QUALITY OF BUFFALO MILK SUPPLEMENTED WITH SELENIUM

ABSTRACT – This study aimed at evaluating the effects of a selenium enriched diet on the composition and somatic cell count of buffalo milk, along with verifying selenium residue in milk and in Minas fresh cheese. Data from 2264 Murrah buffalo milk samples belonging to Tapuio Ltda., located in the agreste region of Rio Grande do Norte were collected in the period from 2010 to 2014 for analysis. To verify the amount of selenium residue in buffalo milk and in Minas fresh cheese, 100 Murrah buffaloes were used and divided into 5 distinct treatment lots, according to milk production (0.08 ppm/Se/kg of concentrate). Three hundred mL of milk from each lot were collected from the tanks, as well as 300g of Minas fresh cheese, from August to November 2014, with collection of the treated lots held only in the month of November. Selenium supplementation reduces somatic cell count in buffalo milk. Selenium residue was not detected in buffalo milk or cheese. Studies with higher levels that 4.8 ppm of selenium in the diets of dairy buffaloes are recommended.

Keywords: dairy buffalo nutrition; milk production; somatic cell count.

INTRODUCTION

Buffalo milk has some physical-chemical peculiarities when compared to cow's milk, including higher levels of fat and protein, sweeter taste and a white opaque color (OLIVEIRA, 2014; PATIÑO et al., 2011; PIGNATA et al., 2014). Moreover, buffalo's milk has a high content of Ca, Fe, P, and vitamins A, C and B6, along with lower levels of vitamin E, riboflavin and cholesterol (ARAUJO et al., 2012; EL-SALAM; EL-SHIBINY, 2011; EL-SALAM & EL-SHIBINY, 2013; MEDHAMMAR et al., 2012).

The increase of mastitis cases in buffalo milk is associated with increase in buffalo milk production in recent years. Aiming to measure the degree of infection, somatic cell count (SCC) is a severity indicator of the inflammatory process, being the usual parameter to assess udder health in relation to milk quality, and for the monitoring program of mastitis control (AMARAL et al., 2004; AMARAL et al., 2005; MORONI et al., 2006; RHODA et al., 2012; RUEGG, 2011). Average values for buffalo milk SCC can vary; 200

thousand/cells/mL is used as the threshold value for the identification of subclinical mastitis (SOLLECITO et al., 2011; TRIPALDI et al., 2010).

In recent years, numerous efforts have been made to stimulate the immune capacity of the mammary gland by increasing the organisms' natural defense mechanisms in an attempt to reduce the incidence of mastitis (SALMAN et al., 2009). Therefore, studies point to a reduction in the incidence of mastitis when using selenium, supported by the negative correlation between somatic cell count (SCC) and the status of the supplemented animals (CORTINHAS et al., 2010; HOGAN et al., 1993; KRUZE et al., 2007; PASCHOAL et al., 2003; SALMAN et al., 2009; SÁNCHEZ et al., 2007).

The supply of Zn, Cu and Se have been associated with a reduction in SCC and an increase in the antioxidant capacity of the enzyme superoxide dismutase (CuZnSOD), ceruloplasmin (CP) and glutathione peroxidase (GSH-Px) (WEISS; HOGAN, 2005; WEISS; WYATT, 2002), and the high concentration of salts in blood plasma was associated with a decrease in incidence of clinical mastitis and lower SCC in the tank (WEISS et al., 1990).

Most recent studies confirm that levels of Se (organic and inorganic) higher than those recommended for animals can maximize natural defense mechanisms, thus increasing resistance to diseases, especially immune function (ALVARADO et al., 2006; GUYOT et al., 2007; MCKENZIE et al., 1998; RAYMAN, 2000; SALMAN et al., 2009).

In addition to reducing mastitis and improving immunity, Se can be incorporated into milk and to promote human health. The maximum concentration of Se allowed to prevent human health problems in milk is 0.14ppm (FDA, 2003). Ceballos et al. (2009) evaluated 42 studies published between 1970 and 2008 and reported that dietary Se supplementation resulted in an increase of 12.6 μg of Se/L of milk.

The importance of selenium in the human diet is well established, since it is an essential element and its determination has fundamental value; this mineral strengthens the immune system, acting as an antidepressant agent and protecting against cancer. However, it is understood that the benefits of increased consumption of this mineral

through fortified dairy products are yet unknown (KIRA; MAIHARA, 2005; STAGSTED et al., 2005).

Thus, the objective of this study was to evaluate the effect of selenium supplementation on the physical and chemical composition and somatic cell count of buffalo milk, and to verify selenium residue in milk and in Minas fresh cheese.

MATERIAL AND METHODS

The experiment was conducted Tapuio Agropecuaria Ltda., in the municipality of Taipu, 50 km from Natal, located in the Agreste region of the State of Rio Grande do Norte, Brazil. The climate, according to Köppen classification is characterized by an *As* climate, meaning it is warm with two distinct seasons: summer (rainy) and winter (dry), with the dry season from August to January and rainy season from February to July. The average rainfall is 855 mm per year, the average temperature is 25.3°C and average relative humidity of 79.0%.

The animals were grazed in pasture under Voisin type rotational stocking, with the predominant pastures being *Brachiaria brizantha* and *Panicum maximum cv. Massai*. In the dry season, the animals' diets consisted of a supply of corn, soybean meal and soybean oil concentrate, along with sugarcane (*Saccharum officinarum*) supplemented with 1% of urea + ammonium sulfate (9:1), in troughs located inside the paddocks. The supplementation with Sel-Plex® organic selenium was performed by adding 0.08 ppm/kg/Se to the concentrate at levels of 1.6 ppm/kg/Se; 2.4 ppm/kg/Se; 3.2ppm/kg/Se; 4.0 ppm/kg/Se and 4.8 ppm/kg/Se.

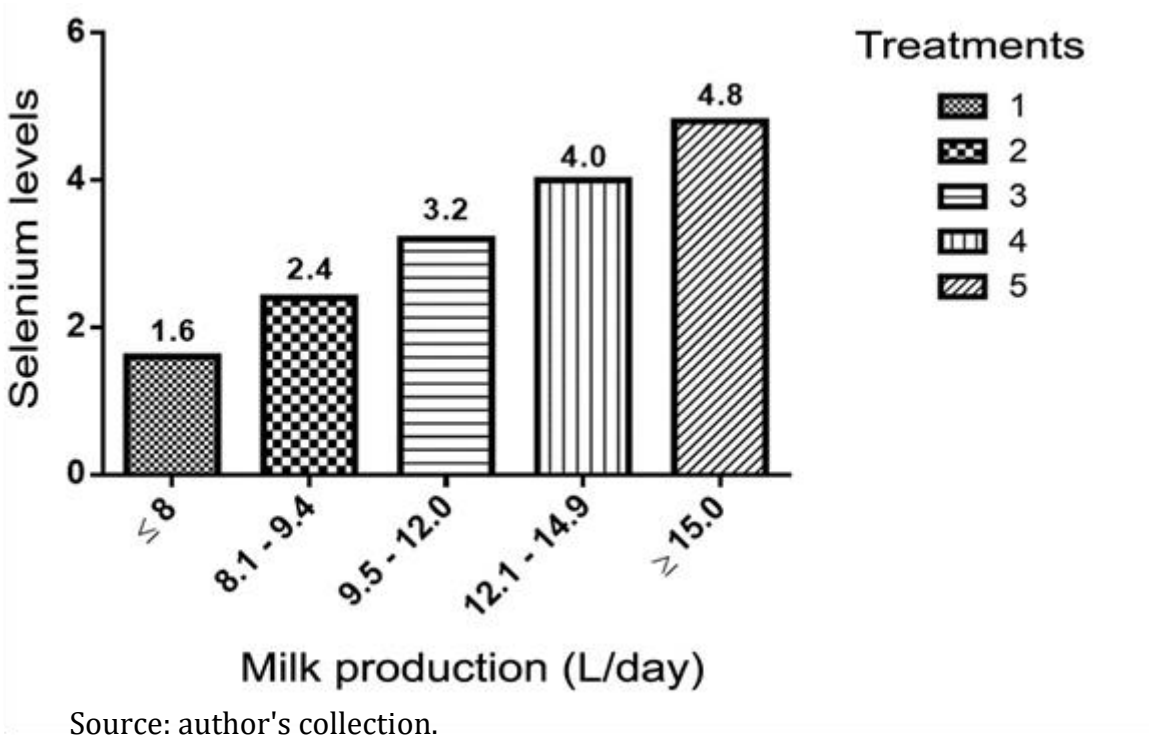
The type of Se used (Sel-Plex®) is a product biosynthesized by yeast containing selenium in the same manner found in nature, which includes the selenoamino acids and related compounds which are ideal for the mineral's absorption and metabolism.

The pre-milking environment consisted of a waiting room's covered with shading, cobblestone floor and water supply. Buffaloes were mechanically milked at 5am and at 3pm, with the adoption of all the procedures of good milking practices, such as the use of

pre- and post-dipping. The milking equipment was a double 20, single line type, with a low line in closed circuit. Milkings were conducted without the presence of calves.

The buffaloes received the concentrate during milking. The formation of the treatments was made according to the lactation duration of the animals and the available amount of concentrate varied in relation to buffalo milk production, as shown in figure 1.

Figure 1. Supplementation according to milk production.



Source: author's collection.

The data used for the analysis of fat, protein and somatic cell count (SCC) were derived from livestock control spreadsheets from the production facility, with daily records of individual information on the buffaloes from April 2010 to June 2014. A total of 2,264 individual milk analysis for all the five milk production level from the total of lactating Murrah buffaloes cows were used.

Milk samples were collected monthly, directly from the meter attached to the milking machine, comprised of samples from morning and afternoon milkings, and packaged in plastic bottles of 40 mL containing Bronopol® (2-bromo2nitropropano-1,3diol).

Samples were homogenized for complete dissolution of the preservative, identified and packed in isothermal box with ice to maintain the temperature below 5°C. Then they were sent to the laboratory of the Dairy Herd Management Program of the Northeast - PROGENE, accredited to the Brazilian Network of Milk Quality (RBQL), part of the National Program for Milk Quality Improvement (PNQL) at the Federal Rural University of Pernambuco (UFRPE). To determine the fat (%) and protein (%) content, the analyzes were performed using infrared absorption Bentley 2000® equipment (Bentley Instruments Inc., Chasca MN, USA) and SCC by flow cytometry using Somacount 300® equipment (Bentley Instruments Inc., Chasca MN, USA).

The experiment to determine selenium (Se) residue was conducted during the dry season of August to November, with collection of tank milk samples and Minas Frescal cheese. To sample the production of milk within each lot, 20 animals were randomly selected in November, 2014.

Each of the animal milk samples were collected in November, directly from the meter, just after the end of the evening milking in plastic 40 mL vials. The vials were properly identified and packed in an isothermal box with ice to maintain the temperature below 5°C, and a homogeneous sample of each batch was kept in 300 mL plastic vials.

Milk from the tanks and Minas fresh cheese collection was carried out from August to November 2014, where the collected cheese was made with the same milk from the tank. Milk from the tanks was transferred to properly identified standard 300 mL vials, and cheese supplied by the property was vacuum packed, weighing 300 gr/each. The collected milk and the samples for each lot were frozen at 0°C.

Milk samples from the tank for each treatment and cheese samples were sent to the Institute of Technology of Pernambuco (ITEP) in Recife - PE, to carry out Selenium residue analysis. The analysis was conducted with a Thermo Scientific® model ICAP 6300 CID optical emission spectrometer with inductively coupled plasma (ICP-OES), by employing simultaneous detection with axial and radial view, a thermally stable polychromator, a radio frequency generator of solid state high capacity equipped with a

concentric nebulizer, and following the methodology indicated by the American Public Health Association (1999).

In order to learn about the quality of consumed forage, the collections were performed on the first Tuesday of the month, in the period from August to November 2014, By hand plunked the forage at the same grazing height to simulate the animal selectivity. In the paddocks which had an average area of 0.8 hectares, we collected four simple samples on site at the time of grazing, obtaining a properly mixed sample. Grazing close to the road and salt troughs were not considered. The concentrated sample was performed on the same day as the pasture collection with the aid of a calador. Concentrate samples were collected monthly in triplicate. Then these samples were sent to the Animal Nutrition Laboratory of the Federal University of Rio Grande do Norte (UFRN).

The methodology described by INCT-CA (2012) was used for determining the content of dry matter, mineral matter, crude protein, ether extract, neutral detergent fiber, acid detergent fiber, insoluble nitrogen levels in neutral detergent and acid detergent and the food and concentrate lignin (Tables 1 and 2).

Table 1. Proportions and chemical composition of the concentrate offered to animals.

Ingredients				Proportions (%)
Soybean meal				50.59
Ground corn				36.55
Soy oil				4.95
Urea				1.00
Mineral mix				6.91
	Chemical composition %			
	August[1]	September[1]	October[1]	November[1]
Dry Matter	91.65	91.68	91.38	91.49
Mineral Matter	9.72	7.29	9.60	9.31
Organic Matter	90.28	92.71	90.40	90.69
Crude Protein	23.42	22.17	28.73	25.72
Ether Extract	8.65	6.63	8.34	8.10
NDF	41.18	35.16	29.46	30.76
ADF	6.04	13.72	7.91	6.48
Hemicellulose	35.14	21.44	21.55	24.28
Total Carbohydrates	58.21	63.91	53.33	56.87
NFC	17.03	28.75	23.87	26.11
Lignin	1.95	3.26	2.04	0.92
Cellulose	4.09	10.46	6.70	5.56
NDIP	0.46	0.70	0.91	0.24

ADIP	0.07	0.53	0.16	0.03
TDN	75.54	74.35	62.81	80.32
DE(Mcal/Kg)	3.33	3.28	2.77	3.54

NDF – Neutral Detergent Fiber; ADF - Acid Detergent Fiber; NFC – Non-Fibrous Carbohydrates; NDIP - Neutral Detergent Insoluble Protein; ADIP - Acid Detergent Insoluble Protein; TDN - Total Digestible Nutrients; DE - Digestible Energy.

Table 2. *Panicum maximum* cv. Massai chemical composition.

Parameters	August[a]	September[b]	October[b]	November[c]
Dry Matter	32.22 ± 5.39	51.52 ± 1.86	38.98 ± 5.61	49.64 ± 1.70
Mineral Matter	7.60 ± 0.71	7.32 ± 1.40	7.80 ± 0.72	7.17 ± 0.72
Organic Matter	92.40 ± 0.71	92.67 ± 1.40	92.67 ± 1.23	92.83 ± 0.55
Crude Protein	5.04 ± 0.85	3.33 ± 0.79	4.17 ± 0.90	4.44 ± 0.78
Ether Extract	1.35 ± 0.12	1.42 ± 0.19	1.88 ± 0.07	1.89 ± 0.45
NDF	75.69 ± 1.90	81.39 ± 1.90	78.85 ± 1.09	75.08 ± 0.34
ADF	43.43 ± 0.80	49.51 ± 1.52	47.08 ± 1.74	45.94 ± 1.27
Hemicellulose	32.25 ± 1.24	31.31 ± 1.96	30.19 ± 3.13	29.14 ± 0.92
Total Carbohydrates	84.25 ± 4.06	86.62 ± 1.40	86.62 ± 1.85	86.50 ± 0.22
NFC	10.38 ± 1.70	5.99 ± 1.03	8.38 ± 1.15	11.42 ± 0.57
Lignin	8.04 ± 0.56	12.47 ± 1.55	17.18 ± 1.99	10.16 ± 0.61
Cellulose	35.38 ± 0.66	37.04 ± 1.27	33.16 ± 4.92	35.77 ± 0.65
NDIP	0.28 ± 0.05	0.22 ± 0.03	0.33 ± 0.04	0.32 ± 0.08
ADIP	0.06 ± 0.01	0.11 ± 0.01	0.07 ± 0.02	0.09 ± 0.00
TDN	50.58 ± 3.98	39.81 ± 3.55	36.98 ± 2.57	46.56 ± 0.97
DE(Mcal/Kg)	2.23 ± 0.17	1.60 ± 0.36	1.57 ± 0.16	2.05 ± 0.04

NDF – Neutral Detergent Fiber; ADF - Acid Detergent Fiber; NFC – Non-Fibrous Carbohydrates; NDIP - Neutral Detergent Insoluble Protein; ADIP - Acid Detergent Insoluble Protein; TDN - Total Digestible Nutrients; DE - Digestible Energy.

Different mineral levels were considered for each treatment for the data analysis of levels of selenium (Se), somatic cell count (SCC), fat, protein and somatic cell score (SCS) (Figure 1).

Based on the calving data, lactating days (LD) were calculated from the average deviation of variance and distributed into four classes: the first class up to 280 lactating days (< 280); the second class between 281 and 305 (281 < x < 305) lactating days; the third class between 306 and 350 (306 < x < 350) lactating days; and the fourth up to 351 lactating days (> 351).

The effect of the seasons was contrasted in two ways: Spring (September 21 to December 20), Summer (December 21 to March 20), Autumn (March 21 to June 20), and winter (June 21 to 20 September), or Dry season (August to January) and Rainy season (February to July).

Values obtained for SCC were transformed into Somatic Cell Score (SCS) using the Equation 1: SCS = log2 (SCC/100,000) + 3. This procedure is intended to circumvent the fact that SCC did not present normal distribution. The following procedures were performed: descriptive analysis, analysis of variance and correlation analysis using the Statistical Analysis System - SAS (2002), and averages were compared by Tukey test at 5.0% probability.

RESULTS AND DISCUSSION

The results in Table 3 show the overall average of the physical and chemical composition, somatic cell count (SCC) and somatic cell score (SCS) of buffalo milk. The quality standard for buffalo milk does not yet exist, however, the literature shows low scores when compared to cow's milk. Cerón-Muñoz et al. (2002), when evaluating the SCC from a sample of 1,630 Murrah buffaloes in São Paulo, obtained an average of 79 thousand/cells/mL.

Table 3. Adjusted averages of buffalo milk composition and sanitary quality.

Characteristics	N	Average ± SD	CV	Min	Max
Fat (%)	2264	5.92 ± 1.61	27.23	1.61	10.16
Protein (%)	2264	4.22 ± 0.43	10.22	3.09	5.35
SCC[1] (thousand/cel/mL)	2264	92.88 ± 178.37	192.05	0.10	990.00
SCS[2] (log cel/mL)	2264	1.47 ± 1.82	124.12	0	6.31

[1] – Somatic Cell Count; [2] - Somatic Cell Score; Information number (N), Average, Standard Deviation (SD), coefficient of variation (CV), minimum value (Min), maximum value (Max).

Few SCC studies in buffalo milk have been conducted in Brazil and in the Northeast, almost nothing is known about this parameter for assessing the health of the mammary gland. Often the SCC parameter for cattle that is used may not be suitable for monitoring mastitis in buffalo cattle (MEDEIROS et al., 2011). Thus, greater SCC in buffaloes than in

cows may not be indicative of mastitis (COSTA FILHO et al., 2015). Thus, it is urgent to develop a specific legislation for the sanitary quality of buffalo milk.

In buffaloes in the Lazio region in Italy, Tripaldi et al. (2010) recommended the amount of 200 thousand/cells/mL as the limit for the early identification of an animal affected by subclinical mastitis. While in Brazil, Medeiros et al. (2011) reported values above 280 thousand/cells/mL being indicative of infection of the mammary gland. However, these authors reported that the microbiological examination of milk is the best method for diagnosing subclinical mastitis in buffaloes. In this study, we evaluated the 2,264 data of the chemical composition and sanitary quality of buffalo milk, which found an average of 92.88 thousand/cells/mL, below the indicative threshold of infection as quoted by the authors above. From the amount of data analyzed in this experiment, it is possible to define a standard for the sanitary quality of buffalo milk.

Somatic cell score facilitates the interpretation of results. In this experiment an average of 1.47 (log/cell/mL) was observed. In a study by Barreto et al. (2010), a negative significant linear correlation ($p < 0.05$) was found between SCS and milk production variables (-0.32).

Lima et al. (2014) found an average of 5.57% fat and 4.22% protein working with the same herd evaluated in this study. These values are similar to those found in the present study (5.92% and 4.22%), respectively. According to Fernandes et al. (2011), in studies conducted in the state of Minas Gerais, the level of fat in buffalo milk varies between 5.5 and 10.4%, and according to Teixeira et al. (2005), protein varies between 3.6 and 5.26%.

The inclusion of selenium reduces fat (%) and protein (%) content and somatic cell count (thousand/cell/mL) in all lactating periods evaluated (Table 4).

Table 4. Comparison of the averages of lactations in each treatment, for composition and sanitary quality of buffalo milk.

< 280 lactating days					
Selenium levels (ppm)	N	Fat (%)	Protein (%)	SCC[1] (thousand/cel/mL)	SCS[2] (log cel/mL)

1.6	350	6.67 [a]	4.38 [a]	138.26 [a]	2.19 [a]
2.4	97	6.13 [ab]	4.22 [ab]	102.72 [ab]	1.60 [ab]
3.2	180	5.91 [ab]	4.11 [bc]	69.37 [ab]	1.23 [bc]
4.0	112	5.40 [bc]	4.07 [bc]	54.40 [b]	0.88 [c]
4.8	38	4.98 [c]	3.98 [c]	37.61 [b]	0.79 [c]
281 and 305 lactating days					
Selenium levels (ppm)	N	Fat (%)	Protein (%)	SCC[1] (thousand/cel/mL)	SCS[2] (log cel/mL)
1.6	249	6.63 [a]	4.50 [a]	141.90 [a]	2.10 [a]
2.4	89	6.06 [ab]	4.11 [b]	101.54 [ab]	1.49 [ab]
3.2	131	5.81 [ab]	4.16 [b]	89.00 [ab]	1.43 [abc]
4.0	75	5.31 [bc]	4.17 [b]	43.71 [b]	0.99 [bc]
4.8	38	4.94 [c]	4.03 [b]	29.19 [b]	0.67 [c]
306 and 350 lactating days					
Selenium levels (ppm)	N	Fat (%)	Protein (%)	SCC[1] (thousand/cel/mL)	SCS[2] (log cel/mL)
1.6	302	6.74 [a]	4.37 [a]	147.63 [a]	2.07 [a]
2.4	86	6.37 [ab]	4.16 [ab]	83.77 [ab]	1.31 [ab]
3.2	137	5.57 [bc]	4.14 [ab]	78.95 [ab]	1.22 [b]
4.0	102	5.55 [bc]	4.11 [b]	66.14 [b]	0.98 [b]
4.8	40	5.42 [c]	4.01 [b]	61.07 [b]	0.94 [b]
> 351 lactating days					
Selenium levels (ppm)	N	Fat (%)	Protein (%)	SCC[1] (thousand/cel/mL)	SCS[2] (log cel/mL)
1.6	305	6.97 [a]	4.49 [a]	136.96 [a]	1.97 [a]
2.4	75	6.22 [ab]	4.42 [ab]	111.99 [ab]	1.75 [ab]
3.2	99	5.73 [b]	4.28 [ab]	65.76 [ab]	1.11 [bc]
4.0	31	5.55 [b]	4.06 [b]	51.95 [ab]	0.98 [bc]
4.8	99	5.45 [b]	4.05 [b]	42.76 [b]	0.76 [c]

[1] – Somatic Cell Count; [2] - Somatic Cell Score; Averages in the same column followed by the same letter do not differ from each other at a 5% significance by Tukey test ($p < 0.05$).

The results of organic mineral supplementation on milk production and composition reported in the literature vary considerably. Some authors report the effects of organic trace mineral supplementation on milk production with no change in their composition (BALLANTINE et al., 2002; GRIFFITHS et al., 2007; KINAL et al., 2007; SICILIANO-JONES et al., 2008).

According to Cortinhas et al. (2010) the supply of organic Se in dairy cows had no effect on milk yield and composition, however, it promoted a reduction in both the somatic cell count as well as the incidence of subclinical mastitis. Paschoal et al. (2006) found no

effects of Se supplementation on SCC or immune response, and credited this lack of effect being related to low levels of Se (2.5 mg Se/day). In 2003, the same authors used a dose of 5 mg Se/day, obtaining a reduction in SCC.

Seasons of the year influenced ($P < 0.05$) milk composition and SCC (Table 5).

Table 5. Average somatic cell count (SCC), somatic cell score (SCS), fat and protein in relation to the seasons.

Characteristics	Season			
	Summer	Autumn	Winter	Spring
N	363	765	591	545
Fat (%)	5.99 ^{ab}	5.84 ^{b}	5.78 ^{b}	6.13 ^{a}
Protein (%)	4.38 ^{a}	4.19 ^{bc}	4.16 ^{c}	4.24 ^{b}
SCC[1] (thousand/cel/mL)	164.53 ^{a}	45.88 ^{c}	92.54 ^{b}	111.50 ^{b}
SCS[2] (log cel/mL)	2.27 ^{a}	0.95 ^{c}	1.49 ^{b}	1.63 ^{b}

[1] – Somatic Cell Count; [2] - Somatic Cell Score. Averages in the same column followed by the same letters do not differ from each other at 5% significance by Tukey test ($P < 0.05$).

SCC was relatively low during all seasons of the year. However, higher averages ($P < 0.05$) for this parameter were found in the summer, while the lowest values were found in autumn, contrary to what would be expected considering that this month has the largest amount of rainfall in the region. Excess moisture creates favorable conditions for increased infection and prevalence of mastitis in herds. Amaral et al. (2004) reviewed the influence of the season and its relationship with SCC and found higher values in summer, a period characterized by high humidity and temperature.

Singh; Ludri (2001) and Araújo et al. (2012) found that seasons had a significant effect on the averages of SCC, being lower in the winter and in the hot and dry seasons, and higher in the hot and humid season, presenting the values 76, 108, and 135 thousand/cel/mL, respectively.

Amaral et al. (2005) reported that seasonal effects should not be considered as the main cause of SCC variation, and in fact what happens is the result of increased ubber bacterial contamination during periods in which the microbial growth conditions are

more favorable and circumstances in which contaminating factors are not avoided by good management practices. It is noteworthy that buffaloes are less susceptible to mastitis than cows for having more muscular papillary ducts with higher amounts of nerve fibers and blood vessels that are an efficient barrier against infections (DELLA LIBERA et al., 2004; KAPRONEZAI et al., 2005; LAU, 1994).

Fat content of buffalo milk had higher values in spring with an average of 6.13% and lower in winter with 5.78%. This contrasts with Costa Filho et al. (2015) when using 70 Murrah buffaloes on the same property studied in this study, which described the higher fat values in summer (6.00%) and lower in autumn (5.40%).

Protein had a higher average in the summer, (4.38%), and lower in winter with an average of 4.16%. This corroborates the work done by Costa Filho et al. (2015) which found an average of 4.28% in the summer and 4.03% in the winter.

For Amaral et al. (2005), most of the changes in milk composition between seasons are derived from different lactation stages in animals, which are due to the reproductive seasonality of the buffalo species.

The fact that the buffalo were supplemented with sugar cane with urea during the dry season, and that concentrate was offered to dairy buffaloes throughout the year, may interfere with the seasonal effect, as the milk composition varies due to various factors, in particular diet composition (AMARAL et al., 2004, LOPES, 2009).

A higher percentage of fat and protein in buffalo milk during the dry season (Table 6) can be attributed to the concentration of these components in the mammary gland due to the lower production of milk during the dry season. The effects of diet supplementation with sugarcane and urea, in addition to the concentrate, were probably not enough to meet the entire requirement of the buffaloes, which had a reduced volume of milk in the course of the period from August to January. This result agrees with the findings by Araújo et al. (2011), describing an average of 5.70% fat in the dry season. Although Andrade et al. (2011) found no differences in the levels of fat between the dry and rainy seasons.

According to Simões et al. (2014), the dry and rainy seasons in the State of Para influenced the composition of buffalo milk, with the dry period having a higher concentration of fat (6.74%) and lower protein (3.92%).

Table 6. Average somatic cell count (SCC), somatic cell score (SCS), fat and protein in relation to the season.

Characteristics	Season	
	Dry	Rainy
Fat (%)	6.05 [a]	5.79 [b]
Protein (%)	4.24 [a]	4.20 [b]
SCC[1] (thousand/cel/mL)	120.85 [a]	65.16 [b]
SCS[2] (log cel/mL)	1.78 [a]	1.16 [b]

[1] – Somatic Cell Count; [2] - Somatic Cell Score. Averages in the same column followed by the same letters do not differ from each other at 5% significance by Tukey test ($P < 0.05$).

Baruselli; Carvalho (2002) document that buffaloes are seasonal polyestrous in short days, with their estrous cycle concentrated in the autumn and winter. Thus, variations in of buffalo milk composition during the year may be due to seasonal reproductive behavior. However, in this study conducted at a site near the equator, these effects were probably more influenced by dry and rainy seasons that lead to changes in the availability and quality of forage and animal welfare, since the variation in the number of hours of sunlight per day throughout the year is very small (ZICARELLI, 2010). However, Oliveira et al. (2014), described that the Murrah buffalo are adapted to the climatic conditions of Rio Grande do Norte state, and therefore do not experience negative effects on their milk production.

SCC varied ($P < 0.05$) depending on the season, being even higher in the dry season. Ludri; Singh (2001) and Araujo et al. (2012) also found that seasons had significant effects on the average of SCC in buffalo milk.

Organic selenium supplementation has not provided ($P > 0.05$) verifiable quantities of selenium in milk or in Minas fresh cheese. To consider the presence of mineral residues

in milk and cheese, it was necessary to obtain a value greater than 0.01 mg Se/kg product. All values were lower 0.01 mg Se/kg.

In a study conducted by Kira; Maihara (2005) to determine the amount of selenium present in milk, cheese and chocolate milk, the highest values of Se were found in the buffalo cheese sample (16.1 μg/100g wet weight). Despite the importance of selenium for human consumption, it is not common to find the description of its levels in the literature of Brazil. The American Society of Enteral and Parental Nutrition suggests an increase in the recommended Se intake from 20 to 60 μg/day to 61 to 100 μg/day for adults (VANEK et al., 2012).

The concentration of selenium in cow's milk ranges from 10 to 25 μg L^{-1} (CONRAD; MOXON, 1979), being dependent on daily consumption. Ceballos-Marquez et al. (2010) reported that an increase in SCC can increase Se concentration in milk. This is due to the influx of neutrophils with high GSH-Px activity of the infected mammary gland. However, in this study, this response was not observed.

CONCLUSION

Selenium supplementation reduces somatic cell count in buffalo milk. Selenium residue was not detected in buffalo milk or cheese. Studies with higher levels that 4.8 ppm of selenium in the diets of dairy buffaloes are recommended.

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