

Requirements of a reference measurement procedure and how they relate to a certified reference material for cTnI that is fit for purpose

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**MATERIAL
MEASUREMENT
LABORATORY**



What is a reference measurement procedure?

For what are reference measurement procedures used?

ISO/TC 15193 “In vitro diagnostic systems — Measurement of quantities in samples of biological origin — Presentation of reference measurement procedures”

- Described the documentation required for reference measurement procedures

— a reference measurement procedure is used to certify the concentration(s) of analyte(s) in reference materials or higher-order calibrants

As such, the performance specifications of a reference measurement procedure are defined by its intended use

Is a reference measurement procedure needed to certify reference materials?

Modes to Certify Reference Materials:
a NIST perspective¹

1. **Certification Using a Single Primary Method with Confirmation by Other Method(s)**
 - i. “A primary method of measurement is a method having the highest metrological properties, whose operation can be completely described and understood, for which a complete uncertainty statement can be written down in terms of SI units”
 - ii. Primary methods are higher-order reference measurement procedures

NIST Special Publication 260-136 “Definitions of Terms and Modes Used at NIST for Value-Assignment of Reference Materials for Chemical Measurements (2000).”

Modes to Certify Reference Materials: the NIST perspective¹ (Cont.)

- 2. Certification using two or more independent critically-evaluated methods**
- 3. Certification using one method at NIST and different methods by outside collaborating laboratories**

Is a reference measurement procedure needed?

Not necessarily

NIST Special Publication 260-136 “Definitions of Terms and Modes Used at NIST for Value-Assignment of Reference Materials for Chemical Measurements (2000).”

What are the specifications of a reference measurement procedure?

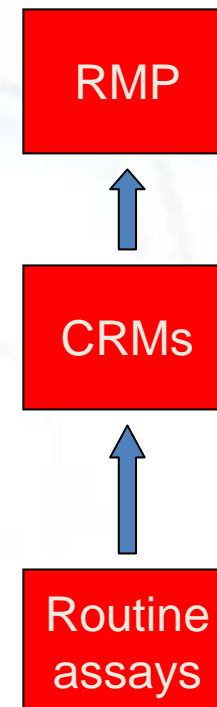
Specifications defined by intended use...



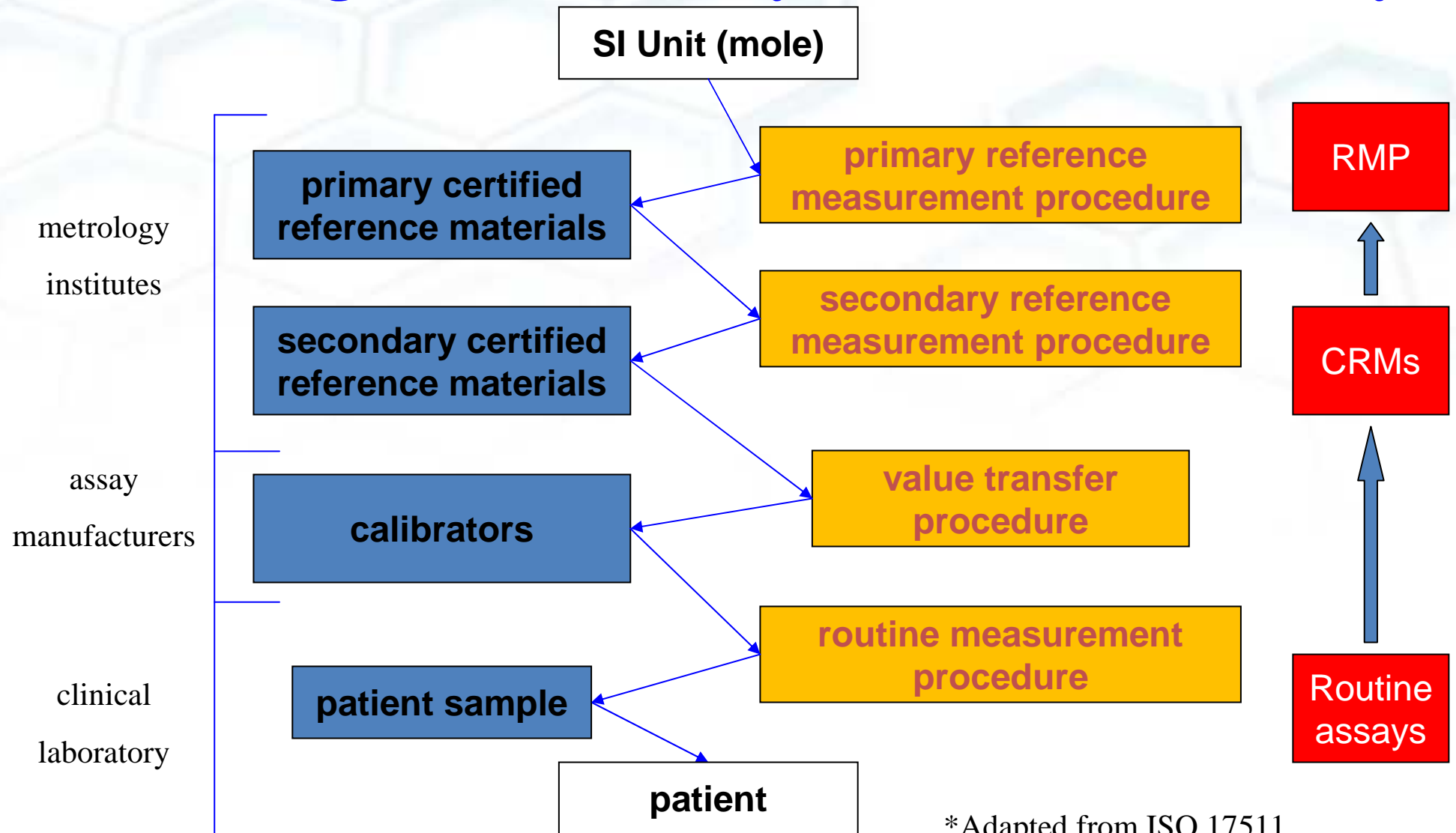
...intended use is the certification of reference materials...



...the specifications of certified reference materials are defined by the standardization needs of the routine assays



Metrological Traceability in Clinical Chemistry



*Adapted from ISO 17511

Measurement specifications for routine cTnI assays:

Measurement accuracy:

- Measurement accuracy is defined by knowledge of a “true” value of the concentration of cTnI in a sample
- A “true” value is usually embodied by a certified reference material
- No secondary (i.e., matrix-based) certified reference materials for cTnI currently exist

Measurement uncertainty:

- Measurement uncertainty (imprecision) of a routine cTnI assay is defined by the clinical needs of the measurement
- See
http://www.ifcc.org/PDF/ScientificActivities/IFCC_Troponin_Table_vDec_2010_FINAL_ng_L_28Jan11.pdf

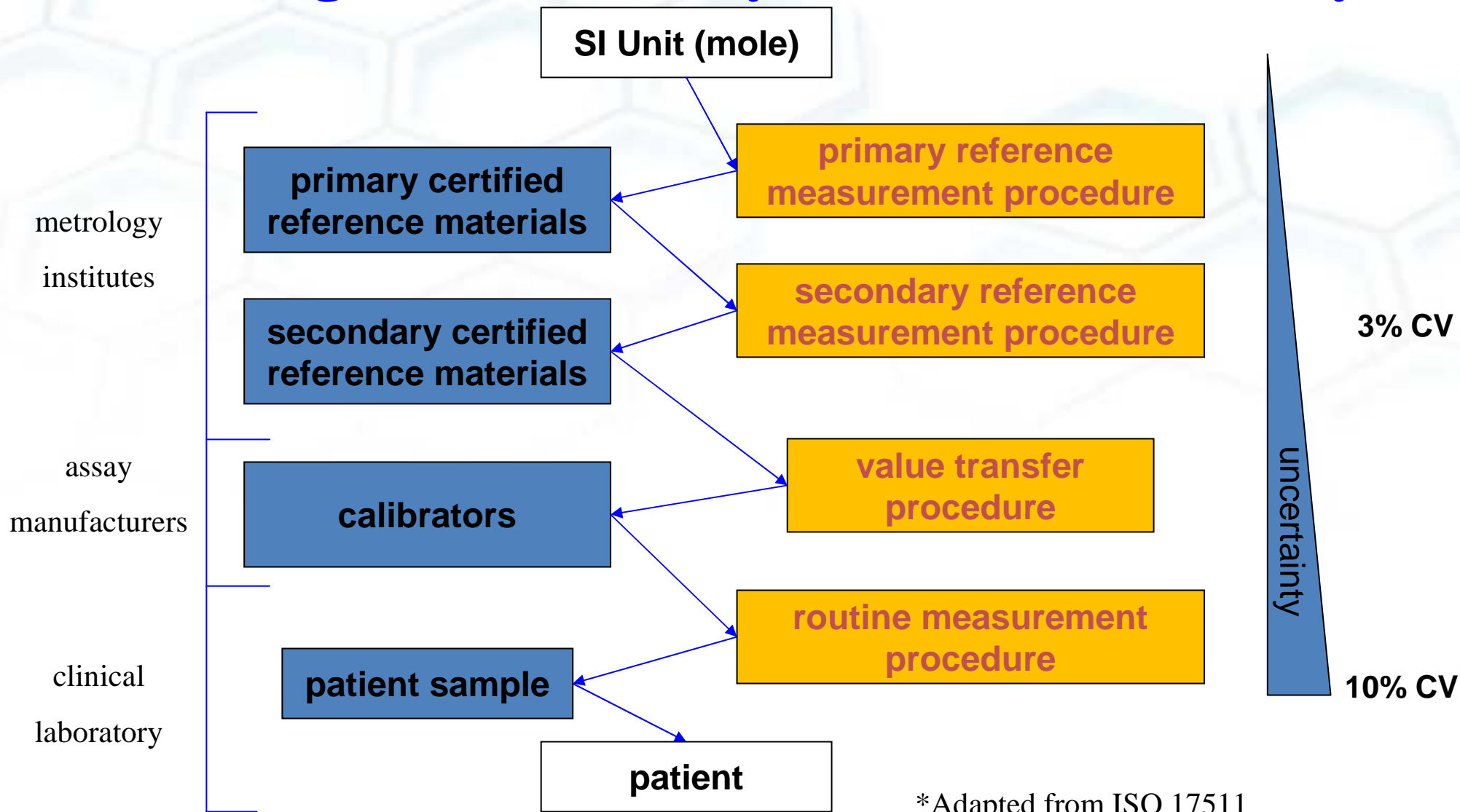
Measurement specificity:

- Defined by the antibodies used in the routine cTnI assay

Measurement uncertainty of a cTnI reference measurement procedure:

- The required measurement uncertainty of a cTnI reference measurement procedure is defined by the measurement uncertainty required in routine cTnI measurement
- Due to the propagation of error in measurement, the required measurement uncertainty of a cTnI reference measurement procedure needs to be lower than the measurement uncertainty of routine assays
- Conventionally, the goal in the development of a reference measurement procedure is to achieve a measurement uncertainty that is approximately 1/3 lower than the measurement uncertainty of routine assays

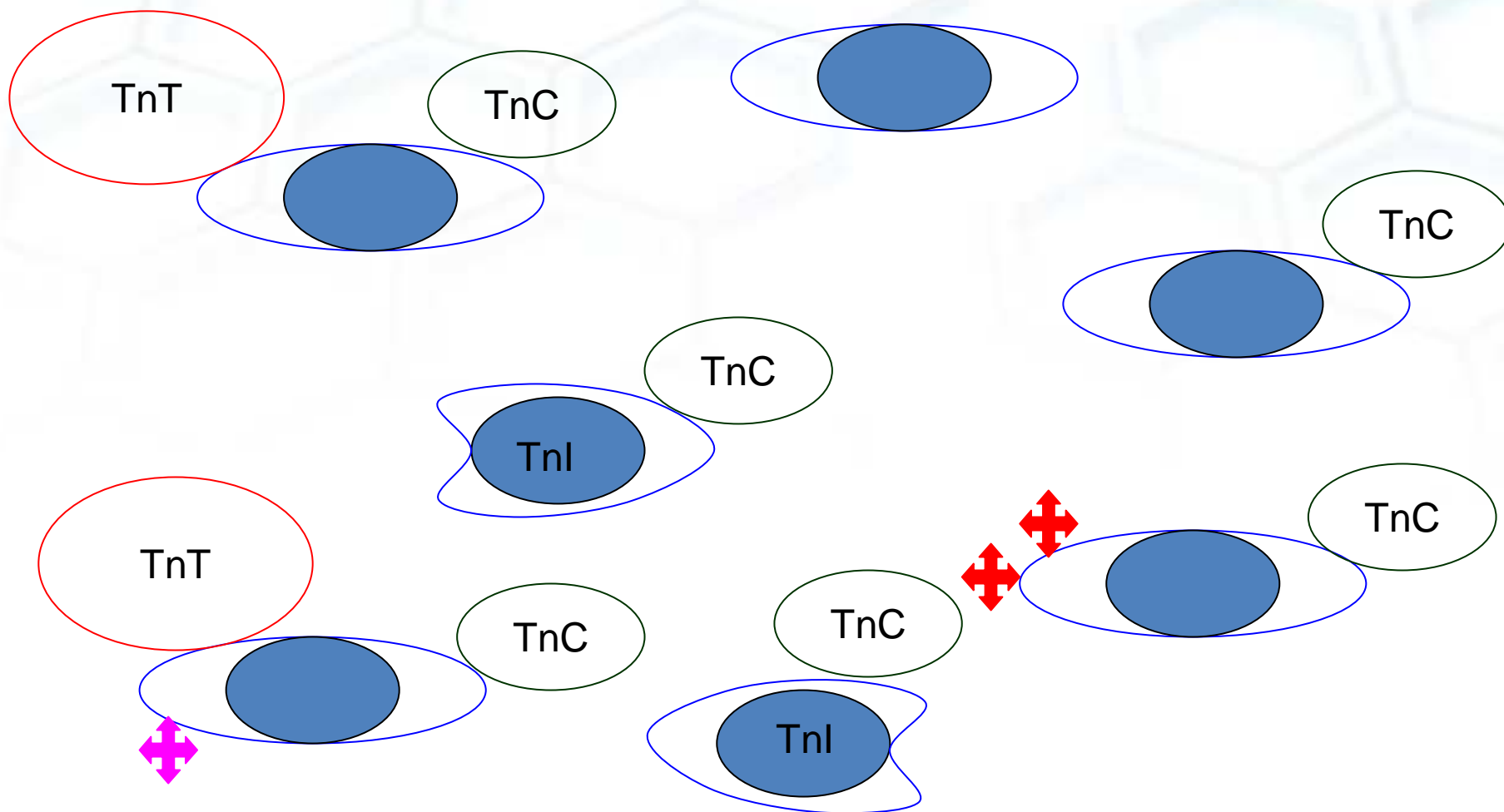
Metrological Traceability in Clinical Chemistry



Measurement specificity of a cTnI reference measurement procedure:

- The measurement specificity of a cTnI reference measurement procedure is defined by the measurement specificity of routine cTnI assays
- However, the measurement specificities of routine cTnI assays vary due to the variety of affinity reagents used in their design and the molecular heterogeneity of cTnI
- The challenge in the development of a reference measurement procedure for cTnI is how to bring order (a single measurement specificity) out of chaos (multiple measurement specificities and cTnI heterogeneity)

Defining a single specificity for cTnI measurement:



cTnI — Definition of the Measurand

IFCC Antibody Specificity Recommendation¹:

Antibodies used for the development of reliable cardiac troponin assays should preferably recognize epitopes that are located in the stable part of the molecule and are not affected by complex formation (such as ICT) and other in vivo modifications.

—Clin Chem Lab Med 2001, 39(2), 174-178

1	ADGSSDAAREPRPAPAP IRRSSNYRAYA <u>TEPHAKKSK ISASRKLQLKT</u>	50
51	<u>LLLQIAKQELEREAEERRGKGRALSTRCQPLELAGLGFAELQDLCRQLH</u>	100
101	<u>ARVDKVDEERYDIEAKVTKNITEIADLTQKIFDLRGKFKRPTLRRVRISA</u>	150
151	<u>DAMMQALLGARAKESLDLRAHLKQVKKEDTEKENREVGDWKRNIDALSGM</u>	200
201	EGRKKKFES	209

Measurement accuracy of a cTnI reference measurement procedure:

- Evaluating absolute accuracy of a reference measurement procedure is difficult as there needs to be something that represents a “true” value
- A true value is usually defined through the gravimetric preparation of calibrants from a primary certified reference material (NIST SRM 2921 for cTnI)
- Accuracy of a reference measurement procedure can be evaluated through recovery and spiking experiments using a primary certified reference material
- Accuracy of a reference measurement procedure can also be evaluated through comparison to another reference measurement procedure or other “higher-order” methods

Transferability of a cTnI reference measurement procedure:

- In order for a reference measurement procedure to be relevant, it must be able to be performed in multiple laboratories yielding statistically equivalent results
- Transferability of a reference measurement procedure requires:
 - Complete documentation of the method
 - Reagents used in the method be readily available
 - Independence to the type of instrumentation used
- The cTnI candidate reference measurement procedure is being evaluated through an interlaboratory study in the CCQM bioanalysis working group involving 12 laboratories in the US, UK, Germany, Belgium, Turkey, Japan, China, Korea, Thailand, Mexico, Argentina, and Brazil

Conclusions:

- The measurement specifications of a reference measurement procedure can only be defined through the evaluation of its intended use to certify reference materials
- The measurement precision required by a reference measurement procedure is defined by the precision of routine assays
- The specificity of a reference measurement procedure must match exactly the specificity of routine assays
- For cTnI measurement, variability in the specificities of routine assays can potentially be reduced by a redefinition of the measurand
- Accuracy of a reference measurement procedure can be evaluated through the use of suitable primary certified reference materials
- A reference measurement procedure must be transferable to other laboratories across time and space