Guideline

Subject: Uncertainty of Measurement
Approval Date: November 2004, November 2009
Review Date: November 2013
Review By: PPAC
Number: 2/2004

Introduction

This Guideline has been developed to provide some preliminary advice to Fellows on how to approach Uncertainty of Measurement. In addition, NPAAC have an accreditation requirement concerning estimation of measurement uncertainty.

It is a requirement of ISO 15189 that laboratories “shall determine the uncertainty of measurement of results, where relevant and possible”………. (5.6.2)

It should be noted that there is nothing new in the concept of uncertainty of measurement. However, it is likely that many different terms have been used to describe it.

ISO15189 (3.17): The uncertainty of measurement is a parameter associated with the result of a measurement that characterises the dispersion of the values that could be reasonably attributed to the measurement.

For example: If the uncertainty of measurement for a test is 10%, and the result is 100, then the true result probably lies between 90 and 110.

This is a useful parameter for indicating how reliable the result is, or to indicate whether any change in test results over time might be clinically significant, or just due to the imprecision of the analytical method used to produce the result.

Common questions pathologists are asked are:
- Is this result significantly abnormal? (usually when it is close to the reference range)
- Is the most recent result for a test significantly different to the previous result? Does the change represent a significant change in the patient’s condition?

Generally these questions are answered by trying to remember the “uncertainty of measurement” for that specific test.

ISO 15189 asks for the degree of uncertainty to be both measured and documented.

IANZ (and NATA) have indicated that as a first step, the measurement of uncertainty should be determined for tests that report the result as a number. This requires an understanding of the imprecision of the analytical process used for those tests.

Imprecision and the Calculation of Uncertainty of Measurement:

The calculation is based on the imprecision (CV%) of the test (see next page).

Ideally, this should be the imprecision of the test as it is performed in the specific laboratory.
This means that at least 30 sets of internal QC data should be considered, obtained over a reasonable period of time (not all in one batch), from which the CV% for that test can be calculated.

The possible sources of the imprecision data for each method include the following:

- **For a new test:**
  Use the CV% data obtained when the test was first established in the specific laboratory. However, the imprecision should be re-estimated as soon as 30 data points are obtained as above, to better reflect the routine performance of the test.

- **For a rare or expensive test:**
  The manufacturer's data on CV% may be used if it is impractical to gather sufficient data to calculate the imprecision of the test as performed in a specific setting.

- **The use of EQA Programme imprecision data,** obtained from a group of laboratories using the same method as the laboratory wishing to use it, may also be acceptable under special circumstances, but this should generally be used only until there is sufficient in-house data to illustrate the performance of the test in the laboratory wishing to use it.

- **For tests where it is simply not possible to determine the imprecision of a test by collecting data,** a best estimate based on general experience with the test may suffice.

- **There are some laboratory results where it is not relevant to determine uncertainty of measurement.**

The imprecision of tests will change over time. Such changes should be monitored to see if they justify a recalculation of the uncertainty of measurement. Using common sense, and rounding off the uncertainty of measurement figures for a particular test, will reduce the need to keep recalculating it on a regular basis (e.g. quoting 7.63% would require constant review, whereas <10% may be near enough for clinical use, and give sufficient leeway to remain unaltered over a longer period).

The recommended calculation of uncertainty of measurement, known as the coverage factor, is:

\[ CV \% \times 2 \]

The uncertainty of measurement may have to be calculated at more than one level if there is a marked change in imprecision across a range of results that could alter clinical interpretation.

There is little point in calculating uncertainty of measurement at concentrations that are well away from clinical decision making limits.

The uncertainty of measurement can be expressed as a %, or as a specific number to relate to a specific test value.

E.g. Albumin: 3 g/L at a concentration of 40 g/L
Potassium: 4%

In determining this process, a number of factors need to be considered:

- **It has to be a simple calculation that is fit for purpose.** Many documents on standards and statistics describe complex calculations, usually based on the numerous sources of possible error in the analytical process. Whereas this is required for the preparation of standards, controls and components of reagents, such meticulous assessment of the
uncertainty or measurement is both impractical and unnecessary for medical laboratory tests. Only those factors having a significant influence on the final result need to be considered.

- The overall imprecision of a test includes most of the significant factors that could practically be assessed.
- Other factors such as non-specificity, method interferences, instability of specimens, sample selection from specimens, etc, are variable and generally not measurable for a particular specimen. They should be noted as potential sources of errors either in the laboratory’s handbook, or on the report form if the potential error could have significant clinical consequences.
  
  E.g. Potential interference in the troponin assay from heparin in the collection tube should perhaps be noted on the report form, whereas ketone interference with the creatinine assay would be more appropriate to mention in a laboratory handbook.

- Factors such as biological variation and age related values are best considered and included as part of the reference range/interval.

**Clinical Application:**

For a single test result:
Uncertainty of Measurement is used to indicate the confidence we have that the reported figure is correct.

If the uncertainty of measurement is calculated as $\text{CV}\% \times 2$:
There is a 95% chance that the true result lies within a range covered by result number $\pm$ the uncertainty of measurement.

For two results to be significantly different:
Uncertainty of measurement is used to indicate that the difference between two results is unlikely to be due to method imprecision alone, and is either due to normal biological variation or a change in the patient’s physiological or pathological condition.

If the uncertainty of measurement is to differentiate two figures with 95% confidence, then the CV% would be multiplied by 2.77. This is generally not practical for every day use. By multiplying the CV% by 2 (use the same figure as for a single test) then the confidence is reduced to 84%. By multiplying the CV% by 3, there can be a 97% confidence level that the figures are different.

It is recommended that CV% be multiplied by 2 in this situation. The term “significantly different” should not be used, as this term has a specific statistical meaning (95%), and so terms such as “highly likely to be different” are preferred. By aiming for 97% confidence ($3 \times \text{CV}\%$), then “almost certainly different” may be used. Such a high level of confidence is not required for most clinical uses of laboratory tests, and there are advantages in having only one (set of) measurement(s) of uncertainty for each test.

**ISO 15189**

The requirements contained in ISO15189 should be studied carefully.

ISO 15189: 5.6.2: *The laboratory shall determine the uncertainty of results, where relevant and possible. Uncertainty components which are of importance will be taken into account. Sources that contribute to uncertainty may include sampling, sample preparation, sample*
portion selection, calibrators, reference materials, input quantities, equipment used, environmental conditions, condition of the sample and changes of operator.

In the case where results are reported as either positive or negative, it is expected that what would be reported is the certainty the result is not a false positive or a false negative.

If it is impossible to gather data to determine uncertainty of measurement, then a reasonable estimate is acceptable, but this needs to be done only if uncertainty of measurement has clinical relevance.

Uncertainty of measurement is irrelevant in disciplines such as Anatomical Pathology for descriptive results in histology reports. However, the “uncertainty of measurement” (unreliability) of the cervical screening test would be beneficial in educating the public that it is a screening test and not a diagnostic test, with the associated increased likelihood of errors.

ISO 15189: 5.8.3 (k) ……… and where applicable, information on detection limit and uncertainty of measurement should be provided upon request.

This second requirement makes it clear that uncertainty of measurement does not have to be included on every report. It simply needs to be available should anyone enquire about the reliability of the result supplied to them.

Calculating Uncertainty of Measurement

The CV% x 2 is a good calculation of uncertainty of measurement, but there are some other factors that may need to be noted in the laboratory’s handbook and/or as a comment on the report form.

Clause 5.6.2 (see above) lists a number of factors that should be considered when calculating the uncertainty of measurement:

- Sampling – for most tests this has a minor effect, and so need not be included. If it is critical (unstable specimen), then a note should be made on the report or in the Handbook indicating the effect of unsuitable collection and delivery of the sample. Sampling is not a constant variable that affects most measurements, and so is difficult to estimate. If it is a constant factor, then this should be allowed for in the reference range/interval. If timing of sampling is important, then notes on specimen collection and the reference range/interval should indicate this.
- Sample preparation – included in the CV%.
- Sample portion selection – not relevant for most tests that report a figure. Variations are usually accounted for by sub-sampling (e.g. sub-sample of a 24 hr urine collection rather than a spot sample), or by expressing the result as a ratio (e.g. a urine concentration as a ratio to urine creatinine output).
- Calibrators – variation in calibration may be included as part of the CV% and may also be reflected in the reference range/interval.
- Equipment used – effect included in the CV%.
- Environmental conditions – effect included in the CV%.
- Condition of the sample – may affect a proportion of specimens, but collection procedures should deal with those tests/samples where this is a potential issue.
- Changes of operator – should be a minor effect, but will be included in the CV% over a period of time.
From the above, it can be seen that CV% derived from internal QC data over a period of time will cover most of the significant factors that will add to the uncertainty of measurement.

**Preparation for ISO 15189**

The College recommends the following:

1. Make a list of all of the tests where the result is reported as a number.
2. From this, make a sub-list of every test which is already in the in-house CV% data.
3. Multiply the CV% by 2 and record this figure as the uncertainty of measurement against each of these tests. This should cover the majority of tests that a particular laboratory reports as a number.
4. Determine the CV% for the remaining tests, either by:
   - reviewing internal QC data (30 sets) and determining the CV%
   - obtaining CV% for method employed from an External QA Programme
   - obtaining CV% for method employed from the manufacturer (reagent kit insert)
5. Check if it is either impractical or irrelevant to determine the uncertainty of measurement for the remaining tests, and document your reasons if these are not obvious.
6. Finally, document the policy for determining the uncertainty of measurement, the calculations used, and the sources of data used for these calculations.

**Related Documents:**

- Guidance on the number of significant figures may be found in Hawkins, R.C. and Johnson, R.N. “The significance of significant figures”, Clinical Chemistry (1990) 36(5):824.