Summary of the MicroVal Validation of the SomaScope LFC and SomaScope Smart (Delta Instruments) against the EURL MMP Criteria for Enumeration of Somatic Cells in Raw Cow's Milk

1. Principle of the alternative method

The SomaScope is a fully automated flow cytometer for the rapid enumeration of somatic cells in raw milk. The working principle of the instrument is based on colouring the somatic cells with the fluorescent dye DAPI (4', 6-diamidine-2'-phenyl indole dihydrochloride), after which the cells are counted electronically.

In the flow cytometer, the mixture of milk and staining solution is surrounded by a sheath liquid and passed through a flow cell. In the flow cell, the stained somatic cells are exposed to light of a specific wavelength (UV) and the cells are individually illuminated. The activated fluorescent molecules emit light of a characteristic colour, DAPI emits in the yellow. For each single cell the fluorescence light intensity is measured and the concentration of cells per mL is determined. The design of the flow cell must ensure that single cells are separately counted.

Between each sample the flow system is thoroughly cleaned to reduce the carry-over to a minimum as well as the risk of build-up and clogging inside the analyser.

This document is a summary of the results of the method comparison study and the interlaboratory study of the SomaScope LFC and Smart (Delta Instruments) against the criteria in the EURL MMP document from January 2013. The MicroVal validation report presents the full results of the validation.

2. Scope

Raw cow's milk

3. Results and conclusions

3.1. Method comparison study

SomaScope LFC performance characteristics determined according to ISO 8196-3|IDF 128-3 and ISO 13366-2|IDF 148-2 are:

- Repeatability (*r*) per cell count level:
  - Low (ca. 108.10^3 cells/mL) 14 %
  - Medium (ca. 487.10^3 cells/mL) 8 %
  - High (ca. $1\,426.10^3$ cells/mL) 5 %
- Carry-over per cell count level:
  - Low (ca. 500.10^3 cells/mL) $C_{H/L}$ = 1 %

    $C_{L/H}$ = 1 %

- Medium (ca. 1 000.10^3 cells/mL) $C_{H/L}$ = 1 %
  $C_{L/H}$ = 1 %
- High (ca. 3 000.10^3 cells/mL) $C_{H/L}$ = 1 %
  $C_{L/H}$ = 2 %

- Linearity (r_C) working range (up to 2 000.10^3 cells/mL) 1,7 %
- Linearity (r_C) (up to 4 500.10^3 cells/mL) 1,98 %
- Lower limit of quantification (LL_Q): 19.10^3 cells/mL

Remark: The lower limit of quantification is calculated on the results obtained from a semi skimmed UHT milk.

- Upper limit of quantification (UL_Q): 4 500.10^3 cells/mL
- Intra-laboratory reproducibility ($R_{intra\text{-}lab}$) per cell count level:
  - Low (ca. 108.10^3 cells/mL) 16 %
  - Medium (ca. 487.10^3 cells/mL) 9 %
  - High (ca. 1 426.10^3 cells/mL) 5 %
- SomaScope LFC functions stable through the working day
- Milk fat content up to 6 % does not markedly affected the somatic cell count results. With cow's milk samples with fat content higher than 6 % a slight overestimation of the results may be observed.
- High protein content of the milk does not influence the somatic cell count results.
- The overall accuracy standard deviation $s_{y,x}$ is:
  - Individual cow milk samples 3 %
  - Herd bulk cow milk samples 2 %

3.2. Interlaboratory study

The interlaboratory study was performed according to ISO 5725-2 (2) and IDF Bulletin 453/2012. The overall precision characteristics of SomaScope LFC and Smart are:

- An interlaboratory standard deviation of repeatability s_r = 17.10^3 cells/mL = 3 % *(indicative limit according to ISO 13366-2|IDF 148-2 = 4 %)*
- A between-laboratory standard deviation s_L = 28.10^3 cells/mL = 4 %.
- An interlaboratory standard deviation of reproducibility s_R = 33.10^3 cells/mL = 5 % *(indicative limit according to ISO 13366-2|IDF 148-2 = 7 %)*

4. Final conclusion validation study

The Method Comparison Study and the Interlaboratory Study show that the alternative method results obtained with SomaScope LFC and Smart (Delta Instruments) comply with all criteria in the EURL MMP document, ISO 8196-3|IDF 128-3 and ISO 13366-2|IDF 148-2.