



## Gir and Guzerat cow milk production and composition according to lactation stage, somatic cell count, physiological state and body condition

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**ABSTRACT.** This work was realized to evaluate lactation stage, physiological state, somatic cell count (SCC) and body condition score (BCS) on production and composition of cow's milk from Gir and Guzerat breeds were studied. Ninety-eight cows were analyzed and classified according to their physiological state, lactation stage, SCC and BCS. Milk production was weighed monthly to obtain production data. Composed samples were collected in vials containing preservatives and sent to the laboratory for analysis. Data were used for statistic descriptive analysis and analysis of variance. Higher milk production in the first 60 days of lactation was observed. Fat, protein and total solids concentration increased according to the increase of days in milk; while lactose concentration decreased. Higher levels of fat, protein and total solids in milk of non-pregnant cows were observed; while pregnant cows had higher daily production and higher lactose content in their milk. Higher lactose levels in milk were observed in animals with lower SCC. The BCS had no effect on milk composition or on somatic cell count. Zebu cow milk production and composition are influenced by lactation stage, physiological state and SCC.

**Keywords:** gestation; milk quality; body reserve; zebu.

## Produção e composição do leite de vacas Gir e Guzerá de acordo com o estágio de lactação, contagem de células somáticas, estado fisiológico e de condição corporal

**RESUMO.** Neste trabalho, objetivou-se avaliar o efeito do estágio de lactação, estado fisiológico, contagem de células somáticas (CCS) e escore de condição corporal (ECC) sobre a produção e composição do leite de vacas Gir e Guzerá. Foram utilizadas noventa e oito vacas, que foram classificadas de acordo com o estado fisiológico, estágio de lactação, CCS e ECC. O leite foi pesado mensalmente para obtenção dos dados de produção. Amostras compostas coletadas em frascos contendo conservante foram enviadas ao laboratório para análises. Os dados foram submetidos a análises estatísticas descritivas e de variâncias. Verificou-se maior produção de leite nos primeiros 60 dias de lactação. Os teores de gordura, proteína e extrato seco aumentaram com o avançar da lactação; enquanto o teor de lactose decresceu. Foram observados maiores teores de gordura, proteína e sólidos totais no leite de vacas vazias; enquanto as vacas gestantes apresentaram maior produção diária e maior teor de lactose no leite. Maiores teores de lactose foram verificados em animais com menor CCS. O ECC não teve efeito sobre a composição e escore de células somáticas do leite. A produção e composição do leite de vacas zebuínas sofrem influência do estágio de lactação, do estado fisiológico e da CCS.

**Palavras-chave:** gestação; qualidade do leite; reserva corporal; zebu.

### Introduction

The Brazilian herd consists mainly of Zebu cattle (Tizioto et al., 2011), being recognized for their better adaptation to tropical conditions in which they are predominantly managed, and for their ability to enable dairy farming on an exclusive grazing regime (Madalena, Peixoto, & Gibson, 2012).

Among Zebu breeds with milk aptitude, Gir and Guzerat have been selected for milk production, with reports of average yields of 8.5 kg day<sup>-1</sup> (Madruga et al., 2016), which can be seen as satisfactory results in tropical production systems (Peixoto, Verneque, Teodoro, Penna, & Martinez, 2006).

Physicochemical composition of cow milk is of great importance when it comes to nutritional value and its industrial processing ability, and varies according to several factors, among which are: breed, diet, udder health, lactation stage and physiological factors (pregnant or non-pregnant) (Aikman, Reynolds, & Beever, 2008; Dufour, Fréchette, Barkema, Mussell, & Scholl, 2011). Proteins, lipids and lactose are the constituents that vary the most during lactation (Kgole, Visser, & Banga, 2012; Sharma, Singh, & Bhadwal, 2011) generally showing higher values when lactation reaches advanced stages (Aganga, Amarteifio, & Nkile, 2002).

Somatic cell count (SCC) is used as an indirect diagnostic criterion of subclinical mastitis (Berglund, Pettersson, Östensson, & Svennersten-Sjaunja, 2007), which are normally counted at 100,000 cells/ml when milk comes from healthy mammary glands (Bytyqi et al., 2010). Higher SCC values have negative influences on the quality of raw milk, which are due to enzymatic activity of somatic cells (Sharma et al., 2011), and the main consequences are associated with reduced shelf life and sensory quality of the final dairy product, resulting in a considerable economic loss for the entire dairy industry (Geary et al., 2012).

Body condition score (BCS) is the cheapest, most practical and non-invasive method for assessing the physiological status of animals in different systems, seasons or environments (Amer, 2008; Bewley & Schutz, 2008; Mohamed, Al-Shami, & Al-Ekna, 2015), quantifying the body's deposited or mobilized reserves, and thus constituting an important tool for setting the nutritional management in lands/farms that develop dairy cattle production. Several studies have quantified the effect of BCS on milk production and composition; however, most of these have been performed with *Bos taurus taurus* cows (Mohamed et al., 2015).

Considering that most of the dairy herd consists of Zebu animals and their half-breeds in tropical regions of Brazil, and taking into consideration the lack of studies and reports dealing with the influence of the factors cited above on the chemical composition of Zebu milk, this study aimed at evaluating the effects of the lactation stage, physiological state, somatic cell count and body condition score on the production and physicochemical composition of milk from both pregnant and non-pregnant Gir and Guzerat cows.

## Material and methods

### Herd selection and location

Data used in this study came from herds of Gir (50) and Guzerat (48) breeds from the Agricultural Research Company of Rio Grande do Norte (EMPARN); which participated in progeny tests of national milk improvement programs from Zebu breeds. Both herds are raised in the same property.

The study was conducted at the Felipe Camarão Experimental Center located in São Gonçalo do Amarante, RN, 13 km away from the state capital. The farm is located in the coastal region of Rio Grande do Norte with the geographic coordinates of 5°37'18"S and 35°35'48"W, and an altitude of 41m. The climate is classified as Aw, tropical with a dry season (Köppen & Geiger, 1928). The average rainfall in the region during the trial period was approximately 1500 mm per year, with an average temperature of 26°C, and average relative humidity of 78.0%, according to data obtained from a meteorological station installed at the farm.

### Management procedures

In the experimental station, a grazing production system (150 ha area) was used with grazing under continuous stocking and individual supplementation with concentrates composed of corn, soybean meal and mineral mix, according to milk production (1 kg of concentrate for every 3 kg of produced milk). The roughage diet varied according to time of year, so that in the rainy season it was based on *Brachiaria brizantha* grazing or dwarf elephant grass (*Pennisetum purpureum Schum* cv. Mott), and in the dry period the cows received a roughage supplementation (sorghum silage or corn with elephant grass), in addition to grazing and feed supplementation.

### Sample collection and testing

Data were collected throughout 2013 from 98 cows, 50 Gir and 48 Guzerat. Information regarding the identification (registration number and name), lactation stage, age at calving, physiological state (pregnant or non-pregnant), total milk production and body condition score (BCS) were recorded for each animal.

Milking was realized twice a day, at 04:00 and at 16:00. On the day of the monthly milk weighing, individual samples of milk straight from the meter attached to the milking system were collected (one sample per cow per month, totaling 12 samples per cow along the experiment). After the end of milking, samples were duly identified and packaged in plastic

vials of 40 mL containing the chemical preservative Bronopol® (2-bromo-2-nitro-1,3 propanodiol). The vials contained milk fractions from both milking (morning and afternoon); 2/3 of the volume of the sample consisting of milk from the first milking and 1/3 from the second milking. After this procedure, the vials were placed in an isothermal cooler containing artificial ice to maintain the temperature between 2 and 6°C, and subsequently sent to a member of the Brazilian Laboratory for Milk Quality Network in Juiz de Fora (Minas Gerais).

For determining the concentration of protein, fat, lactose, and total solids, samples were analyzed by infrared absorption (Bentley 2000®; Bentley Instruments Inc., Chasca MN, EUA). Somatic cell count was determined by electronic count with Fourier transform infrared spectroscopy (FTIR), with MILKO SCAN™ (FOSS, Denmark) equipment.

For BCS evaluation, the methodology proposed by Wildman et al. (1982), adapted by Edmonson, Lean, Weaver, Farver, and Webster (1989) was used. The methodology is based on visual assessments of body reserves in specific parts of the cow's body, adopting a biological scale of 1 to 5, with 0.5 subunits, where 1 represents sub-conditioned cows (very thin) and 5 represents super-conditioned cows (very fat), regardless of weight and/or body size.

#### Data analysis

The effects of the breeds (Gir and Guzerat), physiological state (pregnant or non-pregnant), lactation stage, SCC and BCS were initially considered in the statistical data analysis of milk production and composition.

For physiological state, the animals were grouped into two classes: I - pregnant lactating cows and II - non-pregnant lactating cows. For the lactation stage, cows were classified according to the days of lactation: Class I - up to 60 days of lactation; Class II - 61 to 120 days of lactation; and class III - over 120 days of lactation. SCC was transformed into logarithm base 2 ( $\log_2$ ) to address the assumption of normality, and the following classes were then established: I - up to  $1 \times 10^5$  cells  $\text{mL}^{-1}$ ; II - more than  $1 \times 10^5$  cells  $\text{mL}^{-1}$ .

Statistical analyzes were performed using the statistical package Statistical Analysis System (SAS, 2008) version 9.2. PROC MEANS and PROC GLM procedures were used for descriptive analysis and analysis of variance, respectively. For variance analysis, the maximum likelihood method was used, while the method of least squares was used for obtaining adjusted averages. The averages were compared using the Tukey's test at 5% probability for type I error.

The following mathematical model was used:

$$Y_{ij} = \mu + \text{effect}_i + \varepsilon_{ij}$$

where:

$Y_{ij}$  = dependent variables of fat, protein, lactose, total solids, milk production, DDE and SSC;

$\mu$  = overall average;

$\text{effect}_i$  = effect of the  $i^{\text{th}}$  independent variable.

According to the following independent variables:

Breed = 1 and 2 (Gir and Guzerat).

Days of lactation categories = 1, 2 and 3 (I - up to 60 days of lactation; II - from 61 to 120 days of lactation; III - more than 120 days of lactation).

Physiological state = 1 and 2 (pregnant or non-pregnant lactating cows, respectively).

SCC class = 1 and 2 (I - up to  $1 \times 10^5$  cells  $\text{mL}^{-1}$  or II - over  $1 \times 10^5$  cells  $\text{mL}^{-1}$ ).

BCS class = 1 to 5 (1=2.0; 2=2.5; 3=3.0; 4=3.5; and 5=4.0).

The following characteristics were also included in the mathematical model only for this last independent variable: DDE = degressed dry extract; SSC = score of somatic cells.

#### Results and discussion

Difference in milk composition of the studied breeds was observed (Table 1). The average for milk constituents are close to those set by the National Program for Improvement of Zebu breeds, coordinated by Embrapa Dairy Cattle (CNPGL), averaging 4.5% fat, 3.3% protein and 12.2% total solids for the Guzerat breed; and 4.1% fat, 3.2% protein and 12.0% total solids for Gir. Ruas et al. (2007) working with crossbreeds composed of Holstein, Gir and Guzerat, found lower average milk production values during lactation for cows of Guzerat maternal genetic basis ( $6.5 \pm 2.1$  kg  $\text{day}^{-1}$ ) in comparison to those which were Gir-based ( $7.4 \pm 1.7$  kg  $\text{day}^{-1}$ ).

**Table 1.** Least squares means (LSM) and respective standard deviations (SD) of milk production and constituents.

Variables	LSM $\pm$ SD		MIN MAX		LSM $\pm$ SD		MIN MAX	
	Gir		Guzerat		Gir		Guzerat	
Fat, %	4.12 $\pm$ 0.99 <sup>a</sup>	1.79	7.20	4.22 $\pm$ 1.00 <sup>a</sup>	1.29	6.87		
Protein, %	3.26 $\pm$ 0.30 <sup>a</sup>	2.71	4.24	3.40 $\pm$ 0.34 <sup>b</sup>	2.62	4.30		
Lactose, %	4.63 $\pm$ 0.20 <sup>a</sup>	4.15	5.03	4.62 $\pm$ 0.25 <sup>a</sup>	4.04	5.11		
Total solids, %	12.93 $\pm$ 1.05 <sup>a</sup>	10.61	16.30	13.22 $\pm$ 1.19 <sup>b</sup>	10.50	16.09		
Milk production, kg $\text{day}^{-1}$	11.30 $\pm$ 5.25 <sup>a</sup>	0.50	30.30	11.25 $\pm$ 5.01 <sup>a</sup>	2.00	27.00		

Similar letters within the same column indicate that they did not differ statistically at the 5% level of significance on the Tukey's test. Min = minimum value; Max = maximum value.

Working with Creoles, Guzerat and their crosses, Velázquez et al. (2012) mentioned that the genetic

group is an important factor in determining milk production and found that Guzerat breeds positively influenced several lactation features through individual heterosis.

Higher milk production in the first 60 days of lactation was verified (Table 2). The fat, protein and dry matter concentrations increased with advancing lactation; while lactose content decreased. Regarding milk production and composition according to the lactation stage, the reduction of daily milk production and increase in fat and protein contents corroborate the reviewed literature. According to Papajcsik and Bodero (1988), unlike European, breeds Zebu or crossbred cows tend to have lactation curves that start at the peak of production, or without an inclination stage from calving to peak, and they also present low persistence curves and shorter lactation periods (Velázquez et al., 2012), which would explain the decrease in production observed at 61 days of lactation. Studying the lactation curve in crossbred, Glória et al. (2010) found that in ½ Holstein ½ Gir cows and ½ Holstein ½ Guzerat cows that production started to decline respectively at the 22<sup>nd</sup> and 27<sup>th</sup> days of lactation, demonstrating this peculiarity in milk production of zebu animals and their crossbreeds. Regarding milk composition, Galvão Júnior et al. (2010) mentioned that due to the dilution effect, protein and fat concentrations tend to be lower as milk production increases, and the opposite is also true. Thus, the increase in fat and protein levels could be explained by a reduction in milk production, which leads to increased constituents concentrations. Regarding lactose specifically, Martin and Sauvant (2007) claimed that lactose levels tend to decline after lactation peak, corroborating the findings of this research.

**Table 2.** Least squares means of milk production (MP), fat, protein, lactose and total solids (TS) content of Zebu cows according to lactation phase.

Categories	MP	Fat, %	Protein, %	Lactose, %	TS, %
I	13.34 <sup>a</sup>	3.72 <sup>a</sup>	3.52 <sup>a</sup>	4.71 <sup>a</sup>	13.51 <sup>a</sup>
II	12.01 <sup>b</sup>	4.12 <sup>b</sup>	3.49 <sup>b</sup>	4.69 <sup>b</sup>	13.74 <sup>b</sup>
III	8.71 <sup>c</sup>	4.78 <sup>c</sup>	3.66 <sup>c</sup>	4.54 <sup>c</sup>	14.54 <sup>c</sup>

Similar letters within the same column indicate that they did not differ statistically at the 5% level of significance on the Tukey's test. Categories: I up to 60 days of lactation; II from 61 to 120 days of lactation; III more than 120 days of lactation.

Regarding cows' physiological condition (Table 3), higher concentrations of fat, protein and total solids were observed in non-pregnant cow's milk, while pregnant lactating cows showed higher production and greater content of lactose in the milk. Results have shown that non-pregnant lactating cows had higher levels of milk constituents,

since they do not need to spend energy for fetal growth (Lucy, Jiang, & Kobayashi, 2001); thus, energy is only intended for milk production and maintenance. It is assumed that the largest mobilization of protein in pregnant cows is related to greater mobilization of this nutrient for the formation of fetal tissue in detriment to milk protein production, implying a reduction in the production of this constituent during the remainder of lactation (Santos, Abreu, Souza, & Catto, 2009).

**Table 3.** Means and Standard Deviation (SD) of milk composition (%) and milk production (Kg) of Zebu breeds in relation to animals physiological state.

Variables	Means ± SD	Minimum	Maximum
Non-pregnant lactating cows			
Fat, %	4.47±0.94 <sup>a</sup>	2.61	6.87
Protein, %	3.83±0.30 <sup>a</sup>	3.03	4.24
Lactose, %	4.56±0.22 <sup>a</sup>	4.11	4.87
Total solids, %	13.80±0.97 <sup>a</sup>	12.33	16.30
Milk production, kg day <sup>-1</sup>	9.92±3.43 <sup>a</sup>	4.00	17.20
Pregnant lactating cows			
Fat (%)	4.13±0.98 <sup>b</sup>	1.29	7.20
Protein (%)	3.33±0.33 <sup>b</sup>	2.62	4.30
Lactose (%)	4.63±0.23 <sup>b</sup>	4.04	5.11
Total solids (%)	13.04±1.14 <sup>b</sup>	10.50	16.09
Milk production, kg day <sup>-1</sup>	11.42±5.26 <sup>b</sup>	0.50	30.30

Similar letters within the same column indicate that they did not differ statistically at the 5% level of significance on the Tukey's test.

The classes established according to somatic cell count differed only in lactose content of milk, where higher percentages were observed in Class I cows milk, meaning those with lower SCC (Table 4). Changes in milk composition can be caused by several factors, among which microbial infection of the secretory parenchyma of the mammary glands can be highlighted, having an increase in SCC as a primary consequence. These inflammatory processes may reduce milk production, causing concentration of the solids and consequently configuring a positive correlation between SCC and the total solids milk content.

**Table 4.** Means and standard deviations (SD) of Gir and Guzerat milk composition (%) and production (kg) in relation to the two categories of somatic cell count.

Variable	Mean ± SD	Minimum	Maximum
SCC 1			
Fat, %	4.07 ± 0.83 <sup>a</sup>	1.79	5.59
Protein, %	3.31 ± 0.38 <sup>a</sup>	2.62	4.00
Lactose, %	4.71 ± 0.21 <sup>a</sup>	4.05	5.13
Total solids, %	13.15 ± 1.07 <sup>a</sup>	10.97	15.60
Milk production, kg day <sup>-1</sup>	12.06 ± 1.78 <sup>a</sup>	1.00	22.00
SCC 2			
Fat, %	4.23 ± 1.05 <sup>a</sup>	1.29	5.20
Protein, %	3.35 ± 0.32 <sup>a</sup>	2.67	4.30
Lactose, %	4.60 ± 0.23 <sup>b</sup>	4.04	5.04
Total solids, %	13.13 ± 1.07 <sup>a</sup>	10.50	15.30
Milk production, kg day <sup>-1</sup>	10.73 ± 4.96 <sup>a</sup>	1.70	30.00

Similar letters within the same column indicate that they did not differ statistically at the 5% level of significance on the Tukey's test. SCC 1- somatic cell counts up to 1x10<sup>5</sup> cells mL<sup>-1</sup>; SCC 2 - somatic cell count over 1x10<sup>5</sup> cells mL<sup>-1</sup>.

According to Rangel, Medeiros, Silva, Barreto, and Dorgival Júnior (2009), in the specific case of lactose the increase in SCC implies a reduction of this constituent, probably due to the use of lactose by bacteria as a substrate for growth and multiplication. Still, it is possible that this reduction results from a lower lactose synthesis capacity by the infected mammary epithelium and/or lactose loss to the bloodstream due to increased membrane permeability (Machado, Pereira, & Sarríes, 2000).

There were no records of 4.5 and 5.0 scores during the observation period for either breed. Not one Guzerat cow scored 2.0. Body condition score variation had no effect on milk's composition or somatic cell count (Table 5), highlighting the genetic potential of such animals in maintaining the content of the constituents.

**Table 5.** Body condition score (BCS) means and milk composition of Gir and Guzerat breeds in relation to score class.

Parameter	Body condition score (BCS)				
	2.0	2.5	3.0	3.5	4.0
<b>Gir</b>					
Fat, %	3.95±1.08 <sup>a</sup>	3.79±1.04 <sup>a</sup>	4.60±1.26 <sup>c</sup>	4.01±1.10 <sup>b</sup>	4.03±1.11 <sup>b</sup>
Protein, %	3.35±0.34 <sup>a</sup>	3.19±0.32 <sup>a</sup>	3.34±0.33 <sup>a</sup>	3.31±0.33 <sup>a</sup>	2.99±0.30 <sup>a</sup>
Lactose, %	4.59±0.46 <sup>a</sup>	4.60±0.46 <sup>a</sup>	4.46±0.45 <sup>a</sup>	4.69±0.47 <sup>a</sup>	4.80±0.48 <sup>a</sup>
Total solids, %	12.77±1.33 <sup>a</sup>	12.47±1.30 <sup>a</sup>	13.33±1.39 <sup>a</sup>	12.95±1.35 <sup>a</sup>	12.80±1.34 <sup>a</sup>
DDE, %	8.89±0.51 <sup>a</sup>	8.70±0.50 <sup>a</sup>	8.63±0.49 <sup>a</sup>	8.92±0.51 <sup>a</sup>	8.77±0.50 <sup>a</sup>
SSC, log <sub>10</sub> cfu mL <sup>-1</sup>	6.20±2.30 <sup>a</sup>	5.42±2.02 <sup>a</sup>	5.29±1.97 <sup>a</sup>	3.32±1.24 <sup>a</sup>	3.79±1.41 <sup>a</sup>
<b>Guzerat</b>					
Fat, %	-	3.70±0.96 <sup>c</sup>	4.32±1.12 <sup>c</sup>	4.20±1.08 <sup>c</sup>	4.23±1.09 <sup>c</sup>
Protein, %	-	3.32±0.36 <sup>c</sup>	3.41±0.37 <sup>c</sup>	3.44±0.37 <sup>c</sup>	3.52±0.38 <sup>c</sup>
Lactose, %	-	4.70±0.37 <sup>c</sup>	4.57±0.36 <sup>c</sup>	4.60±0.36 <sup>c</sup>	4.38±0.35 <sup>c</sup>
Total solids, %	-	12.64±1.25 <sup>c</sup>	13.29±1.32 <sup>c</sup>	13.20±1.31 <sup>c</sup>	13.10±1.30 <sup>c</sup>
DDE, %	-	8.94±0.56 <sup>c</sup>	8.98±0.56 <sup>c</sup>	9.04±0.56 <sup>c</sup>	8.84±0.55 <sup>c</sup>
SSC, log <sub>10</sub> cfu mL <sup>-1</sup>	-	4.59±2.11 <sup>a</sup>	5.19±2.39 <sup>a</sup>	4.64±2.14 <sup>a</sup>	6.09±2.80 <sup>a</sup>

Similar letters within the same column indicate that they did not differ statistically at the 5% level of significance on the Tukey's test. DDE = degreased dry extract; SSC = score of somatic cells.

Regarding the influence of body condition score on milk production and composition, although some studies have reported positive correlations between BCS and production of milk constituents Rennó et al. (2006) and Mushtaq et al. (2012), results found in the literature are still inconsistent (Mohamed et al., 2015). According to Lago, Susin, Lago, Faria, and Pires (2001), the influence of BCS on milk production and composition depends, among other factors, of production level of the analyzed cattle, with significant effects being observed more frequently in high producing animals (Waltner, McNamara, & Hillers, 1993). This assumption could possibly explain the lack of variation observed for the composition of milk produced by cows with different body condition scores (Table 5). Indeed, the mobilization of body reserves during lactation - which is directly related to BCS - aims to meet energy demands not met by diet (Waltner et al., 1993), which tend to be higher

when produce levels are high, however, this is not the case with the animals of this study. Still, it is important to point out that the absence of milk composition variation highlights the adaptability and ruggedness of zebu animals, which were able to produce milk rich in fat and protein, regardless of body condition.

## Conclusion

The production and chemical composition of Gir and Guzerat cow's milk are influenced by lactation stage - that leads to reduced daily production and increased protein and fat content with advancing lactation - and physiological condition - is achieving greater levels of milk constituents when dairy cows are not pregnant.

Somatic cell count only influences milk lactose content, which is used by the bacteria causing the infection as a substrate for growth and multiplication.

Body condition score does not alter the chemical composition of zebu cow's milk, probably because these animals have lower production levels compared to animals specialized in milk production.

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