International reference system for somatic cell counting in milk - A world wide challenge

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Abstract

In a globalizing world analytical results play a major role in free and fair trade. However, worldwide equivalence of analytical results can not be ensured by "only" producing standardized analytical methods. For some parameters standardized reference materials are lacking and the reference method shows limited performance. It is there that a reference system should serve to optimally safeguard equivalence.

In this paper it is explained what a reference system is and why Somatic Cell Counting (SCC) was chosen as a first example for implementation. An outline of a reference system for SCC is drawn and the time plan and the next actions of the joint IDF / ICAR project group are described.

Keywords: Reference system, raw milk analysis, somatic cell count, SCC, relation between reference and routine method, joint IDF / ICAR project group.

1.0 What is a reference system?

A reference system is a systematically developed anchoring system that is fed by different types of information from various sources in a laboratory network structure:

Reference method results.

Routine method results.

Results from proficiency testing.

Joint recognition by regulatory bodies, competent authorities and other stakeholders is essential for an effective functioning. A more extensive explanation on the background and aims as well as on the elements of reference systems was published in the Bulletin of the IDF 427/2008.

2.0 Why somatic cell counting as a first example?

SCC is one of the most frequently performed tests worldwide, estimated at over 500.000.000 tests/year.

SCC – as an indicator for udder health status – is relevant in food legislation, payment of raw milk and also has a major impact on farm management and breeding programs.

SCC data are obtained almost exclusively by automated high-throughput fluoro-opto-electronic counting instruments, which are calibrated and controlled with more or less defined milk samples giving a "reference level" for counting. This reference level derives in many cases from the application of the reference method, a direct microscopic cell counting according to ISO 13366-1|IDF 148-1.

2.1 Somatic cell counting as a typical problem

Traditional calibration schemes are especially problematic with SCC, because several preconditions are only poorly met. It is necessary to repeat the reference method in more than one lab to arrive at an acceptable precision and accuracy of resulting reference values. Results from a collaborative study carried

out in October 2005 show that repeatability r and reproducibility R is rather limited (see also table below). The recently revised ISO 13366-1|IDF 148-1 on the microscopic reference method provides a better description on "what to count and not to count". Still, the reference method is tedious and cumbersome and requires experience and frequent execution in order to safeguard adequate competence of the analyst and a proper counting of the "analyte".

Certified reference material ("golden standard") is not available. Secondary reference materials have problems with shelf life and batch homogeneity during storage. Different matrices and cell types are used for the preparation of these secondary reference materials.

In routinely exercised somatic cell counting, well functioning automated fluoro-opto-electronic methods are used. However, the target analyte of these actual routine methods is not commonly accepted as "reference" basis.

Therefore, a true common basis for the calibration of routine instruments is in fact lacking. As a consequence, several routine laboratories have put their own 'reference system' in place in order to anchor their counting level.

For all these reasons, SCC serves as illustration of a typical problem and makes it into a true candidate parameter for implementing a world wide adopted reference system approach.

ISO 13366|IDF 148, part 1 vs. part 2 (all values in '000/ml)

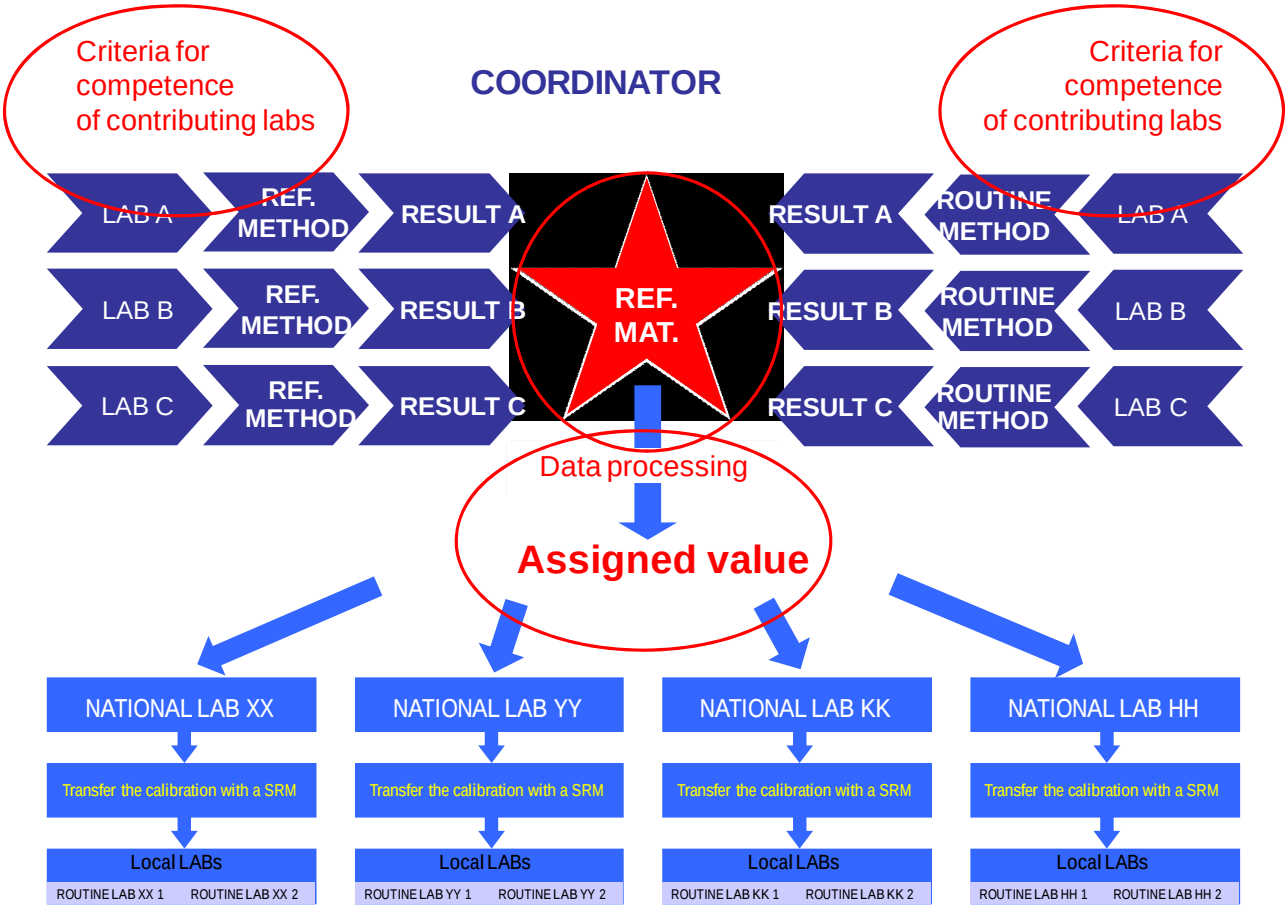
	Mean	s_r	sR	r	R
Reference	245	38 41	107		114
	679	69 79	192		218
Routine	245	13	20	36	57
	679	21 40		59	112

3.0 Outline of reference system for somatic cell counting

A reference system is regarded as a tool to provide a commonly acknowledged reference level for a given analytical parameter. It is characterized as a systematically developed anchoring system that is fed by different types of information from various sources, i.e. from reference materials, reference method analysis, routine method results and proficiency testing in a laboratory network structure. Joint recognition by regulatory bodies, competent authorities and other stakeholders is essential for an effective functioning.

A joint project group of IDF and ICAR has recently outlined a reference system for SCC and aims for its implementation during the next years. The system will be fed by routine and reference laboratories to characterize one or more (secondary) reference materials and systematically assign a "true" value to each material. This "assigned value" will represent the anchor level to which local routine laboratories can relate to. A system, well structured and anchored means avoiding fluctuations between different batches of RMs and subsequent calibrations.

The following scheme shows the principle.



4.0 What has the project achieved yet ?

In the joint IDF / ICAR project group now 4 continents and 16 countries are represented.

A strategic aspect is to communicate the aim of the project in the right way. This is achieved through meetings, by publishing papers, by publication of a newsletter and creating visibility on the ICAR and IDF websites. The communication will be oriented towards both the analytical stakeholders (labs, RM providers) as well as others (animal health bodies, authorities).

Questionnaires for reference materials providers and routine laboratories bring information on how available reference materials produced in several continents and more than 15 countries are used. This information will be useful to draft guidelines for reference materials and to draw a picture of the interlinkages between the different existing local analytical systems.

The first calculation models for assessing both proficiency testing schemes and the performance therein of laboratories involved with the assignment of reference values are under development within a group of statistical experts.

5.0 Next actions

The project group has identified the important parts to come to a reference system and is working out the details of a pilot model, which is to be evaluated in practise.

Crucial parts of this pilot model will be (amongst others):

A suitable reference material (representative and stable).

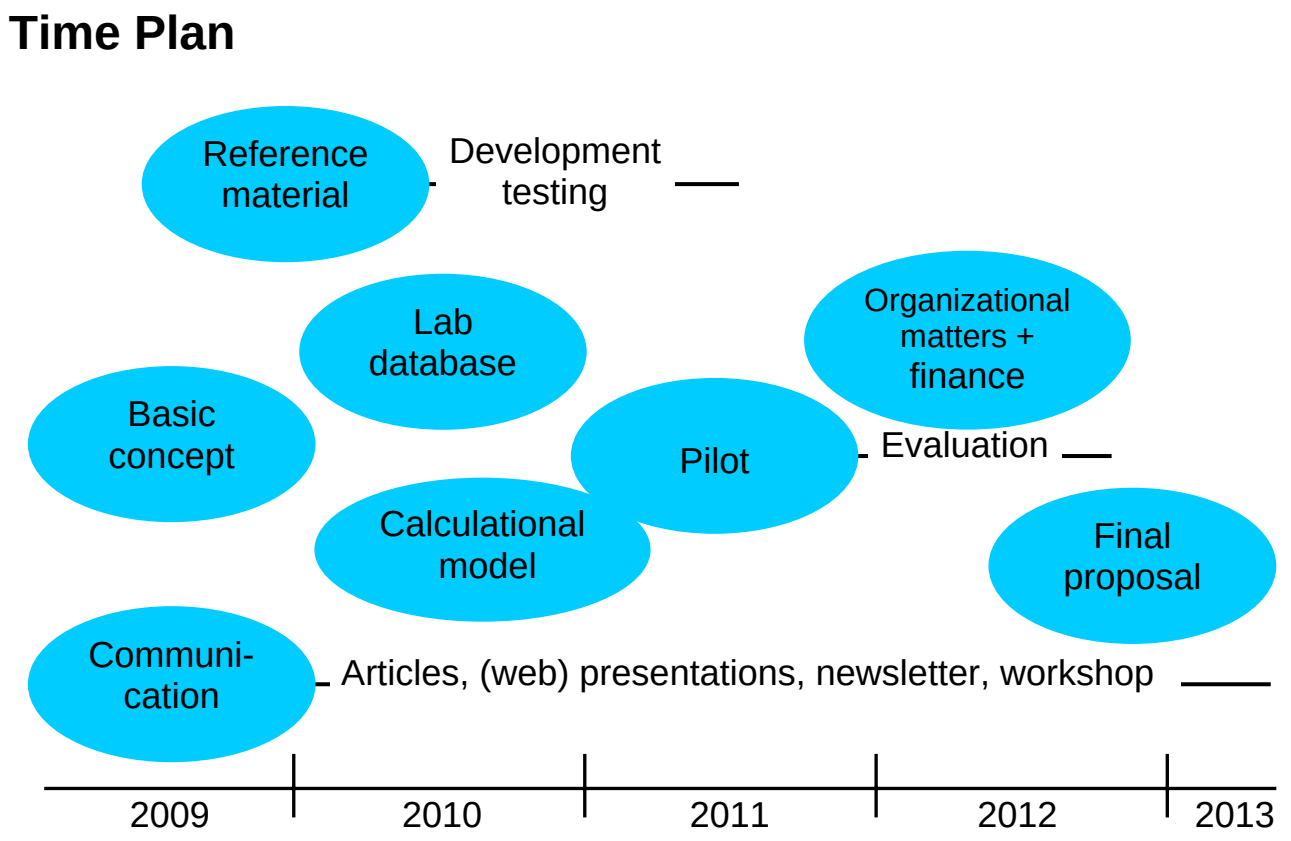
A laboratory database with a system for competence scoring.

A calculation model for determining optimized assigned values.

The project group follows a bottom– up approach. This means to collect and make use of existing and functioning local structures.

Of course some organizational matters and financial issues have to be addressed in the near future, too.

6.0 Time plan



7.0 Conclusions

Somatic cell count in milk is an excellent parameter to explore the feasibility of a reference system approach. It is a very relevant parameter in food legislation, in farm management and in animal breeding and the reference method has distinct drawbacks.

The world wide challenge of this innovative analytical approach is to create mutual trust between the actors involved and to share useful data and experience which are obtained in daily analytical life.

A collaborative atmosphere at the national and international levels will help – and will be needed – to complete the puzzle of this sophisticated but also more robust analytical approach that is focussed at obtaining better analytical equivalence.

A robust reference system should produce results, which are valid in a three-dimensional scale: worldwide, over time and between different methods. ICAR and IDF as dedicated international organisations have taken up the glove to develop and implement this innovative and valuable approach.