Purpose

This procedure describes the process for method verification, method validation, and estimation of uncertainty of measurement to ensure that the standard test methods used within the laboratory are validated and are producing correct results.

Application

This procedure applies to all the sections of PPHRL to ensure the compliance with clause 5.5 of international standard ISO 15189:2012.

Procedure

Verification of examination procedures:

Method verification is applied on validated test method. Following procedure is followed for the method verification.

1. **Proficiency Testing (PT):**

   Lab will participate in PT program for the scope of its test, for which PT services are available. The lab will participate at least one test/year. The method used in the lab will be verified after getting the satisfied result by the PT Provider.

2. **Inter Lab Comparison (ILC):**

   Lab is performing Inter Lab Comparison the laboratory which are already accredited over international standards ISO 15189:2012. No modification is made in the standard test methods and results are being compared. If the laboratory results are in acceptable range then STM is verified. Activity is recorded.

   In case of deviation of the results.

   - Environmental conditions are verified where test was performed.
   - In-house calibration of equipment used in the testing activities are recalibrated.
   - Material used for the test was replaced with reference material.
   - Technical staff is also replace
   - Standard Test Method instructions are followed and verified.
   - The testing activity is performed under the supervision of senior technical staff.
   - All the activity is recorded and the lab again under ILC.

3. **Retesting:**

Resting is also performed on quarterly basis. The samples with known results is being provided to concerned sections with blind coding and testing activity is performed using standard test method.

Validation of examination procedures

STMs used for performance of examination over primary samples are standard test method which are validated and are being used worldwide. In case any test method included in the scope of the laboratory, then the method will be validated and procedure for validation will be developed and implemented in the PPHRL.

Measurement uncertainty

Terms & Definitions

Uncertainty

Uncertainty is deviation from the true value and can only be expressed in the form of a range, e.g. 7.5 $\pm$ 0.03

Here 7.5 is the calculated result and $\pm$ 0.03 uncertainty of the system. These results have a range between 7.47 to 7.53. Greater the range lower will be reliability of results where as less range proves the quality of results.

Published Definition

Uncertainty is a quantification of the doubt about the measurement result.

Error Vs Uncertainty

Error expresses a problem and can only be expressed (in statistical terms) as either positive or negative value, while uncertainty cannot be expressed in the form of a single value. Published Definition of Error

Error is the difference between the measured value and the 'true value' of the thing being measured.

Sources of Uncertainty

Test Performer(s)

It is the uncertainty of the person performing the test, but as a person's error cannot be calculated in the form of numerical values that be used in advanced combined

estimations, we determine Repeatability and Reproducibility through the test results they achieve.

Uncertainty of Repeatability

It is the deviation found in test results when a test is repeatedly performed under the same condition and by the same person.

Uncertainty of Reproducibility

It is the deviation found in test results when a test is repeatedly performed on the same type of sample under different conditions (in our case we suggest changing the test performer only and try to sustain the rest of the uncertainty sources' condition as much as possible).

Method used

It is uncertainty expressed in the test method when some assumptions have to be made due to limitation of resources.

Equipment and Accessories

The equipment, apparatus and accessories being used have uncertainty of their own. In most cases it can be found in their manuals, on their label or their certificates. Experienced personnel who are capable of performing calibration of equipment are also able to find its uncertainty, if it's not given in any other source.

Material, Sample and Chemical

Any material, chemical and/or sample being used has its own uncertainty, which is usually provided with its certificate, literature or container. If it's not provided, personnel can calculate it by comparing it with a reference standard where uncertainty is already stated.

Procedure for Calculation of Uncertainty

Uncertainty in the result may occur due to the certain factors, most often Analyst, Equipment, Chemical / Solution, Environment, etc. all factors constitute the function of combined uncertainty. Factors of uncertainty are independent variants, where the uncertainty is dependent of these variants. This procedure will cover with a function with following factors.

X_1 = Analyst

X_2 = Equipment

X_3 = Chemical / Solution

X_4 = Environment

C.U = $f\ (X_1, X_2, X_3, X_4)$

X_1 = Analyst Performance (Reading)

Standard uncertainty of the analyst will be calculated by the standard deviation in the readings taken by the two analysts. This uncertainty will be combination of repeatability and reproducibility. The analyst who will have large standard deviation will be incorporated. This standard uncertainty is measured at 68% confidence level.

Following formula will be used for the calculation of deviation.

$S.D = \sqrt{\ \sum\ (X_i - x)^2 / n-1}$

X_2 = Equipment

The uncertainty of this factor is taken from the calibration certificate as this uncertainty is measured at 95% confidence level. In case of automated equipment this factor will be kept constant as calibration error is adjusted automatically when standard controls are executed before start of examination.

X_3 = Chemicals / Solution

Influencing factors of chemicals or solution is mentioned in the manual or printing material of chemical / solution. Normally this factor will be given at 95% of confidence. If CI is given at 99% or 68% convergence factor will be incorporated in measurement for homogeneous unit

X_4 = Environment

The variation of results against different temperature levels is calculated. The value of uncertainty is calculated by the derivation of results against temperature levels.

Convergence factor:

Normally all calculated values of uncertainty are at K_1 level. i.e. at 68% CI. All factors uncertain values are converted at K_2 level i.e 95% CI this is done by dividing the uncertainty of factor by square root of 2. This value is call the standard uncertainty of factor.

Sensitivity Factor

Sensitivity factor is a constant, which describes the compassion of uncertainty between dependent and independent variants of combined uncertainty function. Its is denoted by "C" and will be calculated by derivation of dependant w.r.to independent. Multiply the sensitivity factor with uncertainty calculated at 95% CI level. It will be total uncertainty of factor.

Uncertainty Budget:

The calculation of total uncertainty of all factors is called the budget of uncertainty, for each of the test separate uncertainty budget will be calculated. Quality Form code: QSP/QF/L4/027 will be used to prepare uncertainty budget. Taking supposed value the uncertainty budget is prepared as below:

Input qty	**Estimate** X_i	**Standard uncertainty** $U(x_i)$ @ 68% CL	**Sensitivity coefficient** C_i	**Contribution to combined uncert.** $U(y_i) = U(x_i) * C_i$
Analyst	10.06	0.124	1	0.124
Equipment	10.00	0.0102	1	0.0102
Chemical	10.00	0.0011	0.004	0.0000044
Environment	23	2	0	0

Column 1: Input quantity / factor of uncertainty
Column 2: Describe estimated value of factor
Column 3: standard uncertainty by converging uncertain value at 68% CI
Column 4: Sensitivity Factor
Column 5: total uncertain value of factor

Combined Uncertainty:
The square root of sum of squares of all factors is called the combined uncertainty. It is the uncertainty of system at 68% of CI.

Combined Uncer. (Uc) = $\sqrt{(U_{(y\,1)})^2 + (U_{(y\,2)})^2 + (U_{(y\,3)})^2 + (U_{(y\,4)})^2}$

= $\sqrt{(0.124)^2 + (0.0102)^2 + (0.0000044)^2 + (0)^2}$

= $\sqrt{0.0153 + 0.00010404 + 0.000000000019 + 0}$

= $\pm$ 0.124 @ 68% C.L

Expanded Uncertainty:
The value of uncertainty of system at 95% CI is called expanded uncertainty. This is calculated by multiplying combined uncertainty with K = 2

Expended Uncertainty (Ue) = Uc x K

Ue = $\pm$ 0.124 x 2

= $\pm$ 0.248 @ 95% C.L

Result:

10.56 $\pm$ 0.248 @ 95% C.L

Documentation of examination procedures:

PPHRL is using standard test method. These methods are validated and are being used worldwide. STMs are written in a language commonly understood by the staff in the laboratory and be available in appropriate locations. A master copy of STMs is available in the lab whereas controlled copies are available on working benches of technical staff according to the scope of their sections.

STMs include, (when applicable) the following:

- purpose of the examination
- principle and method of the procedure used for examinations
- performance characteristics, type of sample (e.g. plasma, serum, urine)
- steps required for patient preparation

- type of container and additives
- required equipment and reagents
- environmental and safety controls
- calibration procedures
- procedural steps
- quality control steps, interferences (e.g. lipaemia, haemolysis, bilirubinemia, drugs) and cross reactions
- principle of procedure for calculating results including, where relevant, the measurement uncertainty of measured quantity values
- biological reference intervals or clinical decision values and reportable interval of examination results
- instructions for determining quantitative results when a result is not within the measurement interval
- Alert/critical values, where appropriate
- laboratory clinical interpretation
- potential sources of variation, references

1. Related Documents

- Budget for uncertainty
- PT/ILC record
- Retesting record