Harmonisation in Analytical Phase

Hassan Bayat; Clinical Laboratorian
What is the problem?

• Many laboratory measurement procedures give different results for the same specimen
Why does it matter?

- Patients may get the wrong treatment

- Many clinical decisions are informed by laboratory results
- Many clinical guidelines use a fixed laboratory test value for treatment decisions

Example - ACG Practice Guideline: Evaluation of Abnormal Liver Chemistries – 2016:

ULN for ALT: 29 to 33 IU/l for males, 19 to 25 IU/l for females; levels above this should be assessed by physicians.

*Am J Gastroenterol* advance online publication, 20 December 2016; doi: 10.1038/ajg.2016.517
Why else does it matter?

- Clinical studies may use a central lab with a single method
  - Guidelines from the study cannot be implemented until all other methods are harmonized to the central lab

- Clinical studies may use different methods
  - Data cannot be aggregated to develop guidelines until the results are harmonized
Importance of consistent results geographically and over time

Patient perspective
If different measurements systems result in different results for the same patient sample

- Physicians and patients will become confused
- Clinical guidelines will become less useful
- Suboptimal treatments and monitoring practices may be implemented
PTH: Variability Between Methods

PTH concentration (pmol/L) in a single patient.

Treatment variation caused by comparing highest and lowest PTH concentrations in 18 patients.

Almond A, Ellis AR, Walker SW
Current parathyroid hormone immunoassays do not adequately meet the needs of patients with chronic kidney disease.
Example: Calcium

- Analytic biases of 0.1 and 0.5 mg/dL: $60 million to $199 million per year

Gallagher MP, Mobley LR, Klee GG, Schryver P. RTI planning report 04-1, the impact of calibration error in medical decision making. NIST. U.S. Department of Commerce. Technology Administration, April 2004.
Why Anconsistent Analytical Results?

Measuring concentration
Laboratory bias
Reagent bias
Instrument bias
Operator bias

Result
Measurement uncertainty

Bias
What to do?

**Harmonization**

- Equivalence of measurement results among different routine measurement procedures over time and space according to defined analytical and clinical performance goals

- AUS IOM report specified the 6 aims for healthcare:
  - Safe
  - Effective
  - Timely
  - Equitable
  - Efficient
  - Patient centered


These aims provide excellent guidance for the goals of a harmonization process.
Terminology

Standard
The International Vocabulary of Metrology (VIM): “realization of the definition of a given quantity, with stated quantity value and associated measurement uncertainty, used as a reference.”

Traceability
VIM: “property of a measurement result whereby the result can be related to a reference (a standard) through a documented unbroken chain of calibrations, each contributing to the measurement uncertainty.”

Commutability
VIM: “property of a reference material, demonstrated by the closeness of agreement between the relation among the measurement results for a stated quantity in this material, obtained according to two given measurement procedures, and the relation obtained among the measurement results for other specified materials,”
Commutability

(A) Commutable RM

(B) Non-Commutable RM
Non-commutable RMAs: Why?

- Manipulating and alteration of sample matrix
- Nonnative forms of measurands
- Exclusion of clinically important forms owing to purification steps
Non-commutable RMs: Why?

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Standardization:

A harmonization process in which the values assigned to hierarchically lower order standards are systematically determined either by a direct comparison to the highest order reference standard available (i.e., the prototype platinum–iridium alloy kilogram at the BIPM), or indirectly, by comparison with an intermediate (lower order) reference standard, such as a platinum–iridium “copy” prototype kilogram maintained at an NMI.
International Laboratory Accreditation Cooperation (ILAC) Policy on Traceability of Measurement Results:

Essential elements of a traceable measurement:

1. Unbroken chain of comparisons
2. Calculated uncertainty of the final result
3. Fully documented procedures and outputs
4. Evidence of the competence of the laboratories conducting each process
5. The highest order elements of the chain of comparisons should, where available, be primary standards linked to realization of SI units
6. Defined calibration intervals
Traceability

International standard
Preferably SI units

Result from definitive method and calibrator

Result from reference method and calibrator

Result from manufacturer method and calibrator

Result from routine method and calibrator

An unbroken chain of comparisons and uncertainty estimations

1. Name/identity of standard
2. System
3. Unit
4. Concentration
5. Combined uncertainty

Uncertainty
ISO 17511-2003. In vitro diagnostic medical devices -- Measurement of quantities in biological samples -- Metrological traceability of values assigned to calibrators and control materials
Cholesterol

Measurand/Si
Serum cholesterol, substance concentration (mmol/L)

Unit Realization
Cholesterol NIST SRM 911c
99.2 ± 0.4% purity

Gravimetry (weighing)

Primary Calibrator:
NIST SRM 911c in ethanol

Primary measurement procedure: ID-GC/MS

Reference Calibrator:
NIST SRM 1951b, Lipids in Frozen Human Serum

Reference measurement procedure: CDC Abell-Kendall procedure

Working calibrators: cholesterol in serum, or serum-like material

End-user method: cholesterol oxidase method on an automated clinical chemistry platform

Routine patient sample

Patient result: “serum cholesterol = xxx mmol/L”
Hb A1c

Combined standard uncertainty ($\mu_C$)

$\mu_C 0.76\%$

Primary calibrator ($\text{HbA}_2, \text{HbA}_{1c}$)

Secondary calibrator

IFCC reference procedure HPLC-GE performed by CIRME

Accredited reference laboratory

Abbott's working calibrator

Abbott's selected procedure

Abbott's standing procedure

Abbott Diagnostics

Abbott's product calibrator

Abbott enzymatic method on Architect c4000 platform

Clinical samples

Results, mmol/mol

Clinical laboratory
JCTLM
Joint Commitee for Tracability in Laboratory Medicine

- Created in 2002 under a Declaration of Cooperation between the CIPM, IFCC & ILAC
- To support implementation of the EU IVD Directive

- ISO 15193 — Requirements for Reference Measurement Procedures
- ISO 15194 — Requirements for Reference Materials
- ISO 15195 — Requirements for Reference Laboratories

<table>
<thead>
<tr>
<th>Category</th>
<th>Reference measurement procedure</th>
<th>Primary (pure-substance) reference material</th>
<th>Secondary (value-assigned) reference material[^a]</th>
<th>Examples</th>
</tr>
</thead>
</table>

[^a]: More than 1 secondary reference material, with potentially different properties, may be available for the same measurand.
ISO 17511:2003 categories for reference systems

<table>
<thead>
<tr>
<th>Category</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Electrolytes, glucose, cortisol</td>
</tr>
<tr>
<td>2</td>
<td>Enzymes</td>
</tr>
<tr>
<td>3</td>
<td>Hemostatic factors</td>
</tr>
<tr>
<td>4</td>
<td>Proteins, tumor markers, HIV</td>
</tr>
<tr>
<td>5</td>
<td>Epstein-Barr virus, varicella zoster virus</td>
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</tbody>
</table>

* More than 1 secondary (assigned) material available for the same measurand.
Calibration traceability agreed by convention

• International **conventional calibrator** (e.g. WHO)
  • Calibrator with a value that is not traceable to SI
  • The assigned value of the calibrator is based on international agreement

• International **conventional reference measurement** procedures
  • Yields values that are not traceable to SI, but the values obtained are agreed as reference values by international agreement
Measurands without RMPs (Cat. 4 & 5)

- Inadequate definition
- Inadequate analytical specificity
- Inadequate attention to the commutability of reference materials
- Lack of a systematic approach for harmonization
Challenges in ISO Categories 4 and 5

• Definition of the Measurand
  • Example: HCG and cTnI have different molecular forms in different clinical conditions.
    ➢ Harmonization may not be possible for some measurands until the clinically important molecular form(s) are clearly identified.

Linda Theinpoint: The profession should define the component(s) intended to be measured more clearly.

• For calcium, it is calcium; for PTH it is the 1–84 form; for vitamin D it is the sum of vitamin D2 and D3; for TSH it is the mixture of glycoforms but being measured equimolar.

Accuracy in clinical chemistry – who will kiss Sleeping Beauty awake?
Challenges in ISO Categories 4 and 5

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• Analytical specificity

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Commutability Challengene

**Category 4:** 1 or more reference materials with a protocol for value assignment are available for calibration

**Example:** ERM/IFCC-DA470k Proteins in Human Serum from the IRMM

- Many of the current reference materials have not been validated for commutability, ...have been shown to produce nonharmonized patient sample results for several measurands


- Replacement preparations (new lots) may have different relative amounts of the molecular species related to the measurand of interest and different matrix characteristics: Inconsistency & non-commutablity
AACC conference in October 2010:

• To address how to improve harmonization of laboratory test results for which there are no higher-order reference measurement procedures (RMPs), and for which it was unlikely that such procedures could be developed.
International Consortium for Harmonization of Clinical Laboratory Results (AACC) - ICHCLR

- [http://www.harmonization.net/Pages/default.html](http://www.harmonization.net/Pages/default.html)

Harmonization strategies 1(2)

<table>
<thead>
<tr>
<th>Attribute</th>
<th>Method 1</th>
<th>Method 2</th>
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<tbody>
<tr>
<td>Scheme</td>
<td></td>
<td></td>
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<tr>
<td>Reference measurement procedures</td>
<td></td>
<td></td>
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<tr>
<td>Reference materials</td>
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- Scheme: Hierarchical standardization per ISO 17511:2003. Top down approach passing 'trueness' to lower order measurement procedures and calibrators.
- Inter-method comparison as described by International Consortium for Harmonization of Clinical Laboratory Results (ICHCLR) (www.harmonization.net).
- Bottom up approach among routine (commercial) measurement procedures, with no SI traceability.

- Reference measurement procedures: One or more higher order reference measurement procedures available, preferably fulfilling requirements of ISO 15193:2009. None available.
- Reference materials: Certified purified reference materials and/or commutable secondary reference materials. No higher order reference materials available. Panel(s) of commutable human samples assigned consensus values through harmonization studies. Some International Conventional Calibrators may be available (e.g. WHO materials), but usually not commutable.
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<td>Calibration traceability</td>
<td></td>
<td>• The term “harmonized” is generally used when results are equivalent</td>
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<tr>
<td></td>
<td></td>
<td>either by being traceable to a reference material or based on a</td>
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<td>consensus approach, such as agreement to an all-methods mean, but</td>
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<td>neither a higher-order primary reference material nor an RMP exists.</td>
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<td>Sustainability</td>
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General process for assessing and achieving equivalency of clinical laboratory measurement results:

Healthy, diseased, and those who have related pathological conditions that may influence the measurand.

1. Review of Literature
   - Redefinition of measurand
   - Reevaluation of measurement procedure specificities
   - Improved measurement procedures if needed

2. Assessment Study:
   1. Evaluate current degree of measurement equivalence
   2. Determine whether harmonization is possible

3. Data Analysis
   - Panel of samples from healthy and diseased individuals
   - Existing or candidate reference materials
   - Manufacturer’s internal calibrators/controls

4. Harmonization is technically achievable
   - Yes: Harmonization Effort
     - Equivalent Clinical Measurement Results
   - No: Review of Literature

5. Final step: Equivalent Clinical Measurement Results
ISO 15193 — Requirements for Reference Measurement Procedures
ISO 15194 — Requirements for Reference Materials
ISO 15195 — Requirements for Reference Laboratories
PWI — Harmonized Measurement Procedures
The total testing chain

If analytical errors ≈ 10-15%;

*Is it worth to strive with analytical harmonization?*

Test ordered

Clinical phase

Analytic phase

Postanalytic phase

Preanalytic phase

Test ordered

Clinical response

Result interpreted in full clinical context

Results conveyed to clinician

Interpretation in the laboratory

Quality control

Measuring sample

Calibration

Analytic phase

Sample identification

Transporting sample

Result from definitive method and calibrator

Result from reference method and calibrator

Result from manufacturer method and calibrator

Result from routine method and calibrator

International standard Preferably SI units

Traceability

Uncertainty

An unbroken chain of comparisons and uncertainty estimations
Is accuracy important?

• Somewhere in the transition from the widely used manual methods of yesterday to the extensive use of automated today methods.


• Evidently, accuracy as a criterion for the selection of instruments and procedures is now overshadowed by considerations of simplicity and expediency.

Laboratory errors; is the analytical phase really safer?

• Because millions of **TSH/gonadotropin** tests are carried out in UK hospital laboratories alone, ...**thousands** of patients could be adversely affected by errors from interferences.

• **...significant bias was observed in result comparison aldosterone assays,**...

• **Biotin** interference on immunoassay measurement

• **NIBS evaluation of NGS for 25 infectious agents.**
  thologist Neil Almond. Toward higher standards in viral diagnostics. The Pathologist 2017
Laboratory errors; is the analytical phase really safer?


However, despite the impressive improvement achieved in analytical quality, several lines of evidence demonstrate that further improvements in this field are advisable.

• Because millions of TSH/gonadotropin tests are carried out in UK hospital laboratories alone, ...thousands of patients could be adversely affected by errors from interferences.


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If analytical phase ‘probably’ is not really safer;

Why it get better marks in counting?

J. O. Westgard:
A simple "count" is not the right measure of "occurrence." In our Six Sigma Risk Analysis book, we recommended that occurrence be estimated using some form of a "defect rate" so the number of defects would be referenced to the number of results. Common methodology often ranks occurrence qualitatively in terms of number of events per day, week, month, or year.

We've often referenced the paper by Carraro and Plebani, Clin Chem 2007;53:1228-1342, particularly a small part of the discussion of the errors that caused patient harm. ... 46 of these caused inappropriate patient care, and 24 of those were due to analytical errors. That amounts to 52% of the errors that cause patient harm, even though only 15% of the total errors were analytical.
**Opinion Paper**

Mario Plebani*, Michael L. Astion, Julian H. Barth, Wenxiang Chen, César A. de Oliveira Galoro, Mercedes Ibarz Escuer, Agnes Ivanov, Warren G. Miller, Penny Petinos, Laura Sciacovali, Wilson Shcolnik, Ana-Maria Simundic and Zorica Sumarac

**Harmonization of quality indicators in laboratory medicine. A preliminary consensus**

**PRIORITY 1:**

- IQC Failure: Percentage of “Number of tests with CV% higher than selected target, per year/Total number ...”
- No EQAS: Percentage of “Number of tests without EQA-PT control/Total number of tests in the menu”
- EQAS Failur: Percentage of “Number of unacceptable performances in EQA-PT.../Total number...”

**PRIORITY 3:**

- Unacceptable performances in EQA-PT

**Is accuracy important?**
Thank you