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# An attempt to assess accuracy of somatic cell count determination in cow's milk in the dairy herd improvement program – preliminary results

# Piotr Guliński\*, Łukasz Zaremba

Siedlee University of Natural Sciences and Humanities, Institute of Animal Production and Fisheries, Prusa 14, 08-110 Siedlee, Poland

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The aim of this study was to determine accuracy of the assessment of somatic cells count (SCC) in milk of Polish Holstein-Frisian (PHF) cattle obtained at four consecutive weeks: day 1 to day 7, day 8 to day 14, day 15 to day 21 and day 22 to day 29. Based on cytological quality three classes of milk were distinguished, with the following SCC in 1 mL: 1,000-200,000, 200,000-400,000 and >400,000, respectively. Depending on the number of lactations there were six groups of cows with 1, 2, 3, 4, 5 and 6 calvings. The average on-line SCC in 330 milk recordings was 331,000 in 1 mL, ranging from 13,000 to 1,856,000. It was found that the mean SCC in 1 mL of milk across the three cytological quality classes was 89,000, 285,000, and 751,000, while in the consecutive weeks it was 297,000; 326,000, 294,000 and 271,000. Analysis of variance for the natural logarithm of on-line SCC (actual somatic cell count) showed no significant differences between SCC in four different weeks. Determined on the basis of the natural logarithm from the actual SCC (InSCC) the accuracy was very high. The percentage differences between InSCC in the following weeks of the experiment ranged between +0.9 and -1.5%, compared to the recordings taken during the first week of the experiment. The small error in the evaluation of the number of somatic cells determined over a 4-week period was confirmed by the high correlation coefficients (r) between SCC recorded in different weeks. Because the results did not vary significantly across the 4-week period, it was concluded that the 4-week milk recording interval used in the official methodology correctly reflects the actual number of somatic cells present in milk during those 30 days.

KEYWORDS: accuracy /assessment / cows / somatic cell counts

<sup>\*</sup>Corresponding authors: piotr.gulinski@uph.edu.pl

Somatic cell count (SCC) limits are a key component of national and international regulations on milk quality [Moore et al. 2013]. The methodology for the determination of the number of somatic cells varies depending on the type of farms producing cow's milk. In assessing the production of dairy cattle both in Poland and worldwide, three basic methods are used: A4, A8, and AT4 [ICAR 2011]. An important element for the practical usefulness of each methodology is connected with its accuracy. It is necessary, as the reliability of milk recording depends on its frequency. In the case of daily analysis the accuracy would be 100%. However, this approach in commercial breeding practice is not viable economically. Therefore in many countries, including Poland, a variety of methods are used, with different intervals between milk recordings. In Method A4 the recording is carried out at least 11 times a year, each time over a period of 24 hours, whereas in Method AT4 milk samples are collected alternately in the morning or evening. In Method A8 the recording is performed at least 6 times a year, with milk quantity assessed over a period of 24 hours. The main methods applied in Poland are A4 and AT4 [PFHBiPM 2017]; in 2017 they were used for a total of 94.8% of all assessed cows. Their detailed methodology assumes that milking samplings and tests are carried out on average every 4 weeks, i.e. 28 days between each recording. Accuracy is an important element in the practical usefulness of any method. In the case of milk production assessment, its accuracy is determined by its frequency. For daily milk recordings the accuracy would be 100%. However, this approach in commercial breeding practice is no viable economically. Therefore, in many countries of the world, including Poland, various methods are used, with different intervals between milk recording. According to Johansson [1961], the error of daily milk yield recordings carried out at the intervals of 7, 14, 21 and 28 days is 2.5, 3.4, 4.4, and 5.4%, compared to those recorded every day.

Many researchers have investigated the effect of reducing milk recording frequency on the accuracy of lactation yields. Gatner et al. [2008] found that an alternate milk recording method at 4-week intervals provided a low bias and high accuracy of prediction for 100-, 200- and 305-day milk yields, while milk recording methods at 6-week intervals predicted 305-day milk yields with a greater bias and lower accuracy. Crosse and Clift [1988] reported that the relative error in the prediction of lactation milk yield ranged from 2.7 to 6% if the interval lasted from one to ten weeks. Cattin-Vidal [1990] also found an increase in the prediction error induced by the extending of the interval between recordings. Hargrove [1994] and Wangler et al. [1996] reported that with the extension of the interval between successive recordings, prediction accuracy decreased together with the cost of recordings. Pander et al. [1993] determined that the use of recording methods less frequent than A4 resulted in cost reduction without a proportional loss in accuracy when estimating 305-day yields. When total lactation milk yield is considered, the accuracy of all the methods [A4, AT4, A6, and AT6) is greater than 98% [Aleandri et al. 2003]. Berry et al. [2005] reported that for heifers the A8 scheme predicts on average a 305-day yield similar to the A4-predicted 305-day yield.

To date no study concerning the estimation of error in the assessment of the somatic cell count in cow milk was conducted in Poland, while SCC estimation is the requirement in the methodology of dairy herd improvement programs. It needs to be pointed out here that there is a lack of studies in the available literature comparing the number of somatic cells sampled once over four weeks and sampled every day over the same time period.

The main objective of the work was to assess fluctuations of SCC over a time period. To this end, the period of 30 days was divided into four parts: day 1 to day 7, day 8 to day 14, day 15 to day 21, day 22 to day 30. The final aim of the study was to evaluate the importance of the milk sampling period for the accuracy of SCC determination.

## Material and methods

The experiment was conducted between 9 December 2019 and 7 January 2020. The subject of the analyses was milk from *Polish Holstein-Friesian* cows in a dairy farm located in the Czyżew commune, the Wysokie Mazowieckie county in the Podlaskie province. A total of 11 cows of different ages were recorded. Milk samples were taken during the evening milking using the Bentley Flow Cytometer. The somatic cell count in milk was determined with the Fossomatic Minor electronic device, which uses the Charge Coupled Device (CCD) technique.

The start date of the experiment was random. In no case did it coincide with the calving date of the cows included in the experiment. The main criterion for the inclusion of cows in the experiment was the SCC level in the milk that allowed the cows to be included in one of the 3 cytological quality classes. For each of the 11 cows, their inclusion in the experiment took place during the 3rd to the 6th week of lactation.

Data on milk cytological quality and yield came from a total of 330 observations. In the first stage, the basic cytological quality class, i.e. SCC in 1 mL of milk, was determined. Subsequently, changes in the periodic yield of milk (kg) were studied. In the third stage, an attempt was made to estimate the accuracy of SCC assessment method adopting the results determined within the 1<sup>st</sup> week as 100%.

A number of factors that affected the cytological quality of milk were identified. Using somatic cell counts, the animals were grouped into three cytological quality classes with 1,000-200,000, 200,000-400,000 and >400 000 somatic cells in 1 mL milk, respectively. The period of 30 days was divided into four parts: day 1 to day 7, day 8 to day 14, day 15 to day 21, day 22 to day 30. Cows were allocated to 6 groups: 3 cows with 1 lactation, 1 cow with 2 lactations, 2 cows with 3 lactations, 2 cows with 4 lactations, 2 cows with 5 lactations, and 1 cow with 6 lactations. Detailed data on the number of animals and observations within each class are presented in Table 1. The effect of the experimental period on the number of somatic cells was assessed within each of the 3 cytological quality classes.

Since the empirical distribution of somatic cell count in milk departures from a normal distribution, the logarithmic transformation was applied.

The following linear model was used:

$$y_{ijkl} = \mu + A_i + B_j + C_k + e_{ijkl};$$

where:

 $y_{iikl}$  - the observation of SCC;

 $\mu$  – overall mean;

 $A_i$  – random effect of i-th animal;

- Bj fixed effect of j-th milk sampling period (j = 1... 4);
- $C_k$  fixed effect of k-th lactation (k = 1 ... 6);
- e<sub>iikl</sub> residual effect connected with *ijkl-th* observation.

The significance of differences between the means was estimated with the Fisher-Snedecor test (ANOVA) and Duncan's test at P $\leq$ 0.05. In the statistical analysis the PROC GLM (General Linear Model) and CORR procedures of the SAS statistical package version 8.1. were used [SAS Institute Inc., 2014].

## **Results and discussion**

#### Number of somatic cells and daily milk yield

Tables 1 and 2 presents factors affecting the actual SCCs, the natural logarithm from their number (LnLKS), and daily yield of milk. Mean SCC in 330 milk samples was 331,000 in 1 mL (Tab. 1). The study confirmed a high variability in the number of somatic cells, with the smallest SCC of 13,000 and the highest of 1,856,000. The results obtained in the experiment over a period of 1 month for 11 cows indicated only slight differences between the average numbers of somatic cells in milk during each week, as the mean SCC over the 1st, 2nd, 3rd and 4th week was 297,000; 326,000; 294,000; and 271,000 in 1 mL, respectively (Tab. 2). No significant differences between the numbers of somatic cells in milk during different weekly periods were estimated (Tab. 2). There were much larger differences between SCC in milk from cows with different lactations. The lowest SCC was observed for cows with 2 lactations, while it was the highest for two cows with 5 lactations. The LSM (Least Square Mean) of SCC for those cow groups was 39,000 and 675,000 in 1 mL, respectively, with the analysis of variance confirming the statistical significance of the lactation effect on SCC at  $P \leq 0.05$ . It was found that in the designated three cytological quality classes of 1,000-200,000; 200,000-400,000; and above 400,000 the average SCC in 1 mL of milk was 89,000; 285,000; and 751,000, respectively (Tab. 4). The numbers of somatic cells for all the cows in other lactations were comparable, within the limits set by the above extreme groups. In general, the cytological quality of milk in the cow population was found to be high, meeting the standard for extra-class milk applicable

 Table 1. Descriptive statistics for SCC, LnSCC, daily milk yield and the number of observations within selected factors

Item							
Mean (SE) <sup>1</sup> of SCC ( $10^3/1$ mL)	3	31 (17.	2)		range	13-185	6
Mean (SE) of LnSCC		5.30 (	0.06)		range	2.56-7.	53
Mean (SE) of daily milk yield (kg)		25.4 (0	.19)		range	14-32	
Number of cows	11						
Overall number of observations	330						
Observations within factors:							
observation period (day)	1 to 7	'	8 to 14		15 to 22	2	23 to 30
No. <sup>2</sup>	77		77		77		99
milk quality class	1	l		2			3
No.	14	41		97		(	92
lactation (no)	1	2	3		4	4	5
No.	90	30	60		60	60	60

<sup>1</sup> SE standard error of means.

<sup>2</sup> Number of observations within each factor class.

 Table 2. Least square means and standard errors (in parenthesis) of SCC in 1 mL of milk, natural logarithm of SCC, and daily milk yield within analysed factors

Factor	SCC (10 <sup>3</sup> /1 mL)		LnSCC		Daily yield of milk (kg)		
Factor	LSM (SE)	Range	LSM (SE)	Range	LSM (SE)	Range	
		Investigation periods (day)					
1-7	297 (36.3)	18-1383	$5.16^{A}(0.12)$	2.89-7.23	$26.2^{A}(0.34)$	20-32	
8-14	326 (40.2)	15-1856	$5.21^{A}(0.13)$	2.71-7.53	25.7 <sup>AB</sup> (0.35)	20-32	
15-21	294 (35.8)	13-1250	$5.04^{A}(0.13)$	2.56-7.13	25.3 <sup>B</sup> (0.41)	14-31	
22-30	271 (27.2)	21-1148	5.05 <sup>A</sup> (0.11)	3.04-7.05	$26.2^{A}(0.37)$	18-32	
Lactation (number)							
1	238 (22.8)	18-1015	4.95 <sup>°</sup> (0.11)	2.89-6.92	25.4 <sup>C</sup> (0.28)	20-30	
2	39 (5.5)	13-152	$3.46^{E}(0.10)$	2.56-5.02	29.6 <sup>A</sup> (0.27)	26-32	
3	353 (29.9)	79-983	$5.68^{\mathrm{B}}(0.08)$	4.37-6.89	$27.5^{\mathrm{B}}(0.35)$	23-32	
4	379 (24.8)	140-783	$5.82^{\mathrm{B}}(0.06)$	4.94-6.66	23.9 <sup>D</sup> (0.34)	20-30	
5	675 (54.3)	109-1856	$6.26^{A}(0.10)$	4.69-7.53	$22.1^{E}(0.34)$	14-27	
6	101 (8.6)	35-256	$4.52^{\rm D}(0.07)$	3.56-5.55	26.6 <sup>B</sup> (0.46)	18-31	

LSM (SE) - least square mean (standard error).

in Poland and the rest of the European Union. As Moore *et al.* [2013] pointed out, the European Union SCC regulatory limit of 400,000 in 1 mL is gradually adopted as an international export standard in many milk producing countries. An example is the United States, where such an SCC threshold has been introduced for companies exporting products to the EU. It should be stressed that the national limit in the United States is 750,000 SCC/mL for local consumption. In Brazil, on the other hand, 1 million SCC/mL is allowed.

The right part of Table 2 presents the daily yield of milk in the cow population. The average daily yield in 330 observations was 25.4 kg. Considerable differences were observed between cows in different lactations. In this experiment the highest

daily yields were produced by cows with 2 lactations (29.6 kg) and the lowest by those with 5 lactations (22.1 kg).

The results are in line with the data presented by Guliński *et al.* [2016]. The analyses carried out by those authors showed a negative correlation between the number of somatic cells and milk yield. An increase in milk yield was accompanied by a decrease in the number of somatic cells in 1 mL of milk. This meant an improvement in the cytological quality of milk. For the 4 groups of cows indicated in te Methods section with the daily yields of  $\leq$ 15, 15-25, 25-35 and >35 kg, the number of somatic cells in 1 mL of milk was 771,000, 393,000, 240,000 and 180,000, respectively. In a study by Borkowska and Januś [2010], the LnLKS value decreased significantly with the increase in daily milk yield of Montbeliarde cows - from 12.20 at the yield  $\leq$  15.0 kg, to 11.80 at the yield> 35.0 kg.

In a study by Jakiel *et al.* [2011] the correlation between SCS (Somatic Cell Score) and daily milk yield of 5638 Polish Holstein-Friesian primiparous cows was negative (-0.14). The relationships between SCS and daily milk yield decreased when the somatic cell count in milk increased (-0.14 in SCC class I and -0.03 in SCC class IV). According to Grodzki [2011], a decrease in the yield of milk with a deteriorated cytological quality ranged from 10 to 20%. Nelson and Philipot [2006] pointed out that SCC in older cows may be increased due to past infections as well as gland injuries, which is also associated with a reduction in milk yield.

Silva *et al.* [2018] found the coefficient of variation for somatic cell counts in 4586 milk samples at 68.1% for Holstein cows from commercial dairy farms in Brazil. A similar coefficient of variation was reported by Mijić *et al.* [2012] for cows of the Holstein breed in Croatia. The average coefficient of variation for ln-transformed online somatic cell count for cows in the first, second, third and higher lactations was 24.1, 22.3 and 21.6%, respectively. Duckowa *et al.* [2019] analyzed samples of raw milk taken from three industrial dairies and from three on-farm dairies producing traditional Slovak cheese. Somatic cell counts in milk samples from industrial dairies (mean value 326,550 in 1 mL) were lower than in milk samples from on-farm dairies (mean value 507,670 in 1 mL). The average coefficient of variation ranged from 9.2 to 39.1% for industrial dairies and from 47.1 to 155.3% for on-farm dairies.

In a study by Forsbäck *et al.* [2010], udder-quarter and cow composite milk samples were collected during morning and evening milkings for 3 weeks, providing a total of 42 samples. Different milk components showed different levels of day-to-day variation, with the least variation in lactose (0.9%) and the greatest in fat content (7.7%). The day-to-day variation in daily production of total protein, casein, and whey protein contents ranged from 1.4 to 1.8%. Milk yield and SCC had a day-to-day variation of 7.0 and 2.0%, respectively. In the above study the day-to-day variation in SCC was estimated to be approximately 2% in cow composite and udder-quarter milk for healthy cows.

Chagunda et al. [2006] found the coefficient of variation for SCC to be 4.7% in healthy cows and 11.4% in cows with clinical mastitis, which indicates

that this disease increases the variation in SCC. However, that study included a greater number of samples from three different breeds and the coefficient of variation presented was the overall variation. Because high SCC or mastitis was not included in the statistical model used in the study, the variation caused by this was incorporated into the day-to-day variation. Cullen [1967] found that the variation in SCC was numerically greater in udder quarters with high SCC. This implies that cows with udder disturbances might show greater day-to-day variation in the milk from the affected quarter. The average coefficient of variation in composite samples taken at short intervals was 30 to 35% [Cullen 1967], whereas over the whole lactation it may range from 69 to 301% [Duitschaever and Ashton 1972]. According to Schultz [1977], day-to-day variation for individual cows is considerably greater in infected cows than uninfected ones. Therefore, a single SCC test result is relatively inconclusive and classification of the infection status should be determined on the basis of a series of counts. There is variation in SCC throughout milking, as well as diurnal variation from milking to milking [Dohoo and Meek 1982]. Diurnal variation consists of higher SCC in the evening than in the morning milking, and the difference is assumed to be due to the interval between milkings [Syrstad and Ron 1978].

In a study by Quist et al. [2008] a total of 16 farms (14 with cows milked twice a day  $(2^{\times})$  and 2 with cows milked three times a day  $(3^{\times})$  across Canada participated in a 5-d milk-sampling study, with 27,328 milk samples collected and analyzed for fat and protein yields and SCC. Individual cow milk weights and composite samples were collected at every milking for 5 consecutive days between October 2004 and September 2005. Milk was collected into standard Dairy Herd Improvement (DHI) sample vials and sent to the DHI laboratory for analyses of fat, protein, and SCC. Descriptive statistics for both  $2^{\times}$  and  $3^{\times}$  herds for milk yield and fat and protein percentages followed a typical pattern throughout lactation. The somatic cell linear scores were higher in early lactation for first-lactation cows (4.7 vs. 3.8), but were higher at the end of lactation for cows in the second lactation or greater (5.1 vs. 4.9). The 2× herds had higher milk yields in the morning ( $\sim$ 17 vs.  $\sim$ 14 kg), whereas the 3×herds had the lowest milk yields in the morning, with yields peaking at the evening milking (~9 vs. ~11.2 kg). The SCC results of the  $2\times$  and  $3\times$  herds show that throughout the 5-d sampling period no consistent diurnal variation was present; however, morning milkings of the  $3 \times$  herds had the lowest values each day.

Nørstebø *et al.* [2019] analyzed data from 62,471 milkings across 173 lactations of 129 cows. Ln-transformed on-line cell counts (SCC) values (in 1000 cells/ml) as the outcome (LnSCC) in linear mixed models, with random intercepts at the cow-level and lactation-level within cow were used. The arithmetic and geometric mean SCC value in the final data set was 96,629 cells/mL and 35,279 cells/mL, respectively. The lowest SCC value was 1000 cells/mL (detection limit) and the highest was 7,474,000 cells/mL. It was possible to explain 15.0% of the variability in LnSCC with the following fixed effects: lactation stage, parity, milk yield, SCC in residual milk from the previous milking, inter-quarter difference between the highest and lowest

conductivity, season, intramammary infection, and genetic lineage. When including the random intercepts, the degree of explanation was 55.2%. The individual variables explained only a small part of the total variability in LnSCC. It was concluded that physiological or normal variability is probably responsible for a large part of the overall variability in SCC in cows without clinical mastitis. Sensor repeatability was evaluated by analyzing milk from the same sample multiple times. The coefficient of variation was 0.11 at an SCC level relevant for detection of subclinical mastitis. Diurnal variation of SCC between a.m. and p.m. milk may be very important [Reneau 1986]. It has been reported that p.m. milk samples have twice the number of somatic cells as a.m. samples [Cullen 1967]. In a study by Kwaśnicki *et al.* [2005] somatic cell count in the p.m. and a.m. milk samples was 493 and 417 thousands in 1 mL, respectively. The average coefficient of variation for on-line SCC was very high (176%).

#### Numbers of milk samples depending on SCC

Table 3 shows numbers of milk samples of different cytological quality, with different SCC in 1 mL of milk. The data in this table confirm the fact that the cytological quality of milk from the cows in this experiment was of high quality. Nearly 25% of the observations met the highest quality criterion, i.e. they contained up to 100,000 somatic cells in 1 mL. Another 124 samples contained less than 300,000 somatic cells. Overall, nearly 78% of the samples came from fully healthy cows and 22% from cows with subclinical inflammation of the mammary gland. The distribution obtained in this study is classical and confirms the literature data related to the distribution of mammary gland inflammation in dairy herds in Poland.

SCC (10 <sup>3</sup> /mL)	Numbers of samples	Cumulative numbers of samples	Percentage	Cumulative percentage
0-100	80	80	24.24	24.24
100-300	124	204	37.58	61.82
300-500	51	255	15.45	77.27
500-700	27	282	8.18	85.45
700-900	22	304	6.67	92.12
900-1100	15	319	4.55	96.67
1100-1300	9	328	2.73	99.39
1300-1500	1	329	0.30	99.70
1500-1700	0	329	0.00	99.70
1700-1900	1	330	0.30	100.00

Table 3. Numbers of milk samples according to on-line SCC in 1 mL of milk

## Assessment of accuracy of SCC determination in cow's milk

Table 4 provides a summary of the present experiment determining the accuracy of the assessment, based on studies lasting over a 4-week period. The results showed that for the cows assessed the average actual SCC in 1 mL of milk in the  $1^{st}$ ,  $2^{nd}$ ,  $3^{d}$  and  $4^{th}$  week was 297,000, 326,000, 294,000 and 271,000, respectively. These

Milk	SCC	Accuracy of	I mSCC	Accuracy of	
sampling	$(10^{3}/1 \text{ mL})$	determination	LISCU	determination	
period	LSM (SE)	of SCC (%)	LSM (SE)	of LnSCC (%)	
Clas	s 1 of cytological	quality (SCC=1	$\times 10^3 - 200 \times 10^3$	/1 mL)	
Day 1 to 7	95 (11.5)	100*	$4.29^{A}(0.14)$	100*	
Day 8 to 14	88 (9.8)	-7.4	$4.24^{A}(0.14)$	-1.2	
Day 15 to 21	89 (9.7)	-6.3	$4.22^{A}(0.13)$	-1.6	
Day 22 to 30	86 (8.4)	-9.5	$4.23^{A}(0.11)$	-1.5	
Overall mean	89 (4.8)	х	4.24 (0.06)	х	
Class	2 of cytological of	quality (SCC=200	$10^{3} - 400 \times 10^{3}$	0 <sup>3</sup> /1 mL)	
Day 1 to 7	289 (10.4)	100*	$5.64^{AB}(0.02)$	100*	
Day 8 to 14	276 (11.5)	-4.5	$5.60^{AB}(0.04)$	-0.7	
Day 15 to 21	262 (9.2)	-9.0	5.55 <sup>B</sup> (0.04)	-1.6	
Day 22 to 30	301 (9.1)	+4.2	$5.69^{A}(0.03)$	+0.9	
Overall mean	285 (5.3)	Х	5.63 (0.02)	Х	
	Class 3 of cytolog	gical quality (SCO	$>400 \times 10^3 / 1 \text{ m}$	L)	
Day 1 to 7	793 (71.1)	100*	$6.59^{A}(0.10)$	100*	
Day 8 to 14	771 (66.4)	-2.8	$6.57^{A}(0.07)$	-0.2	
Day 15 to 21	740 (46.3)	-6.7	$6.56^{A}(0.06)$	-0.3	
Day 22 to 30	705 (42.8)	-11.1	$6.51^{A}(0.06)$	-1.2	
Overall mean	751 (28.3)	Х	6.56 (0.04	х	
Classes 1, 2, and 3 of cytological quality (SCC $\ge 1 \times 10^3/1$ mL)					
Day 1 to 7	297 (36.3)	100*	$5.16^{A}(0.12)$	100*	
Day 8 to 14	326 (40.2)	+9.7	5.21 <sup>A</sup> (0.13)	-0.7	
Day 15 to 21	294 (35.8)	-1.0	$5.04^{A}(0.13)$	-1.3	
Day 22 to 30	271 (27.2)	-8.7	$5.05^{A}(0.11)$	- 0.5	

**Table 4.** Accuracy of determination of the number of somatic cells across weekly periods and cytological quality classes

Means in columns marked with different letters differ significantly at P $\leq$ 0.05. \*SCC and LnSCC in the first week in each quality class.

differences were therefore minor and the analysis of variances did not confirm their statistical significance.

An attempt to assess the accuracy of SCC determination using the assessment error is presented in Table 4. The accuracy of SCC determination was calculated for all weekly periods and for all cytological classes. The accuracy determined on the basis of the natural logarithm from the actual SCC was very high. The percentage differences between LnSCC in the following weeks of the experiment ranged between +0.9 and -1.5 %, compared to the recordings taken during the first week of the experiment. On this basis it was concluded that the actual SCC did not vary much during 30 days of sampling, which means that the 4-week interval used in the official methodology correctly reflects the actual SCC in milk during those 30 days.

The results regarding the low error of assessment and the actual number of somatic cells determined over a 4-week period was confirmed by correlation coefficients obtained between SCC found in milk in the 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> week (Tab. 5). The correlation coefficients (r) obtained for such correlated characteristics were very high, at 0.72, 0.80 and 0.74, respectively. Analysis of variances confirmed their high statistical significance at P $\leq$ 0.01.

•	-		
Milk sampling time period	Day 1 to 7	Day 8 to 14	Day 15 to 21
Day 8 to 14	0.72*		-
Day 15 to 21	0.80*	0.86*	
Day 22 to 30	0.74*	0.79*	0.82*

 Table 5. Correlation coefficients between natural logarithm from the actual number of 1 mL somatic cells for milk samples obtained during different periods

\*Correlation coefficient with high statistical significance at  $P \leq 0.01$ .

Milk samples collected from 11 cows during 1 month showed only slight differences between the average numbers of milk somatic cells in each weekly period. The average SCC in 1 ml of milk in the 1st, 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> week was 297,000, 326,000, 294,000 and 271 000, respectively. The analysis of variance for the natural logarithm from the actual number of somatic cells indicated no significant differences between the numbers in different weekly periods. The accuracy of the assessment based on the natural logarithm from the actual number of somatic cells was high, with the difference in LnSCC between +0.9 and -1.5% compared to the recordings taken during the first week of the experiment. The low error in the assessment was confirmed by correlation coefficients between the number of somatic cells over the 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, and 4<sup>th</sup> weeks of the experiment. The correlation coefficients (r) for such correlated characteristics were very high, at 1, 0.72, 0.80 and 0.74. On the basis of the results it was concluded that the number of somatic cells did not vary much during the 30-day period. Thus, the 4-week interval used in the official methodology correctly reflects the actual number of somatic cells across four weekly periods.

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