Effect of somatic cell count on composition and hygiene indicators of bulk tank milk

Efeito da contagem de células somáticas sobre a composição e indicadores de higiene do leite de tanque

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Abstract

This study evaluated the effect of somatic cell count (SCC) on composition and hygienic quality of dairy herd bulk tank milk – specifically, the effect of SCC of bulk tank of dairy herds on composition (fat, protein, total solids, nonfat dry solids) and on total bacterial count (TBC), psychrotrophic count (PC) and coliform count (CC) were evaluated. A total of 230 dairy herds located south of Minas Gerais and west of São Paulo were selected based on SCC geometric mean obtained from five monthly analyses preceding the start of the sampling. The dairy farms were classified according to SCC in three groups: low (< 250,000 cells/mL, n = 84), medium (> 250,000 and < 750,000 cells/mL, n = 79) and high SCC (> 750,000 cells/mL, n = 67). After herd selection, bulk tank milk samples were collected every 14 days for three months totaling 1380 samples, which were subjected to analysis of composition, TBC, PC, and CC. A decrease of TBC and CC was observed in herds with low SCC; however, herds with medium and high SCC had an increase in fat, crude protein, and total solids contents. A medium correlation was observed between TBC and PC (r = 0.6215), and also between PC and CC (r = 0.3692). Based on hygiene indicators and milk composition, a low and negative correlation between TBC and fat (r = -0.0585), PC and fat (r = -0.0585), and PC and total solids (r = -0.0662) was observed. Dairy herds with SCC < 250,000 cells/mL had higher bulk tank milk hygienic quality; however, considering the composition, herds with higher SCC produced higher milk fat and protein concentration.

Keywords: Microbiological culture. Diagnostic. Microorganisms. Milk quality. Sanity.

Resumo

Este trabalho avaliou o efeito da contagem de células somáticas (CCS) na composição e na qualidade higiênica do leite de tanque de rebanhos leiteiros. Especificamente, foram avaliados o efeito da CCS do tanque de rebanhos leiteiros na composição (gordura, proteína, sólidos totais, extrato seco desengordurado) e nas contagens bacteriana total (CBT), de psicrotróficos (CP) e de coliformes (CC). Um total de 230 rebanhos leiteiros localizados no sul de Minas Gerais e oeste de São Paulo foram selecionados com base na média geométrica da CCS obtida de cinco análises mensais anteriores ao início das coletas das amostras. As fazendas foram classificadas de acordo com a CCS em três grupos: baixa (< 250.000 células/mL, n = 84), média (> 250.000 e < 750.000 células/mL, n = 79) e alta CCS (> 750.000 células/mL, n = 67). Após a seleção dos rebanhos, amostras de leite do tanque foram coletadas a cada catorze dias durante três meses, totalizando 1.380 amostras, as quais foram submetidas às análises de composição, CBT, CP e CC. Uma redução da CBT e da CC foi observada em rebanho com baixa CCS; entretanto, rebanhos com média e alta CCS tiveram aumento nos teores de gordura, proteína bruta e sólidos totais. Uma média correlação foi observada entre CBT e CP (r = 0,6215) e também entre CP e CC (r = 0,3692). Com base nos indicadores de higiene e na composição do leite, foi observada uma correlação baixa e negativa entre CBT e gordura (r = -0,0585), CP e gordura (r = -0,0585) e CP e sólidos totais (r = -0,0662). Os rebanhos leiteiros com CCS < 250.000 células/mL apresentaram maior qualidade higiênica do leite de tanque; entretanto, considerando a composição, rebanhos com maior CCS tiveram maiores concentrações de gordura e proteína.

Palavras-chave: Cultura microbiológica. Diagnóstico. Microorganismos. Qualidade do leite. Sanidade.

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Introduction

Increased competitiveness in the milk supply chain requires the production of high quality raw milk, which depends on milk processing, yield, and acceptability of dairy products (BOTARO et al., 2013). In order to characterize milk quality, the most commonly used criteria are hygiene milk composition, and sensory characteristics of consumer interest (BRANDT et al., 2010). The main factors that affect raw milk quality are the prevalence of subclinical mastitis, hygiene conditions, and storage during milk production and processing (FORSBÄCK et al., 2010b).

Mastitis is an inflammation of the mammary gland characterized by several changes in the physical and chemical characteristics of the milk (REIS et al., 2011). This disease causes serious economic losses to the dairy industry because it negatively affects the amount of milk produced by dairy herds, milk quality, and shelf life of dairy products (HOGEVEEN et al., 2011).

Due to the increase of somatic cell count (SCC) of milk in response to intramammary infection, milk quality is reduced because of changes in milk composition, such as increased concentration of free fatty acids and serum proteins, decreased casein and lactose content, and changes in milk mineral contents (ÅKERSTEDT et al., 2012). These changes occur in response to the action of inflammation mediators and the activity of enzymes and toxins synthetized by mastitis pathogens (BRANDT et al., 2010).

Hygienic quality of milk at the farm level can be estimated by TBC of bulk tank milk (PANTOJA et al., 2012). However, the wide diversity of raw milk contamination sources contributes to diminish the accuracy of TBC in identifying the contamination sources (BRITO et al., 1998). Thus, in addition to using TBC to evaluate the hygienic quality of milk, counting of specific groups of microorganisms such as psychrotrophic counting (PC) and coliform counting (CC) (ELMOSLEMANY et al., 2010) is also recommended. The use of counting methods for groups of microorganisms in bulk tank milk may indicate the possible milk contamination source, which may be useful in troubleshooting high bacteria counts in farm milk (BAVA et al., 2011).

Psychrotrophic counts estimate the number of microorganisms that can proliferate at refrigerated temperatures of raw milk and that can grow at temperatures from 0 ° to 20 °C (MARTH; STEELE, 2001). Psychrotrophic microorganisms are widely found in the environment and the presence of these bacteria in bulk tank milk is indicative of failure in cleaning of the milking machine (CEMPÍRKOVÁ; MIKULOVÁ, 2009). These bacteria synthesize heat-resistant enzymes, which hydrolyze milk fat and protein, resulting in reduced dairy product quality and shelf life (SVENSSON et al., 2006). The synthesis of these enzymes is directly associated with PC, so that the greater the PC, the more enzyme is produced and lower the shelf life of the dairy products (CEMPÍRKOVÁ; MIKULOVÁ, 2009).

Microorganisms of the coliforms group are commonly found in the feces of cows; however, this group may be distributed throughout the farm environment (JAYARAO et al., 2004). Generally, coliforms may be associated with cases of foodborne intoxication, but this group is easily destroyed by pasteurization (PANTOJA et al., 2011). Nevertheless, if there is some failure during heat treatment, these microorganisms can deteriorate the milk (GAUCHER et al., 2008) and cause disease in consumers (BLUM et al., 2008). High CC in bulk tank milk is indicative of fecal contamination, failure in milk refrigeration or in sanitization of milking machine and tank (COTON et al., 2012).

Thus, the hypothesis tested in this study was that the bulk tank SCC is negatively associated with milk composition and hygienic quality of milk from the herd level. The aim of this study was to evaluate the effect of bulk tank milk SCC on fat, protein, total solids, and non-fat dry solids contents, as well as milk hygiene indicators (TBC, PC, and CC), and to evaluate the correlation between hygiene indicators (TBC, PC, and CC) and milk composition (fat, protein, total solids, and non-fat dry solids) in bulk tank milk of commercial herds.

Material and Methods

Dairy herds selection and milk samples collection

In this study we selected milk suppliers of a dairy located in the southern region of the state of Minas Gerais, Brazil. The dairy performed a payment program by milk quality, which required weekly monitoring of milk composition, SCC, and TBC of herds. The milk payment system was based on bonus and penalty criteria of milk price, according to the geometric mean of TBC, SCC, and protein content of the collection of five monthly bulk tank milk samples from all herds. Moreover, the dairy had a training and qualification program for the truck driver who collected milk samples.

For two years, 230 dairy herds (115 each year) were randomly selected based on geometric mean of SCC of five bulk tank milk samples. The sampling period during two consecutive years was done from August to November annually. The selected herds were distributed in three groups according to SCC: Low < 250,000 cells/mL (n = 84 herds); Medium > 250,000 and < 750,000 cells/mL (n = 79 herds); High > 750,000 cells/mL (n = 67 herds).

During the milk sampling period, selected herds were sampled twice a month for three months, totaling six samplings per herd and 1380 samples throughout the study (690 samples each year). Truck drivers collected milk samples directly from bulk tank after 5 to 10 minutes of stirring in sterile vials without preservatives for microbiological analysis (TBC, PC, and CC) and in vials with preservative bronopol (2-bromo-2-nitro-1,3-propanediol) for composition analysis (fat, protein, total solids, and non-fat dry solids) and SCC. After farm sample collection, all samples were stored in isothermal boxes at 4°C and taken to a sample collection center before analysis. The preservative free samples were frozen at -20 °C before being submitted to microbiological tests in a maximum period of seven days from the date of farm collection.

Microbiological analysis of milk

To determine TBC and PC, milk samples were thawed and kept at 4 °C. After vigorous vortexing for 15 seconds, samples were diluted in 1% buffered peptone water (Merck, Darmstadt, Germany) 1:10. Aliquots of 50 μL were inoculated in petri dishes with standard culture medium for colony counting (plate count agar, Oxoid CM0325, Basingstoke, Hampshire, England), using the automatic plating system (Spiral Plater-ID-DS Plus, Interscience, France) (LAIRD et al., 2004). The plates were incubated at 32 °C (± 1 °C) for 48 hours (± 2 hours) to determine TBC and at 12 °C for seven days to determine PC (FOOD AND DRUG ADMINISTRATION, 2001; LAIRD et al., 2004). To determine TBC and PC, all colonies of the plates were counted using an automatic colony counter (Scan 1200-ID, Interscience, France).

The total coliform count was determined by seeding 1 mL of the sample in Petrifilm™ plates (3M™ Petrifilm™ CC Count Coliform, St. Paul, MN, USA). Two plates for each sample were seeded, one undiluted and other diluted in 1% buffered peptone water (Merck, Darmstadt, Germany) in the ratio of 1:10, according to the methodology described by Elmoslemany et al. (2009b). The samples inoculated in Petrifilm were incubated at 32 °C for 24 hours (± 2 hours). Colony counting was performed using a manual colony counter. All red colonies that produced gas (LAIRD et al., 2004) were considered as total coliforms count.

Milk composition and somatic cell count

Milk composition analysis (fat, protein, total solids, and non-fat dry solids) and somatic cell count were performed in a maximum period of five days after sample collection at the farm. The contents of fat, protein, total solids, and non-fat dry solids were analyzed by infrared absorption, using MilkoScan FT+ (Foss North America, Eden Prairie, MN, USA). The somatic cell count was determined by flow cytometry, using Fossomatic™ FC (Foss Electric A/S. Hillerod, Denmark).

Statistical analysis

Results were analyzed using statistical analysis system (SAS Institute Inc., 2004). The results of microbiological analysis (TBC, PC, and CC) and of SCC were converted to logarithmic scale. Effects of SCC sampling and interaction were analyzed using Proc Mixed of SAS. Comparison of the means was done using Scheffé test that can be applied to any number of contrasts and does not require balanced sampling. A 5% significance level was used for all tests. We used the following statistical model: $Y = \mu + SCC + S + SCC^{*}S\ e$; where: Y = dependent variable; μ = mean of variable; SCC = SCC level: low (< 250,000 cells/mL), medium (> 250,000 and < 750,000 cells/mL) or high (> 750,000 cells/mL); S = sampling time; e = residual error.

Proc Corr of SAS was used to evaluate Pearson correlation between hygiene indicators (TBC, PC, and CC) and variables of milk composition (fat, protein, total solids, and non-fat dry solids).

Results

Milk microbiological analysis

Bulk tank milk samples were collected from 230 dairy herds to evaluate the effects of subclinical mastitis on TBC, PC, and CC. Herds that presented SCC < 250,000 cells/mL had lower TBC (P = 0.0387) and CC (P = 0.0072) values than herds with SCC > 250,001 cells/mL. There were no differences between PC from herds with low, medium, and high SCC. All of the hygiene indicators studied (TBC: $P < 0.0001$; PC: $P < 0.0001$; CC: $P < 0.0001$) and SCC ($P = 0.0211$) showed effects of sampling period. TBC ($P = 0.0305$) and CC ($P = 0.0497$) presented effects of interaction between the different kinds of SCC (low, medium, and high) and collection period of milk samples (Table 1).

Table 1 – Effect of somatic cell count (SCC) of dairy herds on total bacterial count (TBC), psychrotrophic count (PC), coliform count (CC) and somatic cell count (SCC) in bulk tank milk

Variable	n	SCC (× 1,000 cells/mL)*			SEM	*P*		
		< 250	> 250 e < 750	> 750		SCC	S	SCC*T
TBC**	1180	5.11^{a} (129.9)	5.31^{b} (203.7)	5.20^{b} (159.4)	0.021	0.038	< 0.0001	0.030
PC **	1162	4.92 (83.0)	5.16 (144.2)	5.21 (160.9)	0.024	0.091	< 0.0001	0.444
CC ***	1190	2.21^{a} (163.0)	2.43^{b} (268.4)	2.51^{b} (323.4)	0.029	0.007	< 0.0001	0.049
SCC****	1235	5.31^{a} (204.6)	5.66^{b} (457.3)	5.99^{c} (974.4)	0.009	< 0.0001	0.021	0.581

Data are presented as means on log10 transformation and on antilog10 transformation between parentheses; SEM: standard error mean; same letters in the same line do not differ by the Scheffé test (α = 5%); SCC: low, medium and high SCC; S: sampling time; SCC*S: interaction between SCC and sampling time. *$\log_{10}$ (untransformed data); **10^3 CFU/mL; ***CFU/mL; ****10^3 cells/mL

Bulk tank milk composition

Contents of fat, protein, total solids, and non-fat dry solids were determined to evaluate the influence of bovine subclinical mastitis on milk composition of dairy herds. Herds with SCC > 250,001 cells/mL had higher contents of fat ($P = 0.0048$), protein ($P = 0.0029$), and total solids ($P = 0.0173$) than herds that had SCC < 250,000 cells/mL. All of milk composition variables had significant effect of sampling period (fat: $P < 0.0001$; protein: $P < 0.0001$; total solids: $P < 0.0001$; SNF: $P = 0.0010$). Effects of interactions between kinds of SCC (low, medium, high) and collection period were not observed (Table 2).

Table 2 – Effect of somatic cell count (SCC) of dairy herds on mean of bulk tank milk composition

Variable (%)	n	SCC (× 1,000 cells/mL)			SEM	*P*		
		< 250	> 250 e < 750	> 750		SCC	S	SCC*S
Fat	1,222	3.55^{b}	3.72^{a}	3.70^{a}	0.009	0.0048	< 0.0001	0.9499
Protein	1,230	3.16^{b}	3.22^{a}	3.23^{a}	0.004	0.0029	< 0.0001	0.5202
Total solids	1,225	12.28^{b}	12.45^{a}	12.39^{a}	0.013	0.0173	< 0.0001	0.9849
Non-fat dry solids	1,232	8.73	8.73	8.68	0.005	0.3745	0.0010	0.5408

Data are presented as means; SEM: standard error mean; same letters in the same line do not differ by the Scheffé test (α = 5%); SCC: low, medium and high SCC; S: sampling time; SCC*S: interaction between SCC and sampling time

Correlation between variables of composition and hygiene indicators of bulk tank milk

Linear correlation coefficient of Pearson was calculated to evaluate the correlation between indicators of hygiene and milk composition: $|r| > 0.7$: high correlation; $0.3 < |r| > 0.7$: medium correlation; and $|r| < 0.3$: low correlation (BARBETTA, 2006). Medium and positive correlations were observed between TBC and PC (r = 0.6215) and between PC and CC (r = 0.3692). Low and positive coefficients of correlation were observed between SCC and TBC (r = 0.1632), SCC and CC (r = 0.0681), SCC and fat (r = 0.2217), SCC and protein (r = 0.2346), SCC and total solids (r = 0.1328), and TBC and CC (r = 0.2949). Low and negative correlations were observed between SCC and SNF (r = -0.0818), TBC and fat (r = -0.0585), PC and TS (r = -0.0662), and PC and fat (r = -0.0688) (Table 3).

Table 3 – Correlation coefficient between hygiene indicators and bulk tank milk composition

Variables		Log TBC	Log PC	Log CC	Log SCC	Fat	Protein	Total solids	Non-fat dry solids
Log TBC	r	–	0.6215	0.2949	0.1632	-0.0585	0.0098	-0.0488	-0.0172
	P	–	< 0.0001	< 0.0001	< 0.0001	0.0479	0.7396	0.0989	0.5602
Log PC	r	0.6215	–	0.3692	0.0511	-0.0688	-0.0160	-0.0662	-0.0414
	P	< 0.0001	–	< 0.0001	0.0848	0.0208	0.5902	0.0261	0.1630
Log CC	r	0.2949	0.3692	–	0.0681	-0.0478	-0.0360	-0.0549	-0.0313
	P	< 0.0001	< 0.0001	–	0.0204	0.1054	0.2213	0.0624	0.2878
Log SCC	r	0.1632	0.0511	0.0681	–	0.2217	0.2346	0.1328	-0.0818
	P	< 0.0001	0.0848	0.0204	–	< 0.0001	< 0.0001	< 0.0001	0.0042
Fat	r	-0.0585	-0.0688	-0.0478	0.2217	–	0.5306	0.9170	0.3713
	P	0.0479	0.0208	0.1054	< 0.0001	–	< 0.0001	< 0.0001	< 0.0001
Protein	r	0.0098	-0.0160	-0.0360	0.2346	0.5869	–	0.7232	0.7729
	P	0.7396	0.5902	0.2213	< 0.0001	< 0.0001	–	< 0.0001	< 0.0001
Total solids	r	-0.0488	-0.0662	-0.0549	0.1328	0.9282	0.7232	–	0.6891
	P	0.0989	0.0261	0.0624	< 0.0001	< 0.0001	< 0.0001	–	< 0.0001
Non-fat dry solids	r	-0.0172	-0.0414	-0.0313	0.1102	0.0042	0.7729	0.6891	–
	P	0.5602	0.1630	0.2878	0.1174	0.3713	< 0.0001	< 0.0001	–

TBC: total bacterial count; PC: psychrotrophic count; CC: coliform count; SCC: somatic cell count

Discussion

Milk microbiological analysis

In this study milk samples used for microbiological culture were frozen for a maximum period of seven days before microbiological procedures. This procedure was done in order to increase the number of dairy farms included in this study, because fresh milk samples were not available due to logistic restrictions. Therefore, a limitation of the present study is that both CC and PC could be affected by the freezing of milk samples for seven days before analysis (HUBÁČKOVÁ; RYŠÁNEK, 2007) and therefore caution should be taken when comparing results from other studies that did not use frozen samples.

In the present study herds were distributed into three classes of SCC from bulk tank milk: < 250,000 cells/mL; > 250,001 and < 750,000 cells/mL; and > 750,001 cells/mL. Herds with low SCC (< 250,000 cells/mL; 129,900 CFU/mL) had lower average of TBC than both herds from other groups with medium (250,001 and < 750,000 cells/mL; 203,700 CFU/mL) and high SCC (> 750,001 cells/mL; 159,400 CFU/mL). Jayarao et al. (2004), in a study using different SCC classes of bulk tank milk (< 200,000; 200,000-400,000; > 400,000 cells/ml), reported a proportion of TBC according to the increase of SCC in bulk tank milk: TBC = 2,290 CFU/mL in herds with SCC < 200,000 cells/mL, TBC = 4,140 CFU/mL in herds that presented SCC from bulk tank between 200,000 and 400,000 cells/mL, and TBC = 5,970 CFU/mL in herds with SCC ≥ 400,000 cells/mL.

Analysis of hygienic indicators of bulk tank milk (*e.g.,* TBC, PC and CC) is an important tool to evaluate raw milk quality (ZUCALI et al., 2011). Hygiene conditions of milking equipment and bulk tank, subclinical mastitis prevalence at herd level (HAYES et al., 2001; BERRY et al., 2006), and hygiene score of teats and udder before milking are factors associated with hygienic quality of raw milk (SCHREINER; RUEGG, 2003). Zucali et al. (2011) reported that herds with > 50% of cows that presented hygiene score 3 (moderately dirty) and 4 (very dirty) were associated with higher values of TBC, PC, and CC from bulk tank milk than herds with cows presenting lower scores of teats and udder hygiene**.** Although mastitis is one of the factors that contribute to contamination of bulk tank milk, deficiencies of hygiene during milking and milk processing have a huge influence on hygiene indicators, more than contamination caused by microorganisms from the mammary gland (SOUTO et al., 2008).

There were no differences ($p = 0.0919$), in relation to PC, between groups with low, medium and high SCC (PC = 83,000 CFU/mL, 144,200 CFU/mL and 160,900 CFU/mL, respectively). These results are different from those reported by Jayarao et al. (2004), which described that medium values of PC were proportional to the SCC increase: PC = 6,540 CFU/mL in herds with SCC < 200,000 cells/mL, PC = 10,440 CFU/mL in herds that presented 200,000 < SCC < 400,000 cells/mL, and PC = 14,960 CFU/mL in herds that showed SCC of the bulk tank ≥ 400,000 cells/mL.

Usually, high coliform count in raw milk is indicative of poor hygiene during milking, on milking equipment, and on bulk tank, but cases of mastitis caused by coliforms can also result in CC increase of bulk tank (HOGAN; SMITH, 2012; SCHUKKEN et al., 2012). Furthermore, CC is used as an indicator of contamination by fecal microorganisms (BAVA et al., 2011). In the present study herds with SCC > 250,001 cells/mL presented higher CC than herds with SCC < 250,000 cells/mL, as observed by Pantoja et al. (2011). In this study higher CC in herds with higher SCC may suggest a possible occurrence of mastitis caused by coliforms or deficiencies in milking management in herds with SCC > 250,000 cells/mL.

Herds with low SCC (< 250,000 cells/mL; 163 CFU/mL) had lower average of CC than both herds from other groups with medium (250,001 and < 750,000 cells/mL; 268.4 CFU/mL) and high SCC (> 750,001 cells/mL; 323.4 CFU/mL). Similarly to the results of this study, Jayarao et al. (2004) observed increased CC when bulk tank SCC was increased. However, results of CC showed by Jayarao et al. (2004) were lower than those observed in the present study (30, 60, and 70 CFU/mL). Although in the present study hygiene score of cows, housing conditions, and hygiene of milking equipment and on bulk tank were not evaluated, the higher level of CC observed in our study in comparison with those obtained by Jayarao et al. (2004) may be due to the presence of cows with mastitis caused by coliforms, failures in cleaning of milking equipment, and hygiene problems during the housing of cows and during the milking process (JAYARAO et al., 2004; PANTOJA et al., 2011). Additionally, Elmoslemany et al. (2009a) described that the temperature of the solution used to clean milking equipment and poor water quality are factors that negatively affect the hygienic quality of milk, because bacteria proliferation on internal surface of the equipment is facilitated and could contaminate bulk tank milk.

In the present study the effect of sampling period was observed on all analyzed indicators of hygiene (TBC, PC, and CC) and significant interaction was observed between the classes of SCC and the period of sampling in relation to TBC and to CC. Changes on TBC, PC, and CC may occur through contamination of the bulk tank milk by bacteria on the skin of teats and udder, contamination of milking equipment caused by failures cleaning and sanitation, and by the occurrence of mastitis (OLECHNOWICZ; JAKOWSKI, 2012). Furthermore, the effect of sampling time in the present study may have been the consequence of the seasons of the year and climate changes, which can interfere with hygienic quality and on SCC of bulk tank milk (FOX, 2009). In hot and rainy climates, temperature and humidity increase and favor the proliferation of environmental contamination, increasing the risk of intramammary infections and contamination of bulk tank milk (GREEN et al., 2006; ARCHER et al., 2013). In the present study, although climate variables have not been evaluated because of the wide location of farms, collections were made from August to November, the period from late winter and early spring in Brazil. In this way, in addition to hygienic failures on milking and cleaning of milking equipment, the time of collection may also be influenced by seasonality (ZUCALI et al., 2011).

Bulk tank milk composition

Although previous studies have described reduction of the contents of the major milk components according to the increased bulk milk SCC (AULDIST et al., 1998), in the present study herds with SCC > 250,001 cells/mL had higher contents of fat and protein. Najafi et al. (2009) observed that when SCC increased there was a reduction in the content of fat and an increase in protein. According to Najafi et al. (2009), increased milk protein content may occur by the enhancement of concentration of serum proteins in milk from cows with higher SCC, however, a reduction of casein contents in milk according to the increased SCC is also observed.

As observed in the present study, Machado et al. (2000) and Ribas et al. (2004) described increased contents of fat and protein in milk with higher SCC. In milk from herds with higher SCC increased levels of fat and protein can be observed due to the reduction of milk production that occurs when SCC increases (ALMEIDA et al., 1996). If the reduction of milk production was steeper than the decrease of the production of fat and protein, total solids are concentrated in bulk tank milk (RIBAS et al., 2004). Despite the fact that the present study did not estimate individual production of milk to evaluate whether the higher concentration of solids was due to reduced production, this is a possible explanation for the increased contents of fat and protein observed in herds with SCC > 250,000 cells/mL as suggested by Pereira et al. (1999).

The contents of total solids were higher in herds with SCC > 250,000 cells/mL. Similar results were reported by Ribas et al. (2004) and Pereira et al. (1999), as they observed proportional increased total solid contents in herds with higher SCC. According to Ribas et al. (2004), the increase of milk total solids can be explained by the augmenting of the contents of fat with the increase of SCC, due to high correlation between the contents of fat and total solids, which were also observed in the present study. However, Machado et al. (2000) did not report differences for the contents of total solids with the increased of SCC from bulk tank milk. Other factors may affect the milk total solids independently of SCC – for instance, when milk samples were not appropriately collected.

In the present study, an effect of sampling time was observed on contents of fat ($p < 0.0001$), protein ($p < 0.0001$), total solids ($p < 0.0001$), and SNF ($p = 0.0010$), but no interaction between the classes of SCC and the time of sampling was observed. Contents of fat, protein, total solids, and SNF were lower in the last two samplings, which occurred in September and November (springtime in Brazil), in relation to the first sampling, which occurred in August and September (late winter in Brazil). Similar results were reported by Ribas et al. (2004), which observed higher contents of total solids in winter than in summer in Brazil. According to these studies, these differences in milk composition may occur due to the temperature changes observed in this period, which can directly influence dry matter intake of cows and forage availability. Moreover, according to Forsbäck et al. (2010a), even healthy cows showed variations in the contents of fat and milk protein during lactation. Forsbäck et al. (2009) reported that changes in milk composition in cows can interfere with the composition of bulk tank milk and this may explain the effect of the time of sampling observed in the present study.

Correlation between composition and hygiene indicators of bulk tank milk

Considering the hygienic indicators of bulk tank milk, there was a positive correlation ($r = 0.6215$) between TBC and PC. This result corroborates those reported by Jayarao et al. (2004), which described medium and positive correlation ($r = 0.619$) between TBC and PC from bulk tank milk samples. However, Cempírková (2002) reported results higher ($r = 0.92$) than those observed in the present study for the correlation between PC and TBC from bulk tank milk and this correlation was probably higher since in the current study PC may be affected by the freezing of milk samples. The positive correlation between PC and TBC on bulk tank milk can occur because Psychrotrophic bacteria might adapt and proliferate in environments with the same temperature of milk refrigeration. According to Cempírková (2002) and Bava et al. (2011), storage of milk on temperature of refrigeration for over 48 hours can increase by up to 100% the Psychrotrophic counts in raw milk.

Low and positive correlation was observed between TBC and CC ($r = 0.2949$), and between PC and CC ($r = 0.3692$). Arcuri et al. (2006) also observed medium and positive correlation ($r = 0.61$) between TBC and CC. Positive correlation between TBC and CC may indicate poor hygiene during milking or deficiencies in cleaning the milking equipment and tank. The presence of coliforms is an indicator of fecal and environmental contamination (ZUCALI et al., 2011), so the correlation between TBC and CC indicates the negative impact of hygiene during milking on the TBC of raw milk. On the other hand, the positive correlation found between PC and CC may have occurred because some coliforms, such as *Citrobacter* and *Klebsiella*, are Psychrotrophic microorganisms. Although this study did not identify the bacterial species in the milk, the presence of these microorganisms classified as coliforms and Psychrotrophics may have contributed to the increase of the correlation between PC and CC (MARTIN et al., 2011).

Najafi et al. (2009) assessed the effects of SCC on fat and protein of bulk tank milk and observed low and negative correlation ($r = -0.13$) between SCC and fat, and medium and positive correlation ($r = 0.39$) between SCC and protein. These results and the results obtained in the present study differ in relation to the correlation between SCC and fat ($r = 0.2217$) and are similar in relation to SCC and protein ($r = 0.2346$). The increased content of milk protein that occurred in high SCC milk can be explained by the change in milk composition in response to inflammation in the mammary gland, and consequently the increase in concentration of serum proteins in milk (ÅKERSTEDT et al., 2012). Machado et al. (2000) reported that milk from tanks with SCC > 1,500,000 cells/mL presented higher content of fat than milk from herds with SCC < 1,000,000 cells/mL, and herds with SCC < 500,000 cells/mL presented higher content of protein than herds with SCC > 500,000 cells/mL.

In the present study, low and positive correlation between SCC and the raw milk hygiene indicators (TBC and CC) and between SCC and milk composition (fat, protein, total solids) from tank were observed. Cows affected by subclinical mastitis are one of the reasons for positive correlation between SCC and hygiene indicators. Milk from cows with mastitis has high SCC and also increased TBC and CC of bulk tank milk, depending on the causative pathogen mastitis (LAKIC et al., 2011). This increased TBC and CC of bulk tank milk occurs because infected mammary gland is one of the possible contamination sources of bulk tank milk. In this study, positive correlation between SCC and milk composition (fat, protein, and total solids) was observed. According to Pereira et al. (1999), the increase in the contents of fat, protein, and total solids in high SCC milk can occur by the concentration of solids and consequently by the increase in the content of these components on bulk tank milk.

According to Hayes et al. (2001) and Jayarao et al. (2004), the effects of SCC on TBC depends on the size of the herd and the number of cows with mastitis. However, in the present study the size of the herd and the frequency of intramammary infections were not evaluated. Some studies (HARRIS; KOLVER, 2001; BOUYAI et al., 2012; KOECK et al., 2012) suggest that the breed of dairy cow would be another factor interfering on the composition and production of the bulk tank milk and also on some variables evaluated in this study (TBC, SCC, and composition); however, no breed information of the studied herds was recorded.

Conclusion

As a consequence of lower milk yield due to elevated values of SCC, herds with medium and high SCC presented higher contents of fat, protein, and total solids than herds enrolled in the current study with low SCC. On the other hand, herds that presented low SCC had better hygienic quality of bulk tank milk than herds with medium and high SCC. Linear correlation was observed between indicators of hygiene (TBC, PC, and CC) and contents of fat and between SCC and milk composition variables (fat, protein, total solids, and SNF).

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