



SHORT COMMUNICATION

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Influence of the sampling device on somatic cell count variation in cow milk samples (by official recording)

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Abstract

The objective of this study was to investigate the variability in cow's milk somatic cell counts (SCC) depending on the type of milk meter used by dairy farms for official milk recording. The study was performed in 2011 and 2012 in the major cattle area of Spain. In total, 137,846 lactations of Holstein-Friesian cows were analysed at 1,912 farms. A generalised least squares regression model was used for data analysis. The model showed that the milk meter had a substantial effect on the SCC for individual milk samples obtained for official milk recording. The results suggested an overestimation of the SCC in milk samples from farms that had electronic devices in comparison with farms that used portable devices and underestimation when volumetric meters are used. A weak positive correlation was observed between the SCC and the percentage of fat in individual milk samples. The results underline the importance of considering this variable when using SCC data from milk recording in the dairy herd improvement program or in quality milk programs.

Additional key words: dairy cattle farm; milk yield; sampling system; SCC

Abbreviations used: DHIP (dairy herd improvement program); IMI (intra-mammary infection); SCC (somatic cell count)

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Introduction

The use and interpretation of individual cow and bulk tank milk somatic cell count (SCC) data have been well described and widely adopted by the dairy industry as an indirect measure of mastitis risk (Godden *et al.*, 2002). Milk samples are routinely collected at milking time for SCC analysis within an official milk recording. Collecting and using SCC data is essential to reduce the financial loss due to mastitis and to maximize the profitability of selling the highest quality milk at the highest price. The SCC from milk recording provides an efficient and economical way for most producers to monitor the success (or failure) of the herd's mastitis program and these data are used also in the dairy herd improvement program (DHIP).

Bacterial infection in the mammary gland is the most important cause of increased SCC. Some investigations have reported a relationship between milk yield and SCC in cattle without an intra-mammary infection (IMI) (Green *et al.*, 2006; Hagnestam-Nielsen *et al.*, 2009; Boland *et al.*, 2013). Nonbacterial factors that affect SCC include age, genetic, stage of lactation, season, stress, management, milk fractions (foremilk, cisternal milk, alveolar milk or remaining alveolar) or technicians taking the sample (Sawa & Piwczynski, 2002; Hagnestam-Nielsen *et al.*, 2009; Boland *et al.*, 2013). SCC may also vary between milkings and days (Riekerink *et al.*, 2007).

Somatic cells bind to the fat fraction of the milk (Schutz *et al.*, 1990), thus the SCC might be affected if a representative sample in terms of fat measuring is

not obtained. Also a significant positive correlation between SCC and fat content in individual cow milk samples has been reported (Sawa & Piwczynski, 2002).

Official milk recording samples are obtained using 3 types of milk sampling systems: portable, volumetric and electronic. In portable meters, a fixed portion of each milking fraction is diverted into a sample cup, given a representative sample of milk fractions (Fouz *et al.*, 2009).

Volumetric measurers are permanently installed in the milk line and consist of a jar into which all the milk from one milking is placed. At the bottom of the jar, a valve allows samples to be collected. The milk is mixed by opening the valve with the vacuum on so that air bubbles through the milk for 3 to 5 seconds before the vacuum is turned off and milk drops into the collection bottle. However, this is insufficient for complete mixing because fat globules under gravity move toward the top of the jar (Olson & Amick, 1986); mixing is essential and should be more intense the greater the volume of milk in the measuring jar (such as for high-producing cows) (Ma & Barbano, 2000). However, the long mixing time required cause delays in milking and destabilization of fat globules, with the consequent risk of lipolysis and its associated problems, such as rancid off-flavours (Evers, 2004).

Using an electronic meter, the milk from the entire milking passes through a device in which a valve opens when a certain quantity (200-300 g) is reached; this milk returns to the pipeline and the device refills. Each volume released is measured and used to determine milk yield per cow. A portion of the milk that passes through the device is deposited into a collection bottle from which the sample is obtained at the end of the milking. The International Committee for Animal Recording has registered up to 58 different models of electronic meters, although in most cases their performance is adjusted as described. Only a few perform the measurement by other methods, such as infrared light technology. The degree of representativeness varies according to the instrument model: some models collect a portion of each of the milking fractions, whereas other fills up with milk at the start or end of milking. Samples taken using these devices may not be representative in terms of fat content for two reasons: lack of representativeness in milk passage to the sample collection bottle and gravitational displacement of fat globules to the top of the sample collection bottle with no system for prior mixing (Fouz *et al.*, 2009).

Other models, such as LactoCorder® (ICAR, 2006)) are useful for recording production but not for sampling and are expensive and not used in official milk control in Spain.

The hypothesis tested was that differences between methods in their ability to collect and homogenise a representative milk sample affect the SCC. To address

this hypothesis, this study was designed (1) to evaluate the effect of the milk sampling systems on SCC and (2) to analyse the correlation between SCC and fat contents in individual cow milk samples by dairy farms enrolled for Official milk recording.

Material and methods

Area, farms and animals of study

The study was carried out during 2011 and 2012 in Galicia, NW Spain. Galicia is the major dairy cattle region in Spain, accounting for 35% of the whole milk produced in Spain and 1.3% in the European Union.

Galician Official Milk Recording data used in this study were from single lactations of 137,846 Holstein-Friesian cows belonging to 1,912 farms. The mean herd size of the studied farms was 45 cows (minimum 8, maximum 379).

Records were obtained from day-tests by the milk recording, during which the supervising technician measured the daily milk yield and collected a composite milk sample for SCC and fat determination (among other traits) following an alternative am-pm monthly recording scheme throughout the lactation period.

When a cow was dried off, total milk traits per lactation (including total milk yield and mean-adjusted % fat and SCC from whole lactation) were estimated and normalized to 305-d using the Fleischmasnn's method (ICAR, 2014). The relationship between the SCC and the type of milking sampling method (electronic, portable and volumetric) used in the farm was analyzed.

Lactations smaller than 240 days and lactations in which the cow had some episode of mastitis (SCC > 200,000 cells/mL during lactation) were not included for analysis.

Finally, for each cow, the following data (from milk recordings) were available for the study: type of meter used on its farm, standardized 305-day milk production, average % fat during the complete lactation, average SCC (also from the whole lactation) and calving number. For the analysis, calving number was divided into three categories: 1st, 2nd and ≥ 3 rd.

Somatic cell count and fat measurement

Milk samples were collected in 50 mL plastic containers with the preservative bronopol (2-bromo-2-nitro-1,3-propane-diol) previously added.

SCC (cell/mL) values were determined using cell counter FOSSOMATIC™ (MilkoScan, Foss, Hillerød, Denmark) and fat (%) contents by infrared spectroscopy according to the manufacturer instructions.

All analyses were carried out in the Interprofessional Milk Laboratory of Galicia (Mabegondo, A Coruña, Spain).

Statistical analysis

Data were analysed using STATA 11.0 software (StataCorp., TX, USA). Pearson correlation coefficient (ρ) was used to assess the correlation between SCC and fat contents. Initially, the influence of the sampling system on mean SCC from the whole lactation was examined by ANOVA in a univariate approach. After that, a generalized least square linear model was used to study this influence in a multivariate approach. In this model, the mean SCC from the whole lactation was the continuous outcome variable. The explanatory variables included milk sampling device (portable, volumetric or electronic), calving number, mean-adjusted fat (from the complete lactation) and production level (litres/lactation). The interaction terms volumetric meter and mean-adjusted fat $\geq 3.8\%$, and electronic meter and mean-adjusted fat $\geq 3.8\%$ were evaluated (3.8% was the average percentage of fat in the studied population). Herd was included as a random effect variable to account for clustering at herd level.

Results

The milk sampling systems used were electronic in 37.1% of the farms (23.9% lactations), portable in

45.7% of the farms (57.7% lactations), and volumetric devices in 17.2% of the farms (18.4% lactations).

A weak positive correlation was observed between the SCC and the percentage of fat in milk ($\rho=0.154$, $p<0.001$).

A description of the distribution of the SCC values according to milk sampling device and the differences in the mean SCC for the three systems, in a univariate approach, are shown in Table 1. The one-way ANOVA indicated that SCC were significantly higher when electronic meters were used compared to portable or volumetric ones ($p<0.05$). No differences on SCC were observed between portable and volumetric meters when using this method ($p>0.05$).

Regression analysis indicated that samples collected with volumetric meters had a SCC that was, on average, 12,030 cells/MI ($p<0.001$) lower than those from portable meters. On the other hand, samples from electronic meters had significantly higher SCC (23,540 cells/mL, $p<0.001$) than those from portable devices (Table 2). The SCC slightly decreased (by 7,490 cells/mL) when the 305-d milk yield augmented (by 70,520 cells/mL) when the mean fat from the whole lactation increased. The interaction terms between volumetric meter and mean-adjusted fat $\geq 3.8\%$ and between electronic meter and mean-adjusted fat $\geq 3.8\%$ were significant. They indicated that the effect of the type of meter on the SCC was higher in cows with fat $\geq 3.8\%$. In this case, samples from volumetric meters had SCC, on average, 48,710 cells lower than those from portable ones, whereas samples from electronic meters had SCC that were 70,920 cells higher than SCC obtained from portable meters. No evidence of

Table 1. SCC distribution (cells/mL \times 1000) for each type of sampling system.

Sampling system	Mean of SCC (confidence interval)	Minimum	P25	Median	P75	Maximum
Portable	69,240 ^a (69,090-69,390)	10	28	54	101	200
Volumetric	69,110 ^a (68,880-69,340)	10	27	53	100	200
Electronic	74,220 ^b (74,070-74,370)	10	30	60	110	200

^{a-b} Means within different superscripts were significantly different ($p<0.05$).

Table 2. Regression analysis of somatic cell count according to the sampling system, milk production level and % fat.

Variables	β coefficient (SCC \times 1000)	p -value	95% confidence interval of β	
			Lower limit	Upper limit
Constant	86.71	<0.000	85.77	87.66
Portable vs Volumetric meters	12.03	<0.001	6.71	17.36
Portable vs Electronic meters	-23.54	<0.001	-29.29	-17.78
Milk production (L/lactation \times 1000)	-7.49	<0.001	-7.65	-7.33
Mean-Adjusted % fat	70.52	<0.001	68.44	72.59
Volumetric meter \times Mean-Adjusted fat $\geq 3.8\%$	48.71	<0.001	40.37	57.05
Electronic meter \times Mean-Adjusted fat $\geq 3.8\%$	-70.92	<0.001	-76.95	-64.88

association between SCC and calving number was found ($p > 0.05$).

Discussion

In the present paper, only data from lactations where all milking tests showed SCC lower than 200,000 cells/mL were included. Although, in general, SCC did not have a normal distribution, when considering only these lactations, the normality assumption is fulfilled. The results showed a weak positive correlation between SCC and fat content. Previous studies indicated a highly significant positive relation between both parameters (Sawa & Piwczynski 2002; Rajcevic *et al.*, 2003). Correlation between SCC and fat content was not observed when all data were analysed (including lactations with episodes with high SCC).

The results observed in this study are in accordance with those reflected in previous papers. Green *et al.* (2006) indicated that in cattle with no IMI, increases in milk yield results in SCC dilution (Boland *et al.*, 2013). Farms in Galicia, with production level above 35 kg/cow/day had mean bulk tank milk SCC of 87,500 cells/mL while those with production levels lower than 25 kg have an average mean tank milk SCC of 127,000 cells/mL (AFRICOR, 2012). These means were calculated including all samples, not only those with less than 200,000 cell/mL.

Milk composition, including SCC, changes during the course of milking (Sarikaya *et al.*, 2005). Portable meters are the only ones that always obtain a representative sample of all the milking fractions and values of SCC are more reliable (ICAR, 2014). Besides, in both volumetric and electronic flow meters, the milk sample is collected at the end of milking. With the former, the sample is obtained through the valve located at the lowest part of the meter; due to the fact that milk fat globules (as well as the somatic cells associated to them) move upwards, estimated measurements of both parameters may be lower. On the contrary, in electronic flow meters, regardless of the model, the sample is taken from the upper part of a container and measurement estimates may be biased in the opposite direction. Moreover, in portable meters, the sample is taken during the milking process thus avoiding fat concentration at the end of milking, and the volume taken is small, possibly reducing the risk of sample fractioning and inaccurate fat and SCC measurements. As previously discussed, volumetric and electronic sampling devices may fail to obtain milk samples with a homogeneous fat distribution (Fouz *et al.*, 2009) and consequently SCC can be affected since they bind to the fat fraction of the milk.

Standardization of milk sampling procedures would reduce some of the variability in the SCC but would create important inconveniences: (1) the cost of providing sampling systems for each milking point in large milking units, in which samples are obtained during electronic milk quantification; (2) the risk of destabilizing the milking vacuum by installing such devices in parlours with conventional vacuum reserve features, and (3) the difficulty of finding an adequate installation site in parlours with low milk lines, given that the sampling device should be placed lower than the udder level in a vertical position. When proportional meters are installed, effective vacuum level at the teat end could be affected.

Findings indicate that the type of milk sampling system used has a substantial effect on the SCC recorded in individual samples obtained for official milk recording. Since these data are used for the DHIP and for genetic evaluations, this bias can be of great practical importance. Besides, SCC from milk recording is often used in quality milk programs. If a threshold value of 200,000 cells/mL is used to identify animals with IMI, the type of meter may affect the sensitivity and specificity of the diagnosis. The sampling system should fulfil two basic requirements: to collect a sample in which are represented all the milking fractions and not affected in its collection by the gravitational displacement of fat globule. Portable meters are the only that fulfil both conditions.

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