4 H HOKLAS Policy (g))

- Shall have policy and procedure for design, development and subsequent validation of
  - in-house methods,
  - laboratory designed/developed methods,
  - standard methods used outside their intended scope
  - amplifications and modifications of standard methods

## Method Validation – what are required

# HOKLAS Supplementary Criteria No. 20 *"Chemical Testing", "Chinese Medicine", "Construction Materials", "Food", Toys and Children's Products" – Chemical Testing*

## Section 5.3 Validation of methods

## Method Validation – what are required

# **HOKLAS SC No. 20** *Section 5.3*

- Concentration range
- Sample matrices
- “more advanced” techniques may be a deviation
- Confirmation of identity

## Method Validation – what are required

# **HOKLAS SC No. 20** *Section 5.3*

- LoD
- Precision and trueness
- Applicable concentration range
- Applicable sample matrices

## Method Validation – what are required

**HOKLAS SC No. 20** *Section 5.3*

- Method bias assessed by appropriate matrix CRMs
  - Levels of analytes
  - Matrix matches intended sample matrix
  - Uncertainty of assigned values suitable

## Method Validation – what are required

# **HOKLAS SC No. 20** *Section 5.3*

- Method performance characteristics review, and revised, regularly

## Method Validation – what are required

**HOKLAS SC No. 20** *Section 5.3*

- For food analysis, the method validation required depends very much on the analytes of interest and the matrices. Common food matrices include those rich in protein, carbohydrate, oil, dietary fibre, liquid, etc.

## Method Validation – what are required

# **HOKLAS SC No. 20** *Section 5.3*

- If a method is to be accredited for general food, satisfactory validation data shall be obtained for at least five different food matrices with at least three kinds of food for each food matrix.

## Method Validation – what are required

**HOKLAS SC No. 20** *Section 5.3*

- The range of matrices shall also in line with those listed in relevant regulations. Due consideration shall also be taken for the food matrices with potential interferences, e.g. high chloride effect on the ICP-MS determination.

## Method Validation – what are required

# **HOKLAS SC No. 20** *Section 5.5*

- Estimation of uncertainty of measurement

## Method Validation – how much is adequate

Depends on

- the criticality of the measurement
- The scope of the method

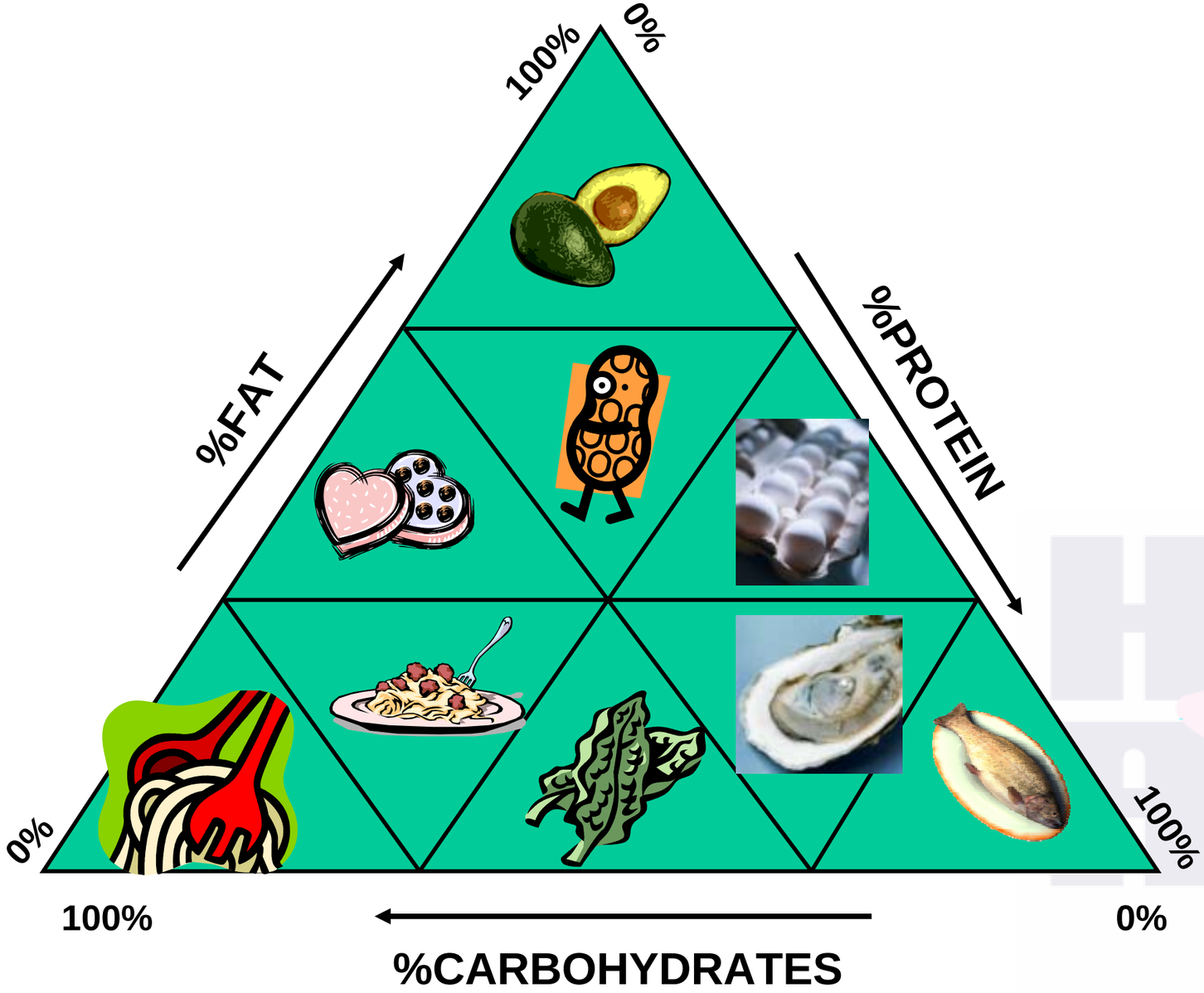
# Method Validation – how much is adequate

- How far can the light shine?
- How far can the validation data on some matrices be extended to other matrices?
- Need professional judgment based on technical knowledge of the limitations of the methodology used.

## Method Validation – how much is adequate

- Example: General foodstuff
- Can never validate using all possible foods
- Classify food according to matrix from an analytical chemistry point of view
- Main components of food – fat, protein, carbohydrate, (fibre, water)
- To obtain reliable data, 3 food types for each matrix
- Performance at specification limits should be available

# Method Validation – how much is adequate




J. AOAC, 83 (2), 413 (2000)
"The Referee", AOAC Int'l, July 1993

# Validation of methods – Accuracy and trueness

- Definition (ISO3534-1)

  bias – the difference between the expectation of the test results and an accept reference value

# Validation of methods – Accuracy and trueness

- Definition (ISO5725-1)

  laboratory bias - the difference between the expectation of test results from a particular laboratory and an accepted reference value.

  method bias – the difference between the expectation of test results obtained from all laboratories using that method and an accepted reference value.

# Validation of methods – Accuracy and trueness

- Assessment of trueness (ISO Guide 33)

  The trueness of a measurement process is checked by comparing the average $\bar{x}$ with the certified value, $\mu$, of a Certified Reference Material (CRM)

# Validation of methods – Accuracy and trueness

- Two factors contributing to the difference between the certified value and the measurement results
  1. The uncertainty of the certified value;
  2. The uncertainty of the results of the measurement process

# Validation of methods – Accuracy and trueness

$$-a_2-2\sigma_D \leq \bar{x}-\mu \leq a_1+2\,\sigma_D$$

$a_1$ and $a_2$ are adjustment values chosen in advance by the experimenter according to economic and technical limitation

$$\sigma_D{}^2 = \sigma_{Lm}{}^2 + s_w{}^2/n$$

n = number of replicate determinations

$\sigma_{Lm}$ = between labs fluctuation $\approx \sigma_1$ (intermediate precision) or $\sigma_L$ supplied by CRM

# Validation of methods – Accuracy and trueness

For many measurement, $s_w$ is small in comparison with $\sigma_{Lm}$ ; consequently when n>10, $\sigma_D$ can be equate with $\sigma_{Lm}$

$$-a_2-2\sigma_{LM} \leq \underline{x}-\mu \leq a_1+2\ \sigma_{LM}$$

## Method Validation – how much is adequate

- Document the food matrix that have been validated in the test procedure.
- Labs should consider additional validation when they receive a food matrix not included in the initial validation.

# Common deficiencies

- Number of food matrices used not adequate
- Number of food types for each matrix not adequate
- Concentration levels used not adequate
- Reporting limits estimated but not verified
- Food samples chosen not representative of the food matrices

# Common deficiencies

- Confirmation of identity technique not available or adequate
- Validation/verification data analyses not done correctly
- Measurement uncertainty not available/not estimated correctly
- Equipment and/or test procedures not conforming to the test std requirements

# Common deficiencies

- Lack of or inappropriate sampling procedure
- QC plan acceptance limits not appropriate

# Thank you