

Hello and welcome to this JCTLM webinar on the Pillars of Standardization  
Elvar Theodorsson speaking

Standardization in general is a consensus process, e.g. between producers and consumers of a certain product to agree on specifications for the proper quality of the product from the perspectives of both parties.

Standardization in the metrology of chemistry is only partially a consensus process – it is fundamentally about identifying the most appropriate reference materials and reference measurement procedures to make sure that different measurement systems and laboratories around the globe measure the same concentrations in the same sample today and in the years to come. The measurement results should be traceable to a pure substance, a primary reference measurement standard and a primary reference measurement procedure. Thus, measurement results become equivalent and consistent in space and time.

The amount of substance and its unit the mole is crucial for standardization. To understand the concept amount of substance we need to understand that metrology of physics has been practiced for more than 100 years, whereas chemistry has been a member of the metrological community only since the early 1970-ies when the base quantity “amount of substance” and its unit “mole” was defined and accepted as one of the base quantities of the SI system of units. Metrology of chemistry inherited many fundamentals of metrology of physics.

Physical chemists were most influential in the current definition of the “amount of substance” as “the amount of substance of a system which contains as many elementary entities as there are atoms in 0.012 kilogram of carbon 12”. Expressed simply – the mole is the Avogadro number of molecules of the pure substance all lumped together. Analytical chemists repeatedly asked for revision of the definition of the “amount of substance” realizing that the effects of atoms, ions and molecules are induced by individual atoms, ions and molecules.

In the draft revised version 9 of the SI brochure to be published in 2018 the Avogadro number of molecules constant instead of the carbon 12, simply represents a conversion factor between the quantity amount of substance (with unit mole) and the quantity for counting entities (with unit one, symbol 1). The mole will be a proportionality constant like the Boltzmann constant  $k$ , which relates the average kinetic energy of gas molecules to the temperature of the gas. The Avogadro constant converts the macroscopic concept of the mole to the sub-microscopic reality of the molecules.

The mole – even with its current definition – is, however, a useful concept in expressing amounts of reactants and products in chemical reactions. It furthermore makes sense that corresponding concepts and terms have the same meaning in all fields of metrology.

Proper definition of the mole of a substance requires exact knowledge of the molecules being measured and their availability in a pure and homogenous form.

In case the molecules are not available in a pure and homogenous form, the concentration cannot be expressed in the molar concentration and SI unit mole/L. Mass concentration of e.g. mg/L, can however, be used for any substance and any calibrator. The SI system of units has also derived units for mass and volume for expressing mass concentrations.

The concept “measurand” is also crucial in the metrology of chemistry. It expresses the understanding that we, as a matter of fact, do not measure the substance/analyte/component of interest, but rather a physiochemical property, e.g. wavelength of light, elution volume on a chromatographic system or immunochemical reactivity which sufficiently characterizes the substance to measurement. The measurand is a quantity which we can measure whereas the substance/analyte/component is not.

Measuring means comparing

We use the measurement system to compare the quantity of the measurand measured in the reference material with a known concentration of the component with the quantity of the measurand measured in the sample where the concentration of the component is unknown. Using the ratio of the quantity values of the measurands in the reference material and in the unknown sample can be compared and the measurement result calculated through a measurement equation.

Knowledge of the uncertainty of the measurement result carried by the reference material together with knowledge of the uncertainty of the current measurement system used enables calculation of the uncertainty of the measurement result of the unknown sample including its uncertainty.

Traceability is a property of the measurement result and not a property of the reference material. We measure/quantify a measurand – a physiochemical property - in the reference material. It is this physiochemical property which is carried by the reference material and stated in its reference documentation including its uncertainty. We cannot compare reference materials directly – only through comparisons of the physiochemical properties they carry.

Let us exemplify this with the measurement of creatinine in plasma. We wish to measure creatinine, but actually measure one of its many possible physiochemical properties, the orange dye produced with the addition of picrate to the sample. This orange color can be measured by means of spectrophotometry. However, other molecules in the sample are also colored in the same orange color by picrate, which means that this measurand – the orange color detected by the spectrophotometer actually overestimates the concentration of creatinine in the plasma sample.

However, the use of another measurand of creatinine – a stereospecific and selective enzymatic reaction involving the creatinine molecule, results in a measurable physiochemical property which is more selective for creatinine.

So traceability is a property of the results of measuring physiochemical properties (measurands) corresponding to a component/analyte in the reference material. Thus traceability is carried by the measurement results.

**JCTLM** established the three pillars of traceability:

1. Reference measurement procedures (RMP)
2. Reference materials (RM) (includes commutability)
3. Network of Reference Measurement Laboratories (RELA studies)

**IFCC** described a fourth pillar:

4. Accuracy based grading of EQA/PT to ensure and maintain international reference systems
5. Universal reference intervals/Medical Decision Limits

Fifth pillar (Mauro Panteghini):

Several international and national organisations serve as sources of appropriate reference materials, reference measurement methods, reference measurement services including trueness based proficiency testing.

There is a very substantial difference between the conditions for physical and chemical measurements. The molecules we intend to measure in chemistry are usually mixed with numerous other molecules (sample matrix) in the sample which may influence the measurand and thereby the measurement result. Physical measurements do not usually destroy the measured sample and are seldom influenced by matrix effects.

Several other confounding factors are also at play in chemical measurements

Less than 10 % of all measurement methods in Laboratory medicine are standardized with standards traceable to SI.

Standardization using traceable reference materials is preferable to harmonization since it includes mechanisms for maintaining stability during extended periods of time whereas harmonization represents consensus processes primarily valid in the time when the harmonization process is performed.

## Supplementary material

### Keywords

Standardization, SI-system, amount of substance

### Learning objectives

1. Standardization in general vs standardization in the metrology of chemistry
2. Understanding the concept of “amount of substance” and its unit the mole
3. Understand the concept “measurand”
4. The meaning of comparisons in measurement
5. Traceability as a property of the measurement result

### Multiple choice questions

What is “mole”

- a) The name of the base quantity in the metrology of chemistry
- b) The symbol for the amount of substance
- c) The number of molecules in a defined amount of pure substance
- d) The number of molecules in a volume of a solution

Answer(s): b)

Why is the availability of pure and homogenous reference materials crucial in the metrology of chemistry?

- a) They can be weighed (gravimetry) accurately and the number of mol estimated
- b) It avoids counting in variants of the molecules, e.g. posttranslational processing
- c) Other molecules are confounders in this context
- d) Purity is fundamental to the ability to reproduce the material over extended periods of time

Answer(s): a), b), c), d)

The difference between standardisation in general and standardisation in the metrology is

- a) The consensus processes
- b) The use of reference materials and reference measurement procedures
- c) Traceability
- d)

3) 5 multiple choice questions to assess knowledge gained (see example attached)