The six Traceability Models of ISO 17511:2020

Authored by Elvar Theodorsson (elvar.theodorsson@liu.se, +46736209471) and reviewed by the JCTLM Traceability Education & Promotion Working group and by other beneficiaries of the JCTLM

BIPM in Paris in 2022

This is a “living document”, version 2022-03-23.

Suggestions for improvements are gratefully received.

JCTLM Secretariat

Bureau International des Poids et Mesures
Pavillon de Breteuil
92312 Sèvres
Cedex France
Tel: +33 1 45 07 70 70
Fax: +33 1 45 34 20 21
Email: jctlm@bipm.org

The six Traceability Models of ISO 17511:2020

Six calibration hierarchies and value transfer models are available depending on the availability traceability to SI, of higher-order reference materials, of reference measurement procedures, and harmonization protocols, as depicted in Figure 1.

Calibration in the traceability hierarchy means using the value of the previous reference material in the traceability hierarchy to assign value through measurement to the next calibrator in the hierarchy. This is primarily accomplished by purchasing a commutable certified reference material at two or more appropriate concentrations, which can be used for calibrating a linear measurement equation used for measurement – which in this case means *value transfer*. If only a single concentration of the certified reference material is available, a serial dilution of the reference material in the corresponding matrix for calibrating the measurement procedure is needed.

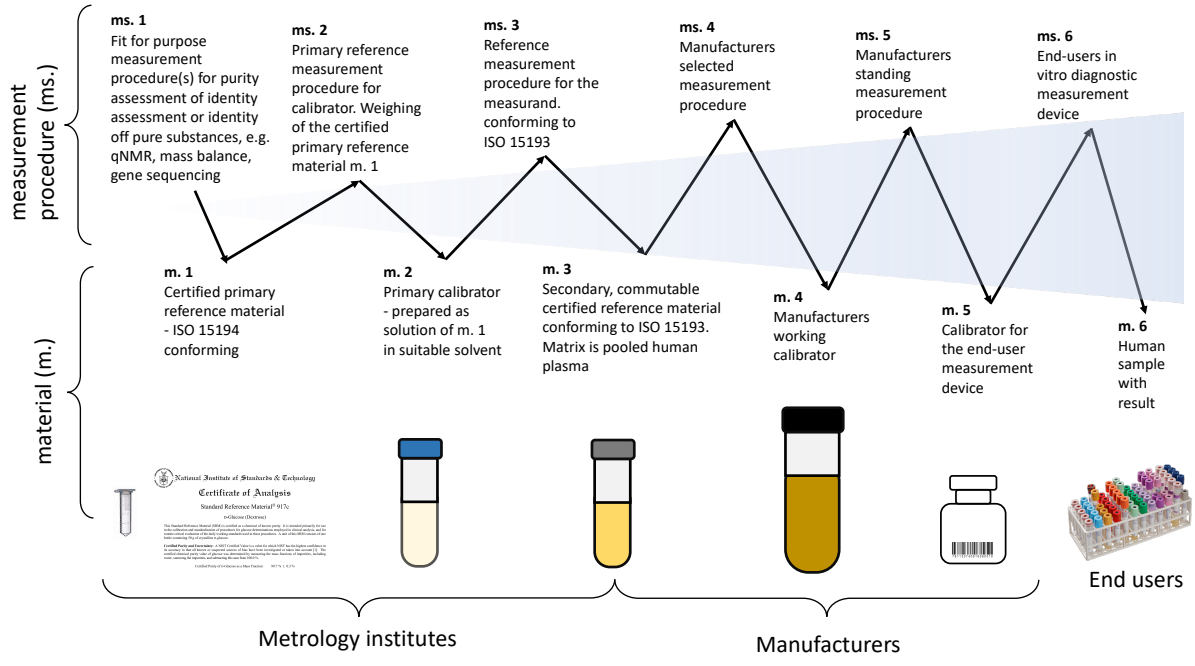

Figure 1: Transferring the value of previous reference material in the calibration hierarchy to the following material by successive measurement steps. The calibrator preceding the measurement step is used for calibration in each measurement step. The assignment of value to the subsequent calibrator is performed by measurement and using the measurement equation using this calibration.

Certified reference materials are self-evident at the top/beginning of the traceability hierarchy and end-user calibrators further on in the hierarchy.

In several calibration hierarchies, the quantity being measured changes along with the steps in the traceability hierarchy. In Figure 1 the value of the primary calibrator in m. 2 is transferred to m. 3 by isotope dilution mass spectrometry using chromatographic characteristics and the mass of the analyte, and when the value of m. 3 is transferred to m. 4, an enzymatic reaction combined with absorbance measurement is used.

The calibration hierarchy is a sequence of consecutive calibrations and value assignments, alternating between measurement procedures found fit for the intended use and appropriate reference materials (calibrators), beginning with certified reference material and reference measurement procedures and ending with values for the measurand in the intended human samples as determined with the end-user in vitro end-user in vitro diagnostic measurement reagents and system.

The technical documentation of the calibration hierarchy needs to include a *figure or other illustration* describing the linkage from the ultimate results using the human samples examined with the specified measuring system up to the highest available metrological reference.

For each step in the calibration hierarchy*, the quantity being measured* in the relevant reference material or human samples, in the case of the final measurements with a measuring system, must be identified, and the relationship between the measured quantity (or quantities) and the measurand must be established.

For a given measurand, *the metrologically highest-ranked measurement procedure*, *measurement protocol, or calibration material* in the calibration hierarchy must be identified and used to define the highest level of metrological traceability for the actual in vitro diagnostic measuring system to be used for analyzing patient samples.

The requirements of the two standards mentioned here apply *ISO-15194* - "In vitro diagnostic medical systems — Measurement of quantities in samples of biological origin — Requirements for certified reference materials and the content of supporting documentation" (1) and *ISO-15193* "In vitro diagnostic medical systems —

Measurement of quantities in samples of biological origin — Requirements for content and presentation of reference measurement procedures" (2).

For a measuring system that claims *metrological traceability of reported values for human samples to the SI*, the defined calibration hierarchy must be supported by available higher-order references, including either or both reference materials and reference measurement procedures that enable the realization of the appropriate SI unit for the corresponding measurand.

To claim metrological traceability for calibration and reported values (International Units) using a *non-SI traceable measuring system*, the calibration hierarchy for the measuring system must be defined in a manner that enables the consistent realization of the corresponding (non-SI) international units (IU).

The number of steps in the calibration hierarchy chain may be modified, provided that the changes are validated, and the metrologically highest elements of the hierarchy are retained.

The measured quantity at different steps in the calibration hierarchy commonly changes as the reference materials/calibrators and measurement procedures change throughout the traceability hierarchy. Changes to the measured quantity may result in different SI reporting units.

For *complex measurands* such as specific proteins in human blood plasma, the measured quantity in a calibration hierarchy at the highest level is often the purity of the intact protein is commonly expressed as a mass fraction, mg/g. At lower levels in the calibration hierarchy, the measurand is often the amount-of-substance concentration of specific epitopes or peptides derived from the protein of interest. In such cases, the measured quantity may be different at different levels in the hierarchy, and the assigned values for various reference materials/calibrators are appropriately expressed in other SI units.

Some measurable quantities cannot be expressed in terms of the seven base or derived quantities of the SI but have *the nature of a count* such as the number of CD4 cells, number of copies of a specific nuclear acid sequence, etc., per unit volume. A complete description of the quantity being counted is essential even in such instances. Traceability to the SI for counts is established through appropriate, validated counting measurement procedures as described in ISO 20391, ISO 20395, and (3).

The ISO 17511:2020 (4) standard details six typical traceability hierarchies principally described below. They are different depending on the availability or not of the following four prerequisites:

1. Traceability to SI
2. Availability of certified reference materials
3. Availability reference measurement procedures
4. Availability of harmonization protocols

Calibration Hierarchy 1 (CH1): Both primary reference material and reference measurement procedure are available

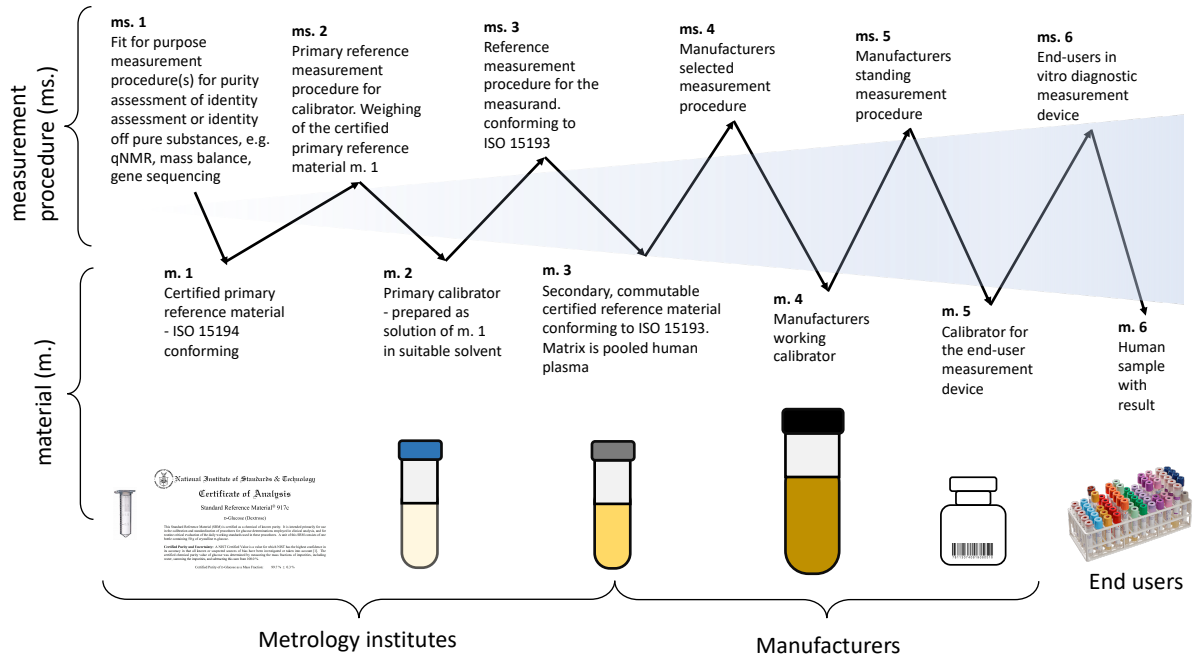

Figure 2: Calibration hierarchy 1 (CH1) Illustration of a traceability hierarchy applicable when both primary reference material and reference measurement procedure available

Primary reference measurement procedures and other fit for the intended use measurement procedures (CH1, ms.1, ms.2) provide metrological traceability to a SI unit of measurement with the slightest achievable measurement uncertainty. More than one primary reference measurement procedure can be used to assign values for quantities of a given kind to primary calibrators. The values obtained by two or more primary reference measurement procedures for a given measurand must not be significantly different within a stated uncertainty at a confidence level for the appropriate analytical quality specifications.

A selected primary reference measurement procedure (CH1, ms.1) must be amongst the best available realization of the unit of measurement, including the slightest achievable relative standard measurement uncertainty. The primary reference material must have its value assigned either directly by a primary reference measurement procedure or by a fit for the intended use measurement procedure for identity and purity assessment of pure substances, e.g., quantitative nuclear magnetic resonance, mass balance, or gene- or amino acid sequencing (5, 6). The value assignment and documentation for a primary reference material must conform to ISO-15194 – “In vitro diagnostic medical systems --

Measurement of quantities in samples of biological origin -- Requirements for certified reference materials and the content of supporting documentation" (1).

The *primary reference materials* (CH1, m.1) are usually highly purified. They contain a physiochemically well-defined analyte, evaluated for compositional stability integrity, and accompanied by a certificate (t - a certified reference material).

A *primary calibrator* (CH1, m.2) must be prepared from a primary reference material [m.1] and value-assigned using a primary reference measurement procedure (HC1, ms.2). The primary reference measurement procedure commonly involves weighing, with the dissolving of a measured mass of the primary reference material in a measured mass of an appropriate solvent.

An appropriate reference measurement procedure (CH1, ms.3) for the measurand must be used to *assign a value to a secondary calibrator or secondary reference material* (CH1, m.3) with a complex matrix. For the documentation of the reference measurement procedure (HC1, ms.3) for the measurand, the requirements of ISO-15193:2009 – "In vitro diagnostic medical systems - Measurement of quantities in samples of biological origin - Requirements for content and presentation of reference measurement procedures" must apply (2).

In cases where more than one reference measurement procedure is available, or when multiple available reference laboratories can perform the same measurement procedures for the measurand, *proficiency testing programs* such as IFCC External Quality Assessment Scheme for Reference (calibration) Laboratories in Laboratory Medicine (7-9) can provide helpful information regarding equivalence among different reference measuring systems and other reference laboratories.

The *secondary calibrators or secondary reference materials* (HC1, m.3) must be commutable with human samples as determined in commutability assessment studies (10-20).

The manufacturer's selected measurement procedure (HC1, p.4) must include a measuring system calibrated by one or more commutable calibrators or reference materials (HC1, m.3) when available.

The *manufacturer's working calibrator* is usually a material with a matrix resembling the human samples intended to be measured by the end-users in vitro diagnostic measuring system. Manufacturers often use panels of clinical samples or a series of pools of human clinical samples for working calibrators. The manufacturer's working calibrator (HC1, m.4) must have its value assigned according to the manufacturer's selected measurement procedure (HC1, ms.4), or - depending on commutability

characteristics of the working calibrator - according to the reference measurement procedure (HC1, ms.3). The secondary (working) calibrators (HC1, m.4) must be *commutable* with human samples as determined in commutability assessment studies comparing the manufacturer's selected measurement procedure (HC1, ms.4) and the manufacturer's standing measurement procedure (HC1, ms.5), or comparing the reference measurement procedure (HC1, ms.3) and the manufacturer's standing measurement procedure (HC1, ms.5) if steps (HC1, m.3) and (HC1, p.4) are omitted from the calibration hierarchy.

The manufacturer's standing measurement procedure (HC1, ms.5) must define a measurement procedure calibrated by one or more of the manufacturer's working calibrators or other commutable matrix calibrators and validated for analytical selectivity.

The *manufacturer's end-user calibrator* (HC1, m.5.) must have its value assigned according to the manufacturer's standing measurement procedure (HC1, ms.5) or the manufacturer's selected measurement procedure (HC1, ms.4) and is intended for calibration of the end-users measuring system (HC1, ms.6).

The *end-user measuring system* (HC1, ms.6) must describe a measuring system calibrated by one or more end-user calibrators. This measurement procedure, the final measurement procedure in the calibration hierarchy for the defined measurand, is used to examine human samples and generate final measured values for the measurand, with combined standard measurement uncertainties of the reported values to be estimated by the end-user, including all known measurement uncertainties gathered at each of the higher steps in the defined calibration hierarchy.

Calibration Hierarchy 2 (CH2): A primary reference measurement procedure defines the measurand

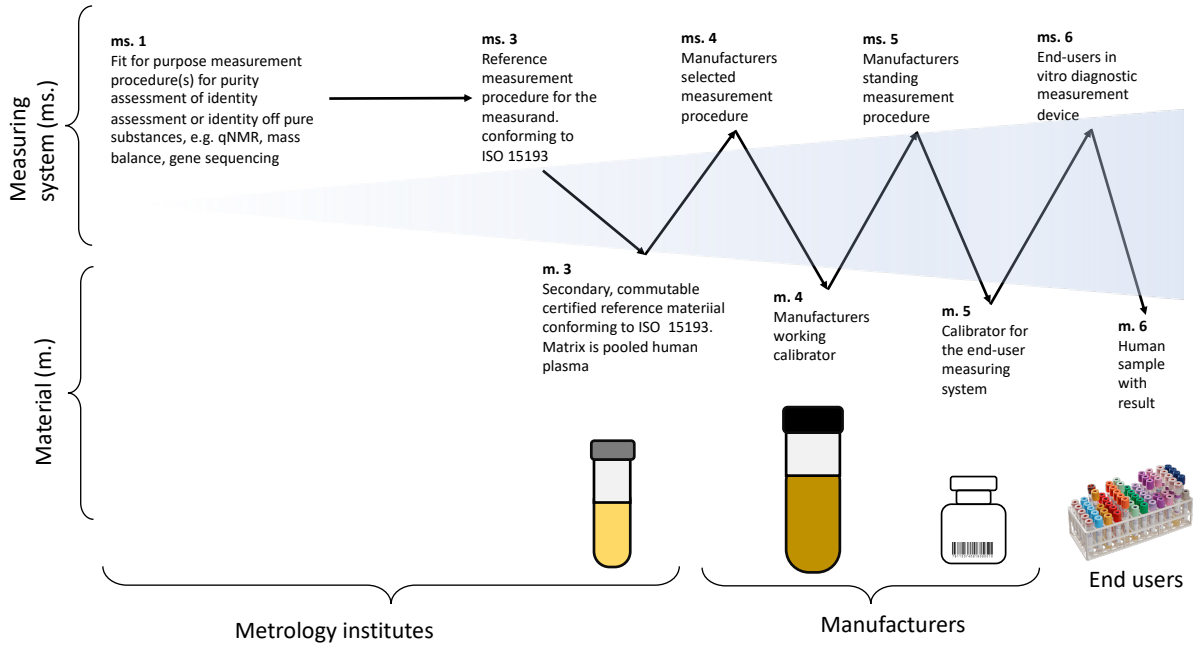

Figure 3: Calibration hierarchy when a reference measuring system defines the measurand, but no primary reference material for the quantity; traceable to SI is available.

Figure 3 describes a model calibration hierarchy for measurands with *a primary reference measurement procedure that defines the measurand with metrological traceability to SI when* no certified primary reference materials are available. In such cases, exemplified by calibration hierarchies for catalytic activity concentration of enzymes, including some blood coagulation factors in human plasma, metrological traceability to SI is based on thoroughly defined and internationally agreed reference measurement procedures.

Catalytic concentration measurements are only comparable among different laboratories if the enzyme activities are measured *under the same conditions*. Therefore, an enzyme measurand must be described both by all the following: kind-of-quantity, name of enzyme and system, and the specified measurement procedure, especially the indicator component of the measured reaction. At the top of the calibration hierarchy, t*he primary reference measurement procedure must be internationally agreed upon*.

For a measurand that is the catalytic concentration of an enzyme, the primary reference measurement procedure (HC2, ms.3) is an integral part of the definition of the measurand. Therefore, the primary reference measurement procedure (HC2, p.3) must be specified in sufficient detail regarding equipment, reagents, reaction conditions, and

calculation from the measured signal. The reference measurement procedure can be reproduced in any qualified laboratory that intends to perform the measurement.

The *documentation for a primary reference measurement procedure* used in a calibration hierarchy as described in HC2, p.3 must meet the requirements of ISO-15193:2009 "In vitro diagnostic medical devices - Measurement of quantities in samples of biological origin - Requirements for content and presentation of reference measurement procedures" (2). In addition, the description of the primary reference measurement procedure for the measurand (HC2, p.3) must include highly detailed information about the measurement procedure detailed in the *ISO-17511:2020* (4) – "In vitro diagnostic medical devices — Requirements for establishing metrological traceability of values assigned to calibrators, trueness control materials, and human samples" standard.

Secondary reference materials or calibrators (HC2, m.3) usually have a matrix resembling the human samples intended to be measured by the end-users routine measurement procedures to improve the likelihood that these materials will be commutable with human samples when used in lower-order measurement procedures in the calibration hierarchy. Panels- or pools of human samples- are secondary reference materials applicable in this context (HC2, m.3), depending on biochemical characteristics (e.g., stability) of the measurand.

The primary reference measurement procedure for the measurand (HC2, ms.3) must be used to *assign a value to a secondary calibrator or secondary reference material* (HC2, m.3) with a complex matrix.

The *secondary reference materials* (HC2, m.3) have certified values with associated uncertainties and are value-assigned by a calibration laboratory using a fit-for-purpose primary reference measurement procedure.

A *manufacturer's selected MP* (HC2, p.4) must define a measurement procedure that is calibrated by one or more secondary reference materials or secondary calibrators (HC2, m.3) and is used to assign values to the manufacturer's working calibrator(s) (HC2, m.4).

The *manufacturer's working calibrator* (HC2, m.4) must have values assigned according to the manufacturer's selected measurement procedure (HC2, p.4) or - depending on the commutability characteristics of the working calibrator - according to a primary reference measurement procedure (HC2, p.3) for the measurand. The secondary (working) calibrators (HC2, m.4) must be commutable with human samples as determined in commutability assessment studies comparing the manufacturer's selected measurement procedure (HC2, p.4) and the manufacturer's standing

measurement procedure (HC2, p.5), or comparing the reference measurement procedure (HC2, p.3) and the manufacturer's standing measurement procedure (HC2, p.5) if steps (HC2, m.3) and (HC2, p.4) are omitted from the calibration hierarchy.

The manufacturer's standing measurement procedure (HC2, p.5) must define a measurement procedure calibrated by one or more of the manufacturer's working calibrators (HC2, m.4) or higher-ranking calibrators. It must also be validated for analytical selectivity.

The *manufacturer's end-user calibrator* (HC2, m.5) is intended to calibrate the end-users in vitro diagnostic measuring system. It must have its value assigned according to the manufacturer's standing measurement procedure (HC2, p.5).

The manufacturer must estimate the total uncertainty of the assigned value of the end-user calibrator (HC2, m.5). It must incorporate all relevant higher-order uncertainties and the uncertainties of each of the subsequent measurements in the calibration hierarchy down to and including the manufacturer's standing measurement procedures (HC2, p.5).

The end-user of the vitro diagnostic measuring system (HC2, p.6) must describe a measuring system calibrated by one or more end-user calibrators. This measurement procedure, the final measurement procedure in the calibration hierarchy, is used to examine human samples and generate the final measured values for the measurand, with combined standard measurement uncertainties of the reported values to be estimated by the end-user, including all known measurement uncertainties obtained at each higher step in the defined calibration hierarchy.

Calibration Hierarchy 3: Measurands defined by a reference measurement procedure calibrated with a particular primary calibrator

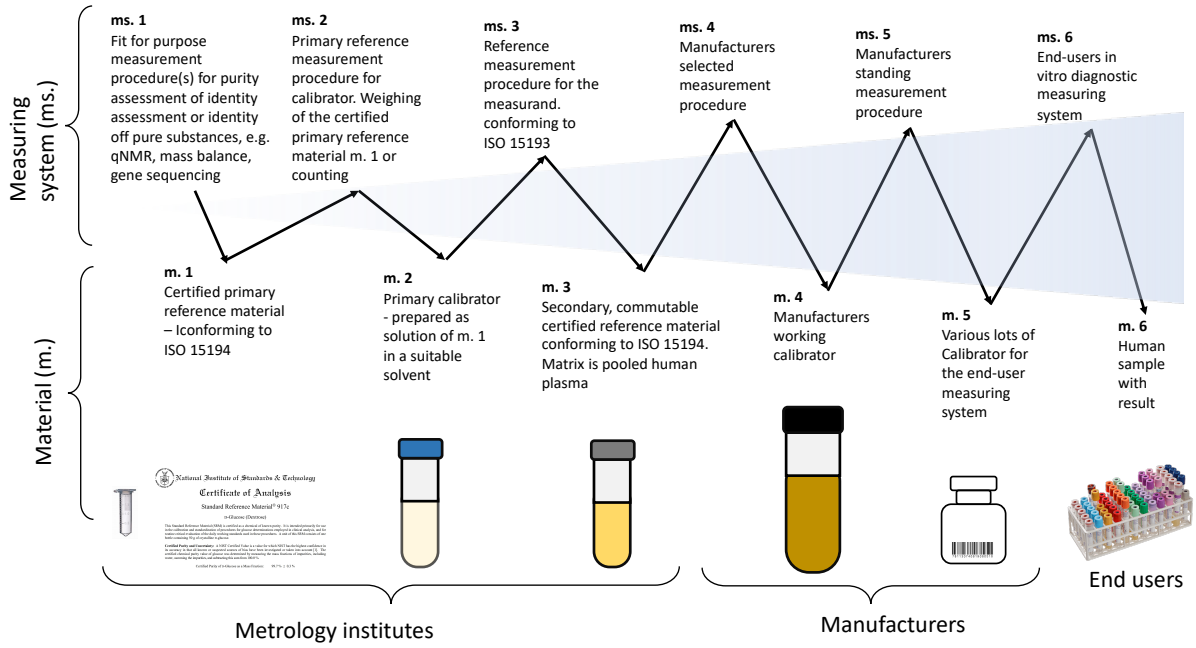

Figure 4: Measurand defined by a reference measuring system calibrated with a particular primary calibrator traceable to SI

Calibration hierarchies for measurands that are defined by a reference measurement procedure calibrated with a particular primary calibrator (with traceability to SI) are described in Figure 4. In such cases, the reference measurement procedure detects a quantity that is a component of the measurand (for example, a peptide fragment or an epitope) rather than the entire molecular structure of the quantity intended to be measured.

A higher-order *reference measurement procedure* (Figure 3, ms.3) that is calibrated with a particular primary calibrator (HC3, m.2) must define the measurand due to its selectivity for a specific epitope or molecular structure that is part of the measurand (21-30).

The *secondary reference materials or secondary calibrators* will usually have a matrix resembling the human samples intended to be measured by the end-user in vitro diagnostic measuring system to improve the likelihood that these reference materials will be commutable with human samples making sure that they are suitable for use with the measurement procedures that they are intended to calibrate (HC3, ms.4 and ms.5)

The *manufacturer's selected measurement procedures* (HC3, ms.4) must define a measuring system calibrated by one or more secondary calibrators or secondary reference materials (HC3, m.3). Its primary purpose is to transfer trueness to the

manufacturer’s working calibrator (HC3, m.4). This measurement procedure shall be selected partly because the calibrators (HC3, m.3, and m.4) are commutable with human samples. A manufacturer's working calibrator is commonly a material with a matrix resembling the intended human samples to be used with the end-user in vitro diagnostic measuring systems, such as a panel or a series of pools of human samples.

The manufacturer's working calibrator (HC3, m.4) must have its value assigned according to the manufacturer's selected measurement procedure (HC3, p.4). The calibration material (HC3, m.4) must have demonstrated commutability with the intended human samples to ensure its suitability for use with the manufacturer's selected measurement procedure (HC3, p.4) and the procedure to be calibrated, i.e., the manufacturer’s standing measurement procedure (HC3, p.5).

The manufacturer's standing measurement procedure (HC3, p.5) must define a measurement procedure calibrated by one or more of the manufacturer's working calibrators (HC3, m.4) or higher ranked calibrator and must be validated for analytical selectivity.

The manufacturer's end-user measuring system calibrator (HC3, m.5) shall have its value according to the manufacturer's standing measurement procedure (HC3, p.5) and is intended to calibrate the end-user in vitro diagnostic measuring system. The total uncertainty of the assigned value of the end-user invitro diagnostic measuring system calibrator (HC3, m.5) must be estimated by the manufacturer and incorporate all appropriate higher-order uncertainties in addition to the uncertainties of each of the subsequent measurement procedures in the calibration hierarchy down to and including the manufacturer’s standing measurement procedure HC3.

The *end-user measuring system* (Fig. 3, ms.6) must describe a measuring system calibrated by one or more end-user calibrators. This measurement procedure, the final measurement procedure in the calibration hierarchy for the defined measurand, is used to examine human samples and generate final measured values for the measurand, with combined standard measurement uncertainties of the reported values to be estimated by the end-user, including all known measurement uncertainties accumulated at each higher step in the defined calibration hierarchy.

Calibration Hierarchy 4: Measurand defined by value assignment protocol for international conventional calibrator (no SI traceability but conforming to ISO-15194:2009)

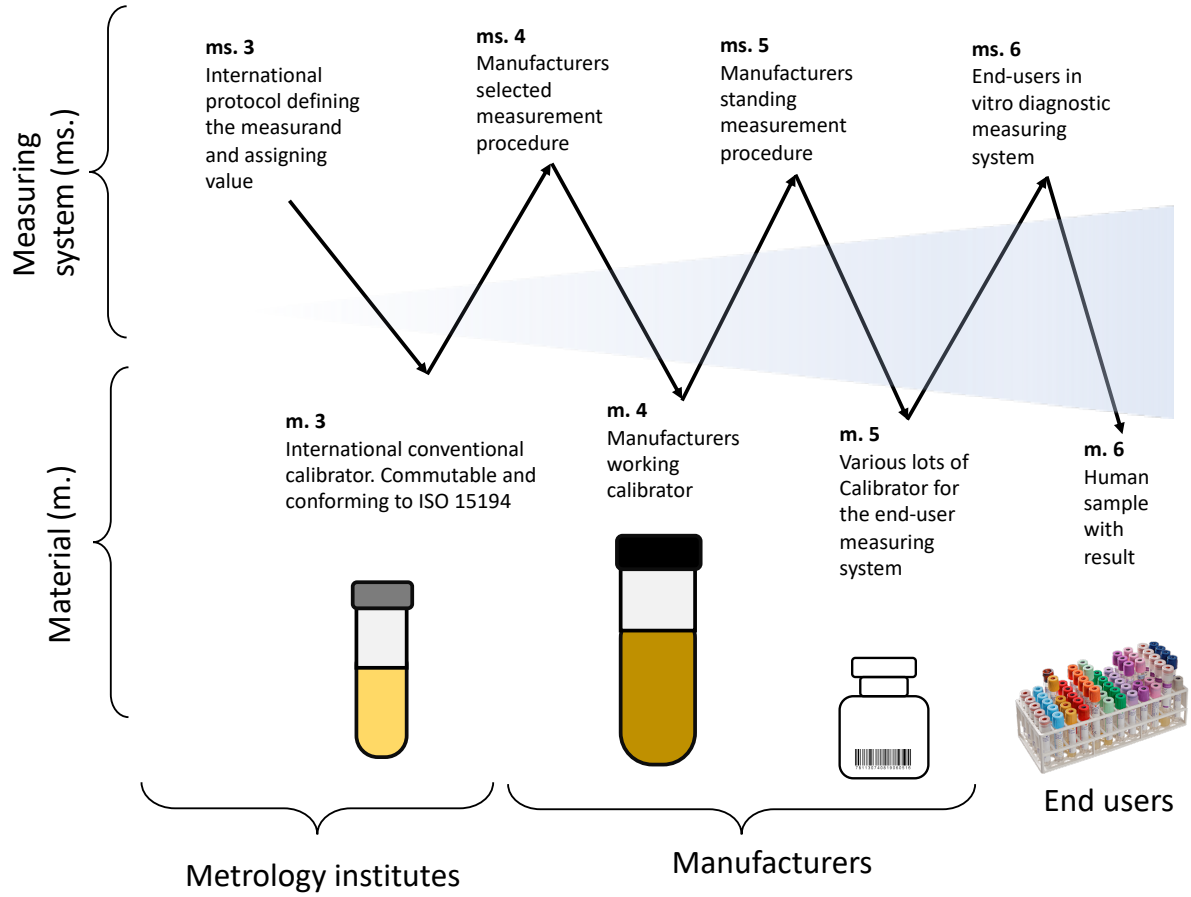

Figure 5: Measurand defined by value assignment protocol for international conventional calibrator (conforming with ISO 15194 but with no SI traceability).

The calibration hierarchy described in Figure 5 applies to cases where there is an *international conventional calibrator that defines the measurand* (CH4, m.3), and which conforms with the requirements of *ISO-15194*:2009 – "In vitro diagnostic medical devices -- Measurement of quantities in samples of biological origin -- Requirements for certified reference materials and the content of supporting documentation" standard. For these kinds of measurable quantities, there are no reference measurement procedures, no primary reference materials or primary calibrators, and no traceability to SI.

The *manufacturer's working calibrators* frequently consist of pools of human samples with a matrix resembling the natural human samples intended to be measured by the end-user measuring systems. The *manufacturer's working calibrator* (CH4, m.4) must have its value assigned according to the manufacturer's selected measurement procedure (CH4, ms.4). The manufacturer's working calibrator (CH4, m.4) must have demonstrated commutability with the intended human samples in the manufacturer's

selected measurement procedure (CH4, ms.4) and the manufacturer’s standing measurement procedure (CH4, ms.5).

The *manufacturer's standing measurement procedure* (CH4, ms.5) must define a measurement procedure calibrated by one or more of the manufacturer's working calibrators (CH4, m.4) or higher types of calibrator such as the international conventional calibrator (CH4, m.3) and is validated for measurement selectivity.

The *end-user measuring system calibrator* (CH4, m.5) must have its value assigned according to the manufacturer's standing measurement procedure. It is intended to calibrate the end-user measuring system (CH4, ms.6). The total measurement uncertainty of the assigned value of the end-user measuring system calibrator (CH4, m.5) must be estimated by the manufacturer, including all steps in the calibration hierarchy, including the manufacturer’s standing measurement procedure.

The *end-user measuring system* (CH4, ms.6) must describe a measuring system calibrated by one or more end-user calibrators. The final step in the calibration hierarchy for the defined measurand is used to examine human samples and generate final measured values for the measurand, with combined standard measurement uncertainties of the reported values to be estimated by the end-user, including all known measurement uncertainties accumulated at each higher step in the defined calibration hierarchy.

Calibration Hierarchy 5: Measurand defined by an international harmonization protocol (not traceable to SI and certified reference material is not available)

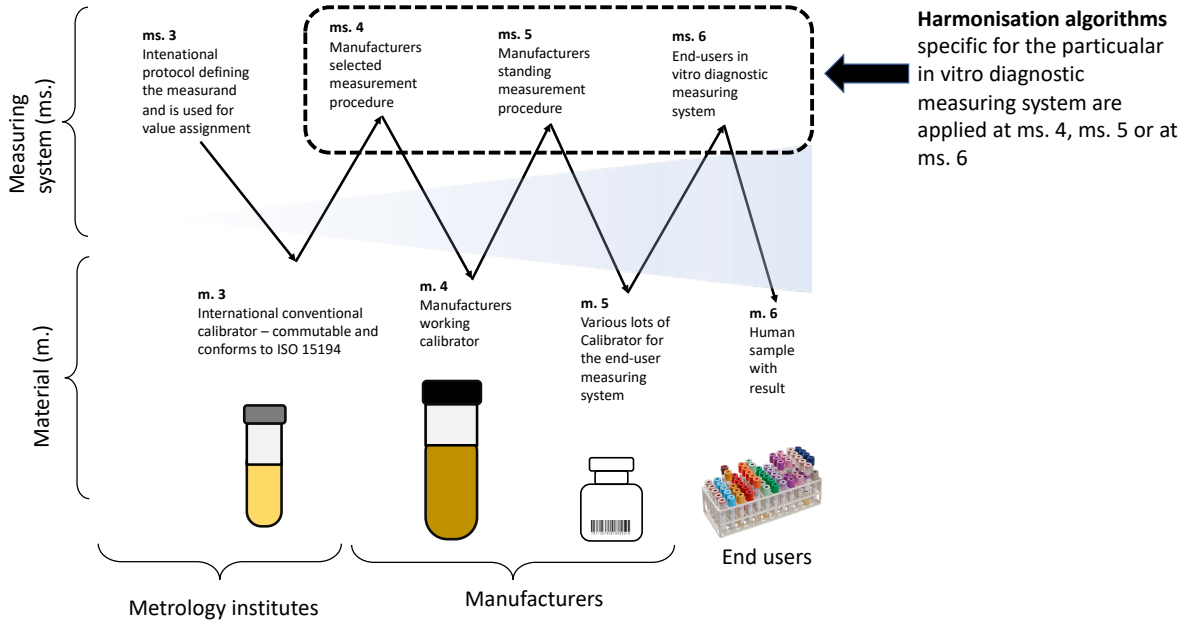

Figure 6: Measurand defined by international harmonization protocol

The calibration hierarchy described in Figure 6 applies to cases in which an international harmonization protocol defines the measurand in human samples. Still, there are no internationally agreed reference measurement procedures, no primary reference materials, no conventional reference measurement procedures or reference materials, and no metrological traceability to the SI.

The *international harmonization protocol* for the measurand (CH5, p.3) defines the highest metrological level in the calibration hierarchy. It is intended to achieve equivalence of reported results for the intended human samples (CH5, m.6) among available harmonized end-user measuring systems (CH5, p.6) for the measurand. The standard *ISO-21151:2020* – "In vitro diagnostic medical devices – Requirements for international harmonization protocols establishing metrological traceability of values assigned to calibrators and human samples" details harmonization protocols in a metrologically traceable calibration hierarchy (31).

The *international harmonization protocol* (CH5, ms.3) must specify the process used to assign harmonized values (arbitrary units, not traceable to SI) to the harmonization reference materials (CH5, m.3). The international harmonization protocol (CH5, ms.3) must specify how the harmonization reference materials are used to estimate relationships between the results for the harmonization reference materials (CH5, m.3) among the measuring systems (CH5, p.6) participating in the harmonization protocol. The overall measurement uncertainties associated with the assigned quantity values for

the harmonization reference materials must be estimated and accounted for in the estimate of the calibrators for the specified in vitro diagnostic measuring system.

Each measuring system manufacturer must determine and document a measuring system-specific algorithm that when applied to (a) their selected measurement procedure (CH5, ms.4) for assignment of values to their working calibrator or calibrators (CH5, m.4), or (b) to their standing measurement procedure (CH5, ms.5) for assignment of values to their end-user in vitro diagnostic measuring system calibrator or calibrators (CH5, m.5), or (c) to their harmonized in vitro diagnostic measuring system (CH5, p.6) for assignment of values to human samples (CH5, m.6), will enable achievement of equivalent results for human samples with their specified in vitro measuring system when compared to other in vitro diagnostic measuring systems participating in the harmonization protocol. The measurement uncertainty introduced by the in vitro diagnostic measuring system-specific harmonization algorithm must also be accounted for and included in the estimate of the measurement uncertainty component at the applicable position in the calibration hierarchy, as well as in the final combined measurement uncertainty of the end-user calibrator(s), for each harmonized measuring system.

The end-user in vitro diagnostic measuring system (CH5, p.6) must describe a measuring system calibrated by one or more specified end-user in vitro diagnostic measuring system calibrators (CH5, m.5). This measurement procedure, the final measurement procedure in the calibration hierarchy for the defined measurand, is used to examine human samples and generate final measured values for the measurand, with combined standard measurement uncertainties of the reported values to be estimated by the end-user, including all known measurement uncertainties accumulated at each higher step in the entire calibration hierarchy.

Calibration Hierarchy 6: Measurand defined by manufacturer's internal arbitrarily defined reference material (not traceable to SI, no certified reference material, no reference measurement procedure, and no harmonization protocol)

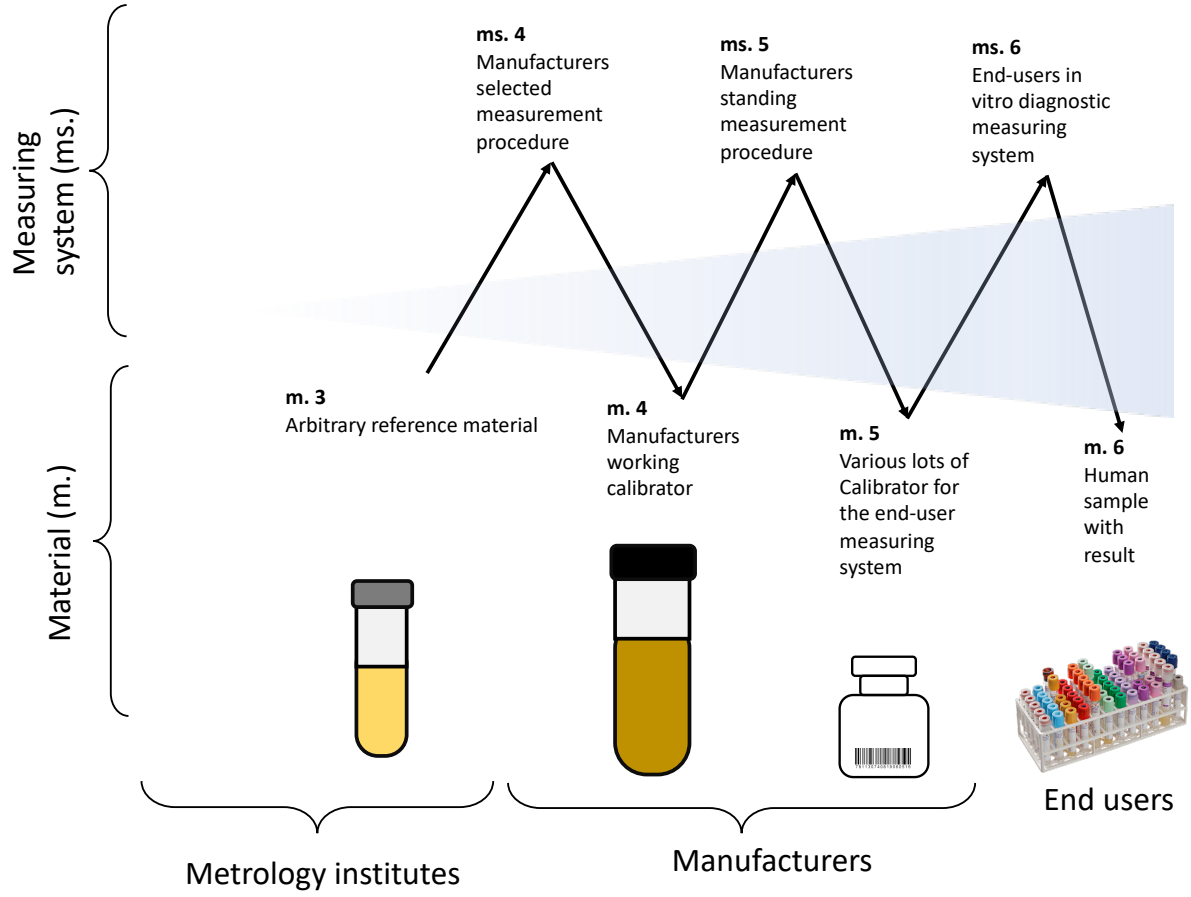

Figure 7: Measurand defined by manufacturer's internal arbitrarily defined reference materials (no primary reference materials or certified reference materials; no reference measuring system; no harmonization protocol; not traceable to SI).

The calibration hierarchy described in Figure 7 applies to measurands with metrological traceability only to a manufacturer's internal, arbitrarily defined reference materials (CH6, m.3), for certain measurands with no certified pure reference materials (CH6, m.1), no primary calibrators (6, m.2), no reference measurement procedures (CH6, ms.3), no harmonization protocol, and no traceability to SI.

Since higher-order reference materials and measurement procedures (CH6, m.1, m.2, ms.1, ms.2, ms.3) do not exist for these types of measurands, arbitrary reference materials (CH6, m.3) can be established as the metrologically highest level in the defined calibration hierarchy for the measurand.

To ensure consistency in the calibration of the specified end-user measuring systems, manufacturers sometimes establish *arbitrary measurement procedures* (CH6, ms.4) and

arbitrary reference materials (CH6, m.3) as the highest level of these calibration hierarchies. Arbitrary reference materials (CH6, m.3) are at times prepared from, e.g., purified biomarkers, then measured with a selected measuring system (CH6, ms.4), enabling further calibration and value assignment to a working calibrator (CH6, m.4). Such working calibrators are often prepared in a human sample matrix material or other appropriate matrices or are comprised of, e.g., panels (or pools) of human samples or "spiked" human samples.

In cases where a *manufacturer establishes an independent calibration hierarchy using a selected measurement procedure* (CH6, ms.4) where there is no availability of reference material for the measurand (e.g., measurement procedures dependent on other properties such as UV absorptivity; procedures based on counting; etc.), the selected measurement procedure must be the metrologically highest level in the specified calibration hierarchy for the defined measurand.

The *manufacturer's standing measurement procedure* (CH6, ms.5) must define a calibrated measuring system with the manufacturer's working calibrator (CH6, m.4), which may be comprised of arbitrary reference materials, including panels of human samples or sample pools. The standing measurement procedure (CH6, p.5) shall be used to determine assigned values for the manufacturer's end-user measuring system calibrator(s) (CH6, m.5).

The *manufacturer's end-user IVD MD calibrators* (CH6, m.5) will have values assigned according to the manufacturer's standing measurement procedure (CH6, p.5) and are intended for use in the calibration of the end-users measuring system (CH6, p.6). The μcal of the assigned value of the end-user measuring system calibrator (CH6, m.5) must be estimated by the manufacturer, incorporating all appropriate higher-order uncertainties in addition to the uncertainties of each of the subsequent measurement procedures in the calibration hierarchy down to and including the manufacturer's standing measurement procedure (CH6, p.5).

The *end-user in vitro diagnostic measuring system* (CH6, p.6) will describe a measuring system calibrated by one or more end-user measuring system calibrators. This measurement procedure (CH6, ms.6), the final measurement procedure in the calibration hierarchy for the defined measurand, is used to examine human samples and generate final measured values for the measurand (CH6, m.6), with combined standard measurement uncertainties in the reported values to be estimated by the end-user, considering all known measurement uncertainties accrued at each higher step in the defined calibration hierarchy.

Documentation of the calibration hierarchy to be included in the manufacturer's technical file for the specified in vitro diagnostic measuring system for a measurand with metrological traceability according to the scheme in CH6 must include specifications and validation documentation for elements critical to the performance and reproducibility of the calibration hierarchy, including at least:

a) *specification of raw materials* to be prepared, purchased, processed, or otherwise acquired for preparation of any arbitrary reference materials (CH6, m.3, m.4) or reagents and other components of the measuring system (CH6, p.4, p.5) including specifications for any patient samples (or pools) or other types of samples intended to serve as reference materials at various levels (CH6, m.3, m.4) of the calibration hierarchy.

b) *specification of the measurand of interest* and any associated measured quantities and influence quantities appropriate to each clinical intended use of the specified measuring system in sufficient detail to enable reproducible human sample selection and pool preparation for subsequent batches of reference materials (CH6, m.3, m.4) as applicable. It is self-evident that the measurand present in each human sample selected as a calibration panel member (CH6, m.3, and m.4) is assumed to represent the measurand of interest for each stated intended use of the in vitro diagnostic measuring system.

c) for steps in the calibration hierarchy that are under the control of the manufacturer, procedures and work instructions for assignment of quantity values to human samples (or pools) or other arbitrary materials intended to serve as reference materials (CH6, m.3, m.4), including descriptions of measures to be taken to ensure consistency of the value assignment process for replacement batches of calibration panels or other arbitrary reference materials. Internal reference materials (CH6, m.3, m.4) are assigned values by the manufacturer using protocols including, for example, (a) arbitrary units of measurement, (b) standard addition of weighed or volumetrically dispensed volumes of a concentrate, (c) direct measurements using the manufacturer's selected MP (see CH6, p.4) (for example an available commercial measurement procedure), or (d) other scientifically valid methods as appropriate to the measurement technology and type of analyte. A sub-panel of human samples from the first calibration panel (CH6, m.3) is often used to transfer assigned values to a subsequent calibration panel.

References

1. ISO. ISO 15194:2009 In vitro diagnostic medical devices — Measurement of quantities in samples of biological origin — Requirements for certified reference materials and the content of supporting documentation. Technical Committee : ISO/TC 212 Clinical laboratory testing and in vitro diagnostic test systems. Geneva, Switzerland: International Organization for Standardization; 2009.
2. ISO. ISO 15193:2009 In vitro diagnostic medical devices — Measurement of quantities in samples of biological origin — Requirements for content and presentation of reference measurement procedures. Geneva: International Organization for Standardization; 2009.
3. Kammel M, Kummrow A, John M, Reitz S, Witt K, Neukammer J. Flow cytometer for reference measurements of blood cell concentrations with low uncertainty. Ieee Int Sym Med Mea. 2015:517-20.
4. ISO. ISO 17511:2020 In vitro diagnostic medical devices — Requirements for establishing metrological traceability of values assigned to calibrators, trueness control materials and human samples. Technical Committee : ISO/TC 212 Clinical laboratory testing and in vitro diagnostic test systems. Geneva, Switzerland: International Organization for Standardization; 2020.
5. BIPM. SI Brochure: The International System of Units (SI) https://www.bipm.org/en/publications/si-brochure/ 2019 [9th Edition:[
6. Westwood S, Choteau T, Daireaux A, Josephs RD, Wielgosz RI. Mass Balance Method for the SI Value Assignment of the Purity of Organic Compounds. Anal Chem. 2013;85(6):3118-26.
7. Kessler A, Siekmann L, Weykamp C, Geilenkeuser WJ, Dreazen O, Middle J, et al. External Quality Assessment Scheme for reference laboratories - review of 8 years' experience. Clinical chemistry and laboratory medicine : CCLM / FESCC. 2013;51(5):997-1005.
8. Kessler A. Traceability in Laboratory Medicine: A Review of the International Activities. Clinical Chemistry and Laboratory Medicine. 2011;49:S146-S.
9. IFCC. IFCC External Quality Assessment Scheme for Reference (calibration) Laboratories in Laboratory Medicine (RELA). International Federation of Clinical Chemistry and Laboratory Medicine (IFCC); 2008.
10. CLSI. CLSI EP 30-A, Characterization and Qualification of Commutable Reference Materials for Laboratory Medicine, 1st Edition. CLSI; 2016.
11. Budd JR, Weykamp C, Rej R, MacKenzie F, Ceriotti F, Greenberg N, et al. IFCC Working Group Recommendations for Assessing Commutability Part 3: Using the Calibration Effectiveness of a Reference Material. Clinical chemistry. 2018;64(3):465-74.
12. Miller WG, Schimmel H, Rej R, Greenberg N, Ceriotti F, Burns C, et al. IFCC Working Group Recommendations for Assessing Commutability Part 1: General Experimental Design. Clinical chemistry. 2018;64(3):447-54.
13. Nilsson G, Budd JR, Greenberg N, Delatour V, Rej R, Panteghini M, et al. IFCC Working Group Recommendations for Assessing Commutability Part 2: Using the

Difference in Bias between a Reference Material and Clinical Samples. Clinical chemistry. 2018;64(3):455-64.

14. Miller WG, Myers GL, Rej R. Why commutability matters. Clinical chemistry. 2006;52(4):553-4.
15. Vesper HW, Miller WG, Myers GL. Reference materials and commutability. Clin Biochem Rev. 2007;28(4):139-47.
16. Kimberly MM, Caudill SP, Vesper HW, Monsell EA, Miller WG, Rej R, et al. Standardization of high-sensitivity immunoassays for measurement of C-reactive protein; II: Two approaches for assessing commutability of a reference material. Clinical chemistry. 2009;55(2):342-50.
17. Miller WG, Erek A, Cunningham TD, Oladipo O, Scott MG, Johnson RE. Commutability limitations influence quality control results with different reagent lots. Clinical chemistry. 2011;57(1):76-83.
18. Miller WG, Myers GL, Lou Gantzer M, Kahn SE, Schonbrunner ER, Thienpont LM, et al. Roadmap for harmonization of clinical laboratory measurement procedures. Clinical chemistry. 2011;57(8):1108-17.
19. Korzun WJ, Nilsson G, Bachmann LM, Myers GL, Sakurabayashi I, Nakajima K, et al. Difference in Bias Approach for Commutability Assessment: Application to Frozen Pools of Human Serum Measured by 8 Direct Methods for HDL and LDL Cholesterol. Clinical chemistry. 2015;61(8):1107-13.
20. Vesper HW, Myers GL, Miller WG. Current practices and challenges in the standardization and harmonization of clinical laboratory tests. Am J Clin Nutr. 2016;104(3):907s-12s.
21. Jeppsson J-O, Kristensson H, Fimiani C. Carbohydrate-deficient transferrin quantified by HPLC to determine heavy consumption of alcohol. Clinical chemistry. 1993;39:2115-20.
22. Jeppsson JO, Arndt T, Schellenberg F, Wielders JPM, Anton RF, Whitfield JB, et al. Toward standardization of carbohydrate-deficient transferrin (CDT) measurements: I. Analyte definition and proposal of a candidate reference method. Clinical Chemistry and Laboratory Medicine. 2007;45(4):558-62.
23. Jeppsson JO, Kobold U, Barr J, Finke A, Hoelzel W, Hoshino T, et al. Approved IFCC reference method for the measurement of HbA(1c) in human blood. Clinical Chemistry and Laboratory Medicine. 2002;40(1):78-89.
24. Schellenberg F, Wielders J, Anton R, Bianchi V, Deenmamode J, Weykamp C, et al. IFCC approved HPLC reference measurement procedure for the alcohol consumption biomarker carbohydrate-deficient transferrin (CDT): Its validation and use. Clinica chimica acta; international journal of clinical chemistry. 2017;465:91-100.
25. Helander A, Wielders JP, Jeppsson JO, Weykamp C, Siebelder C, Anton RF, et al. Toward standardization of carbohydrate-deficient transferrin (CDT) measurements: II. Performance of a laboratory network running the HPLC candidate reference measurement procedure and evaluation of a candidate reference material. Clinical chemistry and laboratory medicine : CCLM / FESCC. 2010;48(11):1585-92.

26. Helander A, Wielders J, Anton R, Arndt T, Bianchi V, Deenmamode J, et al. Standardisation and use of the alcohol biomarker carbohydrate-deficient transferrin (CDT). Clinica chimica acta; international journal of clinical chemistry. 2016;459:19-24.

27. Hoelzel W, Weykamp C, Jeppsson JO, Miedema K, Barr JR, Goodall I, et al. IFCC reference system for measurement of hemoglobin A1c in human blood and the national standardization schemes in the United States, Japan, and Sweden: a method-comparison study. Clinical chemistry. 2004;50(1):166-74.

28. Finke A, Kobold U, Hoelzel W, Weykamp C, Miedema K, Jeppsson JO. Preparation of a candidate primary reference material for the international standardisation of HbA1c determinations. Clinical chemistry and laboratory medicine : CCLM / FESCC. 1998;36(5):299-308.

29. Weykamp C, Wielders J, Helander A, Anton RF, Bianchi V, Jeppsson JO, et al. Harmonization of measurement results of the alcohol biomarker carbohydrate-deficient transferrin by use of the toolbox of technical procedures of the International Consortium for Harmonization of Clinical Laboratory Results. Clinical chemistry. 2014;60(7):945-53.

30. Helander A, Eriksson G, Stibler H, Jeppsson JO. Interference of transferrin isoform types with carbohydrate-deficient transferrin quantification in the identification of alcohol abuse. Clinical chemistry. 2001;47(7):1225-33.

31. ISO. ISO 21151:2020 In vitro diagnostic medical devices — Requirements for international harmonisation protocols establishing metrological traceability of values assigned to calibrators and human samples. Technical Committee : ISO/TC 212 Clinical laboratory testing and in vitro diagnostic test systems. Geneva, Switzerland: International Organization for Standardization; 2020.