Determining the Uncertainty of Calibrator Values

AACC EduTrak Symposium

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References - printed

- **ISO/CEN 17511**
- **Guide to the Expression of Uncertainty in Measurement. ISO, Geneva (1995) “GUM”**
- **Eurachem / CITAC Guide QUAM:2000.P1. Quantifying Uncertainty in Analytical Measurement, 2nd Edition (2000)**

References - internet

a. www.A2LA.org: A2LA policies, links to guidance documents, including the UKAS Guide, and the Eurachem/CITAC Guide, at no cost
b. www.measurementuncertainty.org: Eurachem/CITAC Guide
c. www.fasor.com/iso25: general information, links, and discussion of ISO-IEC 17025
d. www.gum.dk: Danish site to coordinate GUM activities

Why Estimate Uncertainty?

- It's there – we should understand it.
- Assures comparability among tests (national, international)
- Required by most countries for accredited labs (***not*** USA)
- Provides an objective quality measure
- Help with method improvement
- Guide for root cause analysis and corrective action

Caveat

- This introduction is to help get started
- Full understanding takes time

- We are all learning
- NMI's are getting a dose of reality

Uncertainty (of measurement)

"A parameter, associated with the result of a measurement, that characterizes the dispersion of the values that could reasonably be attributed to the measurand."

Requirements of ISO 17511

ISO 17511, clause 4.1.5:

The value assigned to a measurement standard at a given level shall be associated with an uncertainty of measurement that shall include inherited consecutive uncertainty contributions from measurement standards and measurement procedures at all higher levels of the calibration hierarchy.

Requirements of ISO 17511

ISO 17511, clause 4.1.7:

The responsibility of the manufacturer for describing the metrological traceability chain shall start at the value of the manufacturer's product calibrator and end at the metrologically highest reference used by the manufacturer. The uncertainty of this reference shall include any further upwards contributions in uncertainty.

Calibration Traceability

Primary Measurement Standard
Definition of SI unit by CGPM

Primary Reference Measurement Procedure

Secondary Calibrator

Manufacturers Selected Measurement Procedure

Manufacturers Working Calibrator

Manufacturers Standard Measurement Procedure

Manufacturers Product Calibrator

Requirements of ISO 17511

ISO 17511, clause 6:

Uncertainty of measurement shall be expressed in appropriate terms for the assigned value of each measurable quantity pertaining to the reference material

Requirements of ISO 17511

ISO 17511, clause 6:

NOTE: The principles given in the Guide to the expression of uncertainty in measurement (GUM 1993) *preferably* should be followed. The assigned value, *y*, and related expanded uncertainty, *U*, (or combined standard uncertainty, $u_c(y)$) pertaining to a quantity characterizing the reference material should be stated. The minimum information should be:

(numerical value of *y* ± numerical value of *U*) unit

Where $U = u_c(y) \times k$, with the coverage factor $k=2$, giving a level of confidence of approximately 95%

Value Assignment Practices

At least three general approaches:

- Weighed in nominal values
- Adjusted nominal values
- Value assignment by use of analytical testing
  - vs. certified materials (primary standards)
  - vs. value assigned working calibrators
  - vs. reference method
- Other?

Value Assignment Practices

Question:

Would repetitive measurements of a calibrator/reference preparation (Guide 35) satisfy 17511 requirements?

Answer: Probably

- If all uncertainty components, including manufacture and intermediate value assignments are incorporated in replicates at the last level testing.
- If the uncertainty is for the mean of replicates, adjusted for the effect of measurement uncertainty
- If the uncertainty estimate includes homogeneity
- If there are adjustments for all biases

Value Assignment Practices

Question:

Would comparison with a reference method satisfy 17511 requirements?

Answer: Maybe

- Reference method would have to be calibrated with traceable reference materials (including MU)
- Comparison would have to have sufficient range and number of samples to fully describe the relationship
- The reference method must be unbiased or have known biases

Requirements of ISO 15189

ISO 15189, section 5.6.2:

The laboratory shall determine the uncertainty of its measurements, where relevant and possible. Uncertainty components which are of importance shall be taken into account.

NOTE: Sources that may contribute to the uncertainty may include…calibrators, reference materials,…

Requirements of ISO/IEC 17025

ISO/IEC 17025, section 5.4.6.2:

Testing laboratories shall have and shall apply procedures for estimating uncertainty of measurement. *… the laboratory shall at least attempt to identify all the components of uncertainty …*

Requirements of ISO/IEC 17025

ISO/IEC 17025, section 5.4.6.3:

When estimating the uncertainty of measurement, all uncertainty components which are of importance in the given situation shall be taken into account using appropriate methods of analysis.

Note 1: Sources of uncertainty include, but are not necessarily limited to, *the reference standards and reference materials used*, methods and equipment used, environmental conditions, properties and condition of the item being tested or calibrated, and the operator.

GUM approach for estimating uncertainty

Note that GUM is a procedure for Measurement Uncertainty. The uncertainty of a calibrator value is not a case of measurement uncertainty (in most cases).

The GUM approach using components, mathematical models, and propagation of uncertainty still can be used.

GUM caution – from USA GUM

"Although this Guide provides a framework for assessing uncertainty, it cannot substitute for critical thinking, intellectual honesty, and professional skill. The evaluation of uncertainty is neither a routine task nor a purely mathematical one; it depends on detailed knowledge of the nature of the measurand and of the measurement. The quality and utility of the uncertainty quoted for the result of the measurement therefore ultimately depend on the understanding, critical analysis, and the integrity of those who contribute to the assignment of the value. " ANSI/NCSL Z540-2-1997

Types of Uncertainty (GUM)

- Type A: based on repeated analyses & statistics
- Type B: all other types of estimates
- Standard uncertainty: standard deviation estimate
- Combined standard uncertainty: combined standard uncertainties
- Expanded uncertainty: combined uncertainty expanded to attain a defined confidence level

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- Coverage Factor: a multiplier used to achieve a defined confidence level for the estimate.

GUM approach for estimating uncertainty

1. Describe exactly what is being measured.

Define the mathematical model that links the measurand to the influence quantities

Include homogeneity, if single-use aliquots are prepared (if large bulk is retained, ignore homogeneity).

Example: Generic Calibrator via gravimetric preparation

Incoming chemical powder traceable to SI (USP)

Prepare 500μg/mL stock solution by:

- 25 mg powder into
- 50 mL Methanol (class A vol. flask)

$$C = m \cdot P/V \ \mu g/mL$$

m=chemical mass *P*=purity of primary chemical

V=methanol volume *C*=Calibrator concentration

Estimating Uncertainty

2. List ***all*** components of variability.

Consider those components that, by varying slightly, can affect the final concentration

If adjustments are made to correct for known bias (systematic error), then the uncertainty of that adjustment must be included. This should take place after the last stage of testing, prior to value assignment. The component would be added with the MU from the last stage.

Example: Generic calibrator

P=purity of primary chemical

99.5% ± .5% (includes u_c(primary rm))

m=chemical mass Tared weighing, *m*=25mg

components = scale repeatability / linearity / resolution

V=methanol volume Class A flask, 50mL

components = repeatability / flask calibration / temp

Estimating Uncertainty

3. Quantify the uncertainty components

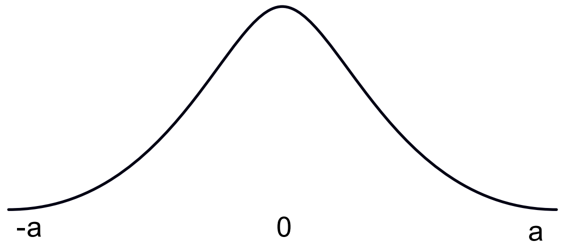
- ✓ Type A or B
- ✓ Size of each component
- ✓ Distribution shape

Express each as a standard deviation

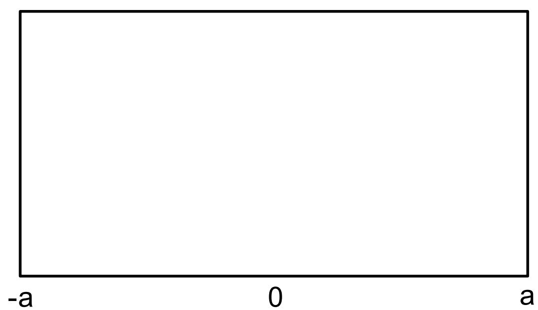
Estimating Uncertainty

Use various sources for the estimates:

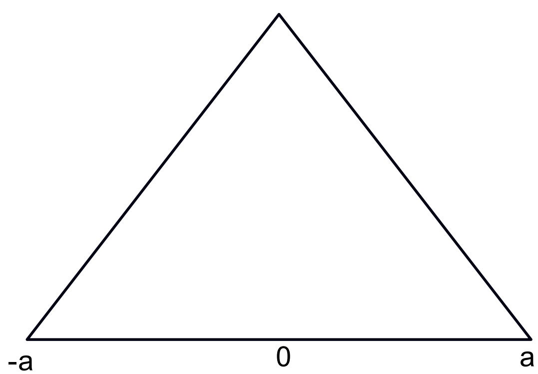
- designed experiments
- method validation studies
- published references
- calibration certificates
- manufacturer/developer specifications
- other sources



Normal
s=.3333a



Rectangular
s=.5774a



Triangular
s=.4082a

Generic Calibrator: purity

P=purity 99.5% ± .5%

from USP

Type B, rectangular

$s_p = .005/\sqrt{3} = 0.29\%$

Generic Calibrator: mass

m=chemical mass (25mg)

- repeatability: from studies (A, normal)
  - s= .0133mg
- linearity: from experience (B, triangular)
  - $s=.022/\sqrt{6} = .0090\text{mg}$
- resolution: from scale, .1 ml (B, rectangular)
  - $s = .05/\sqrt{3} = .0289\text{mg}$

$$s_m = \sqrt{(.0133^2+.0090^2+.0289^2)} = \sqrt{.0011}$$

$$= .0331 \text{ mg} = .13\%$$

Generic Calibrator: volume

V=methanol volume (50mL)

repeatability: from studies (A, normal)

s_r= .0200ml

flask calibration; from certificate (B, triangular)

s =.1ml/√6 = .0408ml

temp: from expansion coefficient – flask calibrated at 20^0C, assume $\pm 4^0$C fluctuation (B, rectangular)

s= (4∗.0182)/√3 = .0420ml

$S_v = \sqrt{(.02^2 + .0408^2 + .0420^2)} = .0619\text{ml} = 0.12\%$

Estimating Uncertainty

4. Combine the components;

- This is “combined uncertainty”.
- GUM specifies use of “Sensitivity Coefficients” (derivatives of equation)
- For simple equations, use RSS method and percentage sd’s.
- For complex equations, may use Monte Carlo simulation (10,000 replicates)

Generic Calibrator

- **Combined Uncertainty:**

  Sum of Squares of components:

  $(.0029)^2 + (.0013)^2 + (.0012)^2 = \mathbf{.00001154}$

  <u>Combined Uncertainty</u> $= \sqrt{(.00001154)} = \mathbf{.34\%}$

Estimating Uncertainty

5. Expand the combined uncertainty;
   This is "expanded uncertainty".
   (This is not required for calibrators)

Use coverage factor k:

- ✓ $k = 2$ or 3 (95% or 99% confidence)
- ✓ k = percentile of t distribution (for less than 20 observations)

Generic calibrator

Expanded Uncertainty:

Use coverage factor *k* = 2, for 95% confidence

Expanded uncertainty = Cx(2x0.34%) =

500µg/mLx.0068 = 3.4µg/mL

Reporting Uncertainty

<u>Report must include:</u>

- Calibrator value
- Combined uncertainty
  - same units as calibrator value
  - same significant digits as calibrator value
- If expanded uncertainty is reported
  - report coverage percentage (e.g., 95%)
  - report expansion factor used (*k*)

Measuring Concentration

- Calculate MU from testing process
- Can use shortcuts, if testing process is well known at the stated level
  - Quality Control (reproducibility)
  - Monte Carlo simulation
- Use MU for the mean of tests (SE), not MU for single tests ($SE = SD/\sqrt{n}$)

Estimates from Control Samples

- Consider extent of method that is replicated by QC samples (e.g., prep)
- Add any components that are not included
- Use reproducibility SD to replace some or all components
- Use 20-50 samples

Uncertainty for transfer protocol

- Take combined standard uncertainty of primary calibrator
- Take uncertainty of measured mean
- Combine using RSS for uncertainty to use in subsequent steps (working calibrator)

Uncertainty of Working Calibrator

- Take combined standard uncertainty from previous step
- Estimate uncertainty as for primary calibrator (or stock solution)
  - All components in manufacturing process
  - Produce combined uncertainty SD
- Repeat for all pairs of steps
- May need to estimate homogeneity

Conclusion

- GUM procedure is *preferred* to estimate uncertainty of calibrators and RM
- Use reference material reproducibility to estimate uncertainty of measured means (divide by $\sqrt{n}$)
- Repeat process for all pairs of steps
- Include uncertainty of bias corrections

Thank you!

Contact Information

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