Independent validation and certification of analytical methods

V.A. Tzeneva & H. van den Bijgaart

Qlip B.V., P.O. Box 119, 7200 AC, Zutphen, The Netherlands
tzeneva@qlip.nl (Corresponding Author)

Summary

Routine testing of milk and milk products on quality and compositional parameters is often performed with alternative methods. An independent validation of an alternative method is key to demonstrate that the method is suitable for application in testing for regulatory purposes, with milk payment and/or with milk recording. Independent validation is an assessment of the performance of the method by an organisation not involved in its development. During the validation process it is established whether the alternative method complies with beforehand stated requirements. Subsequent certification comes with a tangible declaration on the suitability of the analytical method for the intended purpose. It serves to find adoption of the method and its results with laboratories, public authorities, food industry and other relevant stakeholders.

Keywords: milk, analytical methods, validation, certification, independent

Introduction

Milk composition and quality are important parameters in global dairy production. In many milk producing countries it is required that laboratory methods for milk testing comply with criteria published in international documents, such as ISO|IDF international standards, AOAC methods, national or regional regulations, etc.

Routine testing of the compositional parameters in raw milk, such as fat, protein, lactose and urea content, and other parameters, such as total bacterial count and somatic cell count, is in many countries performed with alternative analytical methods. The use of an alternative method, often high-throughput instrumental methods, is acceptable when the method is fully validated by an independent party, including a comparison against the reference method. Since independent validation is a critical factor for the acceptance of an alternative method, the validation criteria are described in official documents, following the requirements published in international standards, e.g. ISO|IDF standards. The validation process and results can be made subject to evaluation by a certification organisation, providing a formal statement on the analytical performance of the alternative method and its fitness for purpose.

Independent validation

Independent validation is a process of establishment of the performance characteristics of a method and provision of objective evidence that the performance requirements for a specified intended use are fulfilled (ISO 16140-1:2016). Independency can be assured via conducting the validation activities by a separate organisation which did not contribute to the development of the analytical method. A major advantage of an independent validation is that the group performing the evaluation is unbiased and emotionally or economically not involved in the method (White paper, 2001). In complex validations it may be difficult for the manufacturer to objectively evaluate the obtained validation results. An independent third party might easier identify issues that have escaped the attention of the developer.

Often the process of an independent validation is monitored by a certification organisation, best in conformity with internationally accepted and documented protocols. A certification organisation relies usually on several entities, e.g. a board, a general secretariat and a network of sub-contractors: laboratories, reviewers and auditors. The certification organisation works with appointed expert laboratories having traceable competence, which prepare and execute the validation, as well as analyse, report and often evaluate the results. The validation procedure and results are reviewed by an (international) technical committee of experts in the field. A certification board closely follows and monitors the whole process and can declare its confidence of the findings by issuing a certificate (Figure 1).

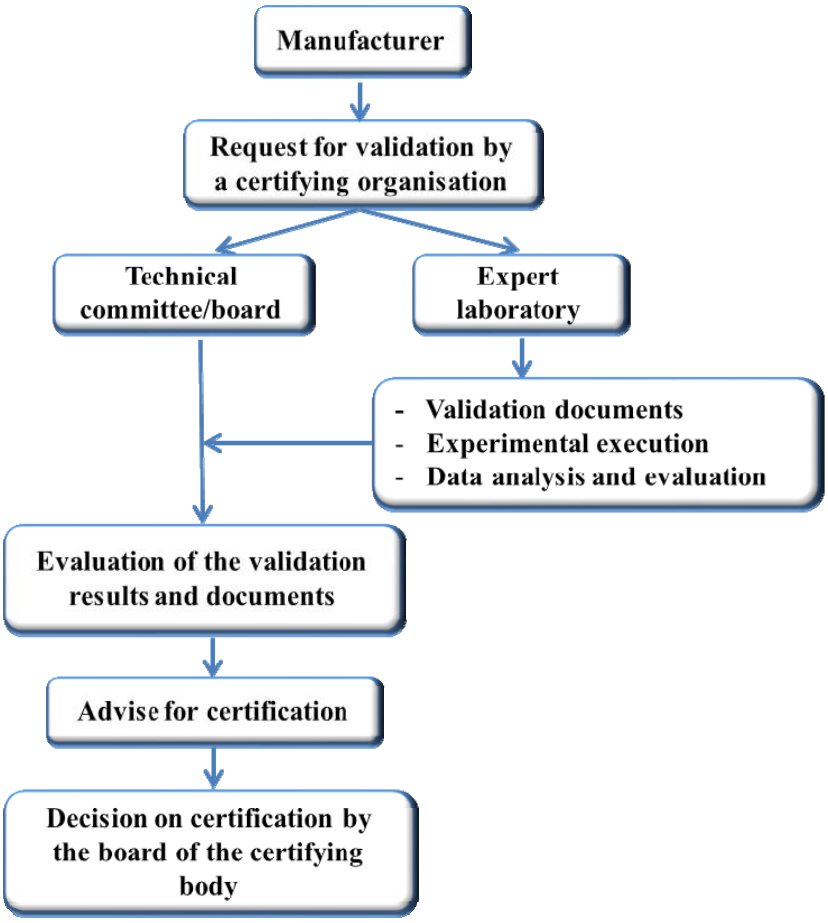

Figure 1. Schematic presentation of the general principle of a certification process (examples MicroVal, AFNOR, NordVal and ICAR).

The validation of an alternative method is assigned to an expert laboratory. The process generally consists of two stages:

- an in-house validation or method comparison study, and
- a method confirmation study or an inter-laboratory study.

The method comparison study demonstrates the performance of the method under validation and checks compliance with the stated acceptability limits. Different performance characteristics are evaluated for qualitative and quantitative alternative methods. For example, the relevant characteristics for qualitative microbiological methods are sensitivity, detection limit, inclusivity and exclusivity. For the evaluation of quantitative instrumental methods these are stability, linearity, repeatability, carry-over and limits of quantification. The estimation of the accuracy of the alternative method against the reference method is done with representative samples, measured with both methods. Moreover, potentially influencing factors affecting the relationship between alternative method results and reference method results are examined.

The precision characteristics of the alternative method, when executed at different user laboratories, are demonstrated by either a method confirmation study or an inter-laboratory study. By a method confirmation study the alternative method is operating under routine conditions for several weeks at several laboratories. Results are obtained with representative routine samples as well as with pilot samples and, if relevant, other check samples. The performance of the method in terms of repeatability, reproducibility and stability is evaluated in each laboratory separately and overall. During this part of the validation study the method is also evaluated for general convenience aspects such as speed, consumables, user-friendliness, security and robustness as well.

When the alternative method is already in routine use its performance could be demonstrated by an inter-laboratory study. Sample sets are prepared by a organising laboratory, usually the expert laboratory, and measured at several laboratories where the alternative and (when required) the reference method are operational. The performance characteristics, e.g. repeatability and reproducibility of the alternative method as well as the agreement of the results with results obtained with the reference method are evaluated by the expert laboratory.

The expert laboratory collects and analyses the results of the method comparison and method confirmation/inter-laboratory studies and prepares a validation report. The validation report is evaluated by a committee of experts appointed by the certifying body. With a positive advice from the expert committee a certificate for performance may be granted for the alternative method. The certificate demonstrates to the end users that the alternative method has been thoroughly tested using an approved and standardised procedure. It means that the method can be used confidently and with the knowledge that the results of that method will be accepted by the national and international authorities (Zegers, 2012).

Examples of independent validations for milk testing

For regulatory purposes and milk payment total bacterial count and somatic cell count are common parameters. National and international authorities in many geographies require proof on proper functioning of the applied methods through independent validation. An example is in the validation of alternative methods for the enumeration for total bacteria and somatic cells as required by EU Regulations 2074/2005 and 1664/2006. The test procedure for these validations are described in two documents issued by the European Reference Laboratory for Milk and Milk Products (EURL MMP document 2011; EURL MMP document 2013), following the relevant ISO standards (ISO 8196-3|IDF 128:2009; ISO 13366-2|IDF 148-2:2006; ISO 16140-2:2016; ISO 16297|IDF 161:2013). Recently several instruments for total bacterial count and somatic cell count have been granted with MicroVal certificates (www.microval.org).

For the purpose of milk recording, the ICAR Certificate of Quality program requests independent validation of alternative methods for measurements of milk compositional parameters. The ICAR certification process follows the ISO protocol for validation of alternative methods (ISO 8196-3|IDF 128:2009) and the obtained results should comply with requirements in the relevant ISO standards (ISO 9622|IDF 141:2013, ISO 13366-2|IDF 148-2:2006).

Conclusion

The globalising milk market requires confidence in an uniform, reliable and traceable way of testing of milk characteristics all over the world. Independent validation of analytical methods demonstrates and assures that alternative methods are fit for purpose. Subsequent certification provides tangible proof of adequate performance. It paves the way for acceptance of a method and the results by public authorities, food industry, laboratories and end users.

List of References

Commission Regulation (EC) No 1664/2006 of 6 November 2006 amending Regulation (EC) No 2074/2005 as regards implementing measures for certain products. Off. J. of Eur. Union 2006 L320/13.

Commission Decision 2002/657/EC of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results. Off. J. Eur. Union 2002 L221:8-36.

Commission Regulation (EC) No 2074/2005 of 5 December 2005 laying down implementing measures for certain products. Off. J. of Eur. Union 2005 L338/27.

EURL MMP document - Criteria for the validation of instrumental (epifluorescent) methods for the enumeration of somatic cells in raw cow's milk, version 2, 21/01/2013.

EURL MMP document - Validation criteria of instrumental methods for enumeration of total flora in raw milk, version 2, 21 December 2011.

ISO 8196-3|IDF 128-3:2009 Milk - Definition and evaluation of the overall accuracy of alternative methods of milk analysis - Part 3: Protocol for the evaluation and validation of alternative quantitative methods of milk analysis.

ISO 9622|IDF 141:2013 Milk and liquid milk products - Guidelines for the application of mid-infrared spectrometry.

ISO 13366-2|IDF 148-1:2006 Milk - Enumeration of somatic cells - Part 2: Guidance on the operation of fluoro-optoelectronic counters.

ISO 16140-1:2016 Microbiology of the food chain - Method validation - Part 1: Vocabulary.

ISO 16140-2:2016 Microbiology of the food chain -- Method validation -- Part 2: Protocol for the validation of alternative (proprietary) methods against a reference method.

ISO 16297|IDF 161:2013 Milk – Bacterial count – Protocol for the evaluation of alternative methods.

MRL Commission Regulation (EU)No. 37/2010 of 22 December 2009: On pharmacologically active substances and their classification regarding maximum residue limits in foodstuffs of animal origin. Off. J. Eur. Union 2010, L15:1-72.

White paper (2001) Independent verification and validation. Comprehensive Consulting Solutions, Inc. - "Business savvy. IT Smart".

Zegers, N. (2012) Rapid microbiological methods for food safety testing. International Food Hygiene, Volume 22, Number 7.