Reference and calibration system for routine milk testing

Advantages & Disadvantages - Choice criteria

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Meeting of ICAR Reference Laboratory Network, 6 June 2006 - ICAR Session Kuopio 2006

The error of measurement includes:

⇒ the precision error of the routine method:
  - repeatability & within day reproducibility (short term stability)
  - under control
  - cannot be avoided or reduced in routine testing

⇒ the precision error of the reference method:
  - repeatability: negligible as reduced by sample and replicate numbers
  - reproducibility: Possible systematic error of the lab allowed by the method and normally distributed according to $s_R$

⇒ the error of calibration:
  - statistical error of adjustment: can be improved with appropriate samples made to maximize the correlation coefficient
  - error of sample representativeness resulting of matrix effects

Introduction

Reference and calibration systems

⇒ refer to a general analytical system chosen for a prior defined purpose: i.e. milk recording
⇒ part of a strategy to achieve the objectives of organized users, thus resulting from a collective choice.

Objectives

⇒ to optimise the accuracy of results (or lower the related uncertainty)
with providing sufficient confidence in the quality of results
and with an acceptable balance between quality & cost

Analytical systems available for milk recording

Reference

Centralised calibration:
- Low cost (purchase RM samples; no reference method by labs)
- Matrix effects (to be limited)
- Competence for testing

No/negligible error $\rightarrow$ estimating true values:
$s_{\rightarrow d}^2 \approx s_L^2 / p$ with: $p$ labs

Possible systematic bias $\bar{3}$ of the lab from the standard precision of the method:
$s_{\bar{3}}^2 = s_L^2 + s_{R}^2$

Local calibration:
- High cost (sample collect; reference analysis)
- Additional competence
- No bias related to matrix effects (representativeness)

Nonnegligible error $\bar{3}$ in estimating true values:
$s_{\bar{3}}^2 = s_L^2 / p$ with: $p$ labs

Centralised calibration:
- Low cost (purchase RM samples; no reference method by labs)
- Matrix effects (to be limited)
- Competence for testing

Mid infra red spectroscopy and matrix effects on classical wavelengths

Elements for choice

Choice of appropriate methods

Wavelengths: minimise bias laboratory spreading within the region or country, thus no or only little influence of the milk matrix variation

Ex: Fat A < Fat B < Fat by FT-MIR Full Spectrum
CP 6.5 µm < CP by FT-MIR Full Spectrum
TP 6.5 µm < TP by FT-MIR Full Spectrum
CP’ by FT-MIR Full Spectrum < TP by FT-MIR Full Spectrum

Expression units: routine methods and reference methods to take into account same components in the measurement principle

Ex: Mass of component: Fat A < Fat B
NPN: Crude Protein 6.5 µm < True Protein 6.5 µm

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1°) Experimental evaluation

Over a one year period and on the whole region:

1- Analyse: by a single laboratory representative test samples of different collect areas (labs) by the routine methods with unchanged calibration and the reference methods.

2- Evaluation: of ranges of variation of theoretical calibration bias between labs and between periods

3- Decision: by reference to maximum acceptable range of calibration bias (fit for purpose).


Geographical and seasonal effect on mid infrared fat and protein determination in France:

Samples of 8 regions of France with various geographical/seasonal situations analysed at the same time in reference and mid infrared and calibration (same instrument) optimised on the whole of data for each season to measure local effects (ANOVA).

<table>
<thead>
<tr>
<th>Measurand (g/100g)</th>
<th>Season</th>
<th>Range of</th>
<th>d mean</th>
<th>sd (g/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat 5.7 µm</td>
<td>Nov. 1981</td>
<td>0.102</td>
<td>0.077</td>
<td>0.072</td>
</tr>
<tr>
<td></td>
<td>Feb. 1984</td>
<td>0.042</td>
<td>0.043</td>
<td>0.042</td>
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<tr>
<td></td>
<td>June 1985</td>
<td>0.086</td>
<td>0.051</td>
<td>0.044</td>
</tr>
<tr>
<td>Fat 3.5 µm</td>
<td>Nov. 1981</td>
<td>0.063</td>
<td>0.052</td>
<td>0.047</td>
</tr>
<tr>
<td></td>
<td>Feb. 1984</td>
<td>0.017</td>
<td>0.027</td>
<td>0.027</td>
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<tr>
<td></td>
<td>June 1985</td>
<td>0.031</td>
<td>0.031</td>
<td>0.030</td>
</tr>
<tr>
<td>True protein 6.5 µm</td>
<td>Nov. 1981</td>
<td>0.018</td>
<td>0.023</td>
<td>0.023</td>
</tr>
<tr>
<td></td>
<td>Feb. 1984</td>
<td>0.019</td>
<td>0.028</td>
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<tr>
<td></td>
<td>June 1985</td>
<td>0.022</td>
<td>0.019</td>
<td>0.018</td>
</tr>
</tbody>
</table>

Units: g/100g

O. Lera, Le Lait, 69, 1989
Example: BCR MIR European Programme 1991
15 labs (countries) x 2 bulk milks x 8 periods of 1 year

Matrix effects:
- MW / Fat A
- UFA / Fat B
- NPN / CP
- Citrate / TP

Elements of choice for a centralised calibration system

Comparison of laboratory bias distributions in Proficiency Testing
- In centralised calibration:
  - laboratory bias (1) = bias with the reference method (2) + calibration bias (3)
- MIR PTs:
  - measures the distribution of laboratory biases (1) = (2) + (3)
- PTs on reference methods:
  - measures the distribution of biases with the reference method (2)

- Comparing the standard deviations (or ranges) of biases between labs:
  - Routine method SD ≤ reference method SD = improvement or equivalence (4)
  - Routine method SD > reference method SD = discrepancy of uncertainty (5)

(4) => Centralised calibration system applicable.
(5) => It is to users (milk recording) to consider whether or not the extent of discrepancy is acceptable for the intended use.
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Example: Comparison of laboratory bias distributions

Example: Comparison of laboratory bias distributions

CRUDE PROTEIN in MILK by KJELDAHL

TRUENESS OF LABORATORIES

(Distribution mean biases (lab-ref.)

0 2 4 6 8

-0.12 -0.08 -0.04 0.00 0.04 0.08 0.12

Classes of d

Absolute frequency

(units: g CP / 100 g of milk)

ICAR Interlaboratory Proficiency Study - March 2006
International Proficiency Testing - March 2006
ICAR Reference Laboratory Network

TRUE PROTEIN in MILK by MIR SPECTROSCOPY

TRUENESS OF LABORATORIES

(Distribution mean biases (lab-ref.)

0 2 4 6

-1.2 -0.8 -0.4 0.0 0.4 0.8 1.2

Classes of d

Absolute frequency

(units: g TP / litre milk)

ESSAI D’APTITUDE CECALAIT - FEBVRIER 2006
National Proficiency Testing - February 2006

Routine laboratories

METHODS

CRUDE PROTEIN in MILK by KJELDAHL

TRUENESS OF LABORATORIES

(Distribution mean biases (lab-ref.)

0 6 12 18 24

-1.20 -0.80 -0.40 0.00 0.40 0.80 1.20

Classes of d

Absolute frequency

(units: g CP / kg milk)

ESSAI D’APTITUDE CECALAIT - MARIS 2006
International Proficiency Testing - March 2006
All types of laboratories (routine & reference)

METHODS

CRUDE PROTEIN in MILK by MIR SPECTROSCOPY

TRUENESS OF LABORATORIES

(Distribution mean biases (lab-ref.)

0 2 4 6

-0.12 -0.08 -0.04 0.00 0.04 0.08 0.12

Classes of d

Absolute frequency

(units: g CP / 100 g milk)

International Proficiency Testing - February 2006
Routine laboratories

METHODS

Conclusion

• Where applicable, centralised systems for reference and calibration are:
  – more convenient tools for laboratories
  – more securing systems for users

• Centralised calibration requires:
  – either routine methods insensitive to matrix effects
  – or to concern areas with negligible variation in matrix composition.

• Otherwise its applicability relates to:
  – the loss of precision accepted compared to the advantages of a centralisation of the reference.
  – the uncertainty of analytical results needed by users.

Conclusion on appropriateness

• Tools for the application of centralised calibration systems already exit and are published in ICAR Sessions proceedings:
  – Appropriate method for calibration sample preparation (RMs)
  – Means for mid (chemicals) and long term (deep freezing) preservation
  – Structure for reference values checking or determination (ICAR Ref Lab network)

• Centralised calibration can be also an answer to the question of checking/fitting calibration of in farm analytical devices...

Conclusion

Thank You for your attention!