Sections in This Document

1. Purpose

This procedure describes the process for the estimation of measurement uncertainty (MU).

2. Scope

These procedures apply to regulatory testing carried out by the Office of Regulatory Science (ORS) laboratories.

3. Responsibility

ORS laboratories are responsible for the collection of data and the determination of the estimated uncertainty for analytical methods in use.

4. Background

Every measurement or test has an error of measurement. If repeated, a test or measurement often gives a different result, even though it usually is very similar to the original result. Therefore, a test or measurement gives only an approximation of the true value of the quantity to be measured. A measurement or test is only complete if it includes the measurement uncertainty of the test. This can be thought of as a quantitative indication of the quality of the result.

Defining the measurement uncertainty is the same as determining accuracy of results.

5. References

A. National Institute of Standards and Technology (NIST). Technical Note 1297, Guide for Evaluating and Expressing the Uncertainty of NIST Measurement Results.

B. EURACHEM. CITAC Guide, Quantifying Uncertainty in Analytical Measurements.

C. PL-3 Policy on Uncertainty Measurement, Perry Johnson Laboratory Accreditation

D. ISO/IEC 17025:2017, General Requirements for the Competence of Testing and Calibration Laboratories. Section 7.6

E. AOAC International Guidelines for Laboratories Performing Microbiological and Chemical Analysis of Food, Dietary Supplements, and Pharmaceuticals – An Aid to Interpretation of ISO/IEC 17025:2017; August 2018.

6. Procedure

6.1. Categories of Tests

A. Each laboratory is required to identify and record the measurement uncertainty category (listed below) for each method on the scope of accreditation. Additionally, each laboratory is required to list all the components of uncertainty and estimate the uncertainty for all quantitative methods or technologies on their scope of accreditation.

B. The three different categories of tests as well as the extent of uncertainty estimation are:

1. Qualitative tests and tests reported on a categorical or nominal scale. These do not need a measurement of uncertainty; however where possible the laboratory should identify possible areas for error in order to control these factors.
2. Well recognized test methods that specify limits to the values of the major sources of uncertainty of measurement and specify the form of presentation of calculated results. In such cases, the laboratory is considered to have satisfied the uncertainty requirements by following the method and reporting instructions; however when verifying the method the laboratory should confirm their MU compares with the established MU for the method.
3. If a new quantitative test method does not specify or define a measurement uncertainty, the laboratory must determine the method's measurement uncertainty. Uncertainty can be estimated using laboratory control samples or the root sum square (RSS) method. After the application of the required calculation or uncertainty estimation technique provided in this procedure, other calculations and techniques can be used as supporting evidence.

6.2. Estimating Uncertainty

The steps for estimating uncertainty are discussed below. Measurement uncertainty does not usually include uncertainty due to sampling or bias.

A. Steps using relative standard deviation of laboratory standards/control samples run through all method steps.

1. Perform spiked determinations at different concentrations including tolerance limit.
2. Calculate concentration and percent recovery.
3. Calculate the standard deviation (SD) and relative standard deviation (RSD) on results where the process is in statistical control (no outliers or out of control results).
4. Calculate the measurement uncertainty at the 95% confidence level as follows:

$$U = k \times RSD$$

where:

U = uncertainty

k = coverage factor (for 95% and 50 points, use 2; for less than 50 points, use the appropriate t statistic for 95%)

5. To calculate the measurement uncertainty interval for a measured value, calculate as follows:

   Interval = U X c

   where:

   c = concentration

B. Steps using the root sum square (RSS) method.

1. Clearly define what is being measured.
2. Review the method and identify every possible source of uncertainty. Sources include, but are not limited to:
   a. Reference standards and reference materials
   b. Methods and equipment used
   c. Environmental conditions
   d. Properties and condition of the unit under test
   e. Operator
3. Review the sources and determine whether or not the components are included when running laboratory control samples.
4. Quantify all the components. Estimates can come from a variety of sources and do not need to be overly rigorous. Possible sources of quantitative estimates include:
   a. method validation studies,
   b. information from published methods or textbooks,
   c. calibration certification,
   d. manufacturer's specifications, and
   e. experience.
5. Consider the components. Assume that the components are independent. Every source may not have to be evaluated if they are deemed insignificant. Components that are less than a fifth of the largest component can be eliminated.
6. Combine the components. The usual method is to square all independent components, add them, and then take the square root of the sum. This is called the root sum square method and gives the combined standard uncertainty.

7. Expand the combined standard uncertainty. Multiply the combined uncertainty by a coverage factor based on the level of confidence needed. In most cases a confidence level of 95% is sufficient, so k =2. If 99% confidence is required, then k =3. When estimates are based on limited data, k may vary according to the student t distribution. For example, if the estimate is based on analyses of 20 laboratory control samples, the coverage factor of k = 2.1 for 95% confidence.

C. Measurement Uncertainty budgets are periodically reviewed and updated to reflect changes in the laboratory, equipment, procedures, or personnel.

6.3. Reporting Uncertainty Estimates

A. The extent of reporting of the estimates of uncertainty depends on the needs of the client, the specifications for the test, and the intended use of the result. Measurement uncertainty is not a major contributor in basing regulatory actions since the FDA will err on the side of public safety when making those decisions.

B. MU estimate is not reported unless specifically requested by the customer. Nonetheless, the estimate is to be calculated, along with the means of making the estimate. Records are sufficient to allow replication of the calculation.

C. If reported, the uncertainty is reported to the same number of significant figures as the result. In most cases, two significant figures suffice. The estimate should also state the level of confidence associated with the coverage factor. Finally, the uncertainty is reported in the same units as the result.

D. The bias is reported in addition to the uncertainty when the method has a known bias and the bias adjustment was not performed.

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7. Glossary/Definitions

A. Coverage factor – The coverage factor is the number that is multiplied by the standard uncertainty to produce an uncertainty estimate that will contain a large fraction of all values that might be obtained on a test. The coverage factor is commonly noted as k, and k=2 is used for 95% coverage, and k=3 for 99% coverage.

B. Expanded uncertainty – The expanded uncertainty is the combined standard uncertainty (or standard uncertainty, if there is only one component), multiplied by the coverage factor.

C. Measurement uncertainty – The measurement of uncertainty is the parameter associated with the result of a measurement that characterized the dispersion of the values that could be reasonably attributed to the measurand.

D. Measurand - The quantity being measured is the measurand (i.e. the concentration of an analyte).

E. Repeatability conditions – Identical samples prepared at the same time, by the same analyst, under identical conditions, run on the same instruments are repeatability conditions.

F. Reproducibility conditions – Reproducibility conditions are identical samples analyzed under different conditions, including any of the following: different times, different equipment, different analysts, or different laboratories.

G. Standard uncertainty – The standard deviation for uncertainty is either for the test or for a component of the test. It can be expressed in the units of the measurement, or a percentage, but all components are expressed in the same terms before they can be combined.

8. Records

A. Measurement uncertainty determinations, calculations and values for laboratory methods in use

9. Supporting Documents

B. ORA Laboratory Manual, Volume II, ORA-LAB.5.4.5 Methods, Method Verification and Validation

10. Document History

Revision #	Status* (D, I, R)	Date	Author Name and Title	Approving Official Name and Title
1.2	R	03/23/2005	LMEB	LMEB

02	R	05/15/2019	LMEB	LMEB

* - D: Draft, I: Initial, R: Revision

11. Change History

Revision #	Change
02	Revisions made to align this procedure with new ISO/IEC 17025 and AOAC requirements. Revision to formatting and policy clarifications were also made.

12. Attachments

None