ARTICLE

EVALUATION OF THE BENTLEY FCMTM SOMATIC CELL COUNTER ON COW'S MILK

The Somacount FCM, manufactured and distributed by Bentley Instruments, is an apparatus for the enumeration of somatic cells in milk using a fluoro-opto-electronic method: firstly, the test sample is mixed with a dye (buffered detergent solution of ethydium bromide) in order to disperse fat globules and to stain the nuclei of the somatic cells. An aliquot is then injected into a laminar flow carrier fluid. The stained cells are separated by the flow and exposed to a laser beam. The fluorescent light emitted by the somatic cells is then detected and amplified by a photomultiplier tube. Intensities above a specific threshold are counted. This light intensity is then converted into a cell concentration using a calibration equation.

The instrument is composed of two separate counting units which work alternately (Blue/red channel). The signal is processed by a computer connected to the apparatus.

The tests:

The evaluation tests were carried out in Actilait-Cecalait's physico-chemistry laboratory (reference and instrumental analyses) from December 2009 to February 2010. The repeatability and the accuracy were evaluated.

The criteria for evaluating the estimated parameters were taken from ISO 13366-2/IDF 148: 2007, or from the CNIEL/IE handbook concerning the use of somatic cell counters within the context of milk payment and milk control in France.

The following instrumental parameters were used:

- Rate: 500 samples / hour;
- No contamination adjustment;
- Use in combined mode with the Bentley FTS infrared analyser (draining assistance).

1- Samples

The tests were carried on 100 milk samples from 4 farms in Jura. Bronopol was added to the samples to give a final concentration of 0.02%.

2- Repeatability

2.1- Procedure

The repeatability of the instrument was evaluated using all the milk samples. The quantitative determination was carried out in automatic analysis mode, in duplicate for each set of 10 samples according to the following sequence: (Set 1 rep 1 -Set 1 rep 2 -Set 2 rep 1 -Set 2 rep $2 \dots$ Set n rep 1 -Set n rep 2). A control milk was analysed every 30 samples to check the stability of the analyser.

2.2- Results

The results obtained are summarised in the table below.

	n	Min	Max	Μ	Sx	Sr	Sr (%)	r
RED CHANNEL	112	4	1225	105	175	7,1	6,7	20
BLUE CHANNEL	112	4	1280	111	180	5,9	5,3	16

Table 1: FCM repeatability criteria on cow's milk samples

n: number of results; min and max: minimum and maximum value; M and Sx: mean and standard deviation of the results; Sr and Sr%: absolute and relative standard deviation of repeatability; r: maximum deviation of repeatability in 95% of cases.

2.3- Conclusion

The standard deviations of repeatability Sr and Sr% observed are in agreement with the recommendations of the CNIEL/IE handbook which fixes a maximum limit of 15.10^3 /ml and 10% (level 100.10^3 /ml) and of the standard NF EN ISO 13366-2/FIL 148 which fixes a maximum limit of 6% (average level 150.10^3 /ml).

<u>3- Accuracy</u>

3.1- Procedure

The accuracy of the analyser was evaluated using 110 samples. The quantitative analyses were carried out in accordance with the evaluation of the repeatability (cf 2.1). The evaluation concerned the values obtained after calibration of the instrument with commercial SRMs (cow's milk) produced by Actilait-Cecalait.

The reference method ISO 13366-1/IDF 148: 2007 (microscopic method) was used for the enumeration of the somatic cells.

The samples were also analysed using the Bentley Somacount 150 (SCC150), previously calibrated with the same SRMs (single test).

3.2- Results

The results obtained are summarised in the table and figures below.

	BLUE CHANNEL	RED CHANNEL	SCC 150	Red channel /SCC 150	
n		110			
min		1			
max		1248			
Y (10 ³ /ml)		109			
X (10 ³ /ml)	106	111	109	106	
Sy (10 ³ /ml)		184			
d (10 ³ /ml)	10	15	13	-3	
Sd (10 ³ /ml)	22	27	29	12	
Sy,x	14	14	14	10	
Sy,x (%)	14,8	14,7	14,2	9,2	
b	0.899	0.871	0.859	1.045	
a	1	-1	2	-2	

Table: FCM accuracy criteria on cow's milk samples

n, min, max: number of results, minimum and maximum value; Y,X: mean of the results using the reference and instrumental methods; Sy: standard deviation of the results from the reference method; d, Sd: mean and standard deviation of deviations; Sy,x (Sy,x%): absolute and relative residual standard deviation; b, a: slope and intercept; Sy,x' (Sy,x'%): absolute and relative standard deviation forced through zero; b': slope forced through zero





Figures 1 and 2: Relationship between the FCM and reference results on cow's milk samples



Figure 3: Relationship between the results of the FCM and the SCC 150 on cow's milk samples (regression forced through zero)

It can be noted that the means and standard deviations of deviations are similar between the two channels and are respectively around 13.10^3 /ml and 25.10^3 /ml. The regression lines are significantly different from 1.00 (P = 1%). The residual standard deviations (about 14.10^3 /ml, relative 15%) are equivalent. The FCM results are very close to the values obtained with the SCC 150 analyser (a = 1.04; b = -1.7; Sy,x =10, d = -3).

3.3- Conclusion

With no regulations for milk payment or standards, it can however be noted that the slope obtained is significantly different from 1.00 for the FCM and the Somacount 150. The origin of this deviation could be due to the relatively low cell count of the samples used in this study (average 96.10^3 /ml). In comparison with the reference values, the absolute deviations remain low, approximately 10.10^3 /ml.

The comparison of the FCM with another instrument (Bentley Somacount 150) calibrated in the same conditions produced satisfactory results with a slope and an intercept not significantly different from 1.00 and 0.00 respectively.

Phase I of the evaluation of the FCM within the context of milk payment and control (cow, goat, ewe) was validated by the Scientific and Technical Committee of the French Ministry of Agriculture.

According to the evaluation report of the Bentley FCM^{TM} somatic cell counter on cow's milk – X. QUERVEL et Ph. TROSSAT – Actilait / Cecalait – April 2010

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