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Milk fatty acid profile of dairy cows fed diets based on sugarcane bagasse in the Brazilian semiarid region

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ABSTRACT

Little is known about milk fatty acid (FA) profile of cows fed diets based on sugarcane (*Saccharum officinarum* L.) bagasse. This study aimed to evaluate FA profile and nutritional quality indexes of milk fat of cows fed sugarcane bagasse and concentrate in the Brazilian northeast semiarid region. Multiparous Girolando cows were allocated in four diets based on different levels of sugarcane bagasse (30%, 38%, 46%, and 54% total DM) added to concentrate. Fatty acids C18:1 *trans*-11 (lowest value for 46% inclusion) and conjugated linoleic acid (CLA) *cis*-9, *trans*-11 (lowest value for 46% inclusion) in milk fat presented a quadratic pattern ($P < 0.05$). This trend was also observed for C18:1 *cis*-9 ($P < 0.05$), but at a lower magnitude. There was a reduction in concentration of C18:1 *trans*-10 (0.320 to 0.148 g 100 g^{-1} total FA) and CLA *trans*-10, *cis*-12 (0.008 to 0.004 g 100 g^{-1} total FA) according to the levels of sugarcane bagasse increase. The greater inclusion of bagasse and consequent reduction in the proportion of concentrate, notably corn, cause an increase in concentration of medium-chain saturated FA and a linear reduction of C18:3 n-3 and C18:2 n-6, with a consequent increase in the indexes of atherogenicity (2.82 to 3.2) and thrombogenicity (3.39 to 3.84) and a decrease in ratio of hypo/hyper-cholesterolemic (0.44 to 0.37) and ω-6:ω-3 (12.44 to 8.80) in milk fat. Results indicate that different levels of sugarcane bagasse change the FA content in cows' milk when bagasse is an exclusive roughage source in the diet. The greater inclusion of sugarcane bagasse reduces the quality of milk fat of dairy cows. The lowest level of sugarcane bagasse inclusion tested (30%) promotes greater milk yield and healthier milk fat.

Key words: Conjugated linoleic acid, human health, milk fat, ruminant milk, *Saccharum officinarum*, tropical roughages.

INTRODUCTION

The irregularity of rainfall distribution in semi-arid regions directly influences forage and animal production. Due to the difficulties in roughage production in the semi-arid region, arising from physical-chemical constraints of soil, water resources or climatic conditions, alternatives forages are limited. However, due to the high amount of waste generated by the sugar-alcohol industry (about 30% of all milled material), sugarcane bagasse has become an ingredient frequently used in the diets of dairy cattle (Hofsetz and Silva, 2012), and the availability of this product coincides with the period of shortage of roughage.

Despite this possibility, sugarcane (*Saccharum officinarum* L.) bagasse has a low nutritive value, presenting neutral detergent fiber (NDF) above 80% and crude protein content around 1.5%, which makes it necessary to include a

high proportion of concentrate in ruminant diets to meet nutritional requirements. Besides raising the cost of the diet, concentrated feed, alters the ruminal fermentation pattern, with an increase in the propionate:acetate ratio (Han et al., 2014). Silva et al. (2015) studying diets with sugarcane bagasse and concentrate observed that great proportions of concentrate increased propionate, the ratio propionate:acetate and decreased linearly the pH. These modifications in fermentation patterns also modify the ruminal biohydrogenation routes, with possible modification of the fatty acid (FA) profile of the milk (Jenkins et al., 2008).

The manipulation of FA profile of ruminant milk through changes in diets has been the subject of several studies, since milk fat contains numerous bioactive compounds with potential beneficial effects on human health. In addition, certain diets may induce the ruminal formation of specific FAs (e.g., conjugated linoleic acid [CLA *trans*-10, *cis*-12), which inhibits mammary lipogenesis, with a consequent reduction in milk fat content (Garduño et al., 2018).

It was hypothesized that altering the levels of sugarcane bagasse and concentrate in dairy cows' diet promotes modifications in milk FA profile. Therefore, the objective of this study was to evaluate the fatty acid profile and nutritional quality indexes of milk fat of crossbred cows, reared in the semi-arid region and fed diets containing different levels of sugarcane bagasse.

MATERIALS AND METHODS

This study was carried out in Roçadinho Farm, located in Capoeiras city (18º36'33" S, 36º37'30" W; 733 m a.s.l.), in the middle of Agreste and Ipojuca Valley micro-region, Pernambuco State, Brazil, that presents semi-arid climate classification as Bsh (Köppen, 1948). The management and animals' care were performed by the guidelines and recommendation of the Committee on Ethics on Animal Studies at the Federal Rural University of Pernambuco (License Nº033/2014), Recife, Brazil.

Eight lactating, multiparous Girolando cows (3/4 to 7/8 Holstein × Gyr) at 90 ± 10 d (mean ± SD) of lactation, and an average initial body weight (BW) of 600 ± 34.3 kg (mean ± SD), were randomly assigned to replicated 4×4 Latin square designs, according to their genetic group. The trial lasted for 84 d, with four consecutive 21-d periods divided into 14-d adaptation and 7-d sampling periods. The animals were housed in individual stalls, half-covered 16 m^2 each, equipped with sand bed and rice straw, automatic waterers and individual feeders.

The chemical composition of ingredients is shown in Table 1. The experimental diets consisted of different sugarcane bagasse levels (30%, 38%, 46% and 54% DM). The diets were formulated to be isonitrogenous, according to the requirements calculated by the NRC (2001) for 600 kg cows producing 20 kg d^{-1} and 3.5% milk fat. The sugarcane bagasse was acquired from the local industry.

The animals were fed twice daily, after the morning (04:00 h) and afternoon (16:00 h) milking sessions. The amount of feed supplied was corrected daily, allowing *ad libitum* intake with 10% refusal in the fresh matter; water was also available *ad libitum*. Samples of the feeds and refusals were collected during the last 7 d of each experimental period and stored at -20 °C in airtight plastic bags.

Samples of feeds, refusals, and feces were analyzed for DM (method 934.01) according to AOAC (2005) and ether extract (EE) according to AOCS (2004). Also, samples of diet ingredients and orts were taken and stored at -18 °C, for further fatty acid (FA) determination by the gas-phase chromatography technique (Tables 1 and 2), as described by Ribeiro et al. (2014). The analysis was performed at the laboratory of chromatography of Embrapa Gado de Leite (Juiz de Fora, Minas Gerais, Brazil) using a 6890 N gas-phase chromatograph (Agilent Technologies Inc., Santa Clara, California, USA), equipped with HP-FFAP column (25 m × 0.20 m × 0.33 μm; Agilent Technologies, Santa Clara, California, USA) and flame ionization detector.

The cows were milked twice a day (04:00 and 16:00 h), and milk yield (MY) was registered from the 15th to the 21st day of each experimental period. The milk samples were collected on days 19th and 20th during both milking periods, after the last collection, and the composed samples were made for each cow. The procedures for analysis of MY and content, as β-hydroxybutyrate (BHBA) concentration in plasma were made as described by Freitas et al. (2018).

Milk samples were collected, transferred to bottles without preservatives, and stored at -18 °C, for the future determination of FA profile at the laboratory of chromatography of Embrapa Gado de Leite. This FA profile determination was performed according to the description by Ribeiro et al. (2014), in a 7820 A gas-phase chromatograph (Agilent

Table 1. Ingredients proportions, chemical composition and major fatty acid composition of experimental diets.

	Sugarcane bagasse inclusion (% DM)			
Item, g kg^{-1} DM	30	38	46	54
Sugarcane bagasse	300.0	380.0	460.0	540.0
Corn meal	493.0	410.5	328.0	245.5
Soybean meal	173.0	173.0	173.0	173.0
Urea + ammonium sulfate[1]	4.0	6.5	9.0	11.5
Salt	5.0	5.0	5.0	5.0
Sodium bicarbonate	10.0	10.0	10.0	10.0
Minerals[2]	15.0	15.0	15.0	15.0
Chemical composition, g kg^{-1} DM				
Dry matter	719.0	685.1	654.3	626.1
Organic matter	932.2	927.1	921.9	916.8
Crude protein	145.3	146.3	147.5	148.9
Ether extract	32.5	28.9	25.2	21.5
Neutral detergent fiber	281.4	344.9	404.8	462.5
Non fiber carbohydrates	439.6	381.5	323.3	265.5
Total digestive nutrients	698.3	663.4	662.1	651.7
Fatty acids profile[3], g 100 g^{-1} total FA				
C10:0	0.53	0.66	0.80	0.95
C12:0	0.46	0.57	0.68	0.80
C14:0	0.54	0.65	0.77	0.88
C16:0	21.77	22.69	23.59	24.50
C16:1	0.59	0.75	0.91	1.07
C18:0	0.53	0.67	0.81	0.96
C17:0	2.83	3.17	3.51	3.85
C18:1 *cis*-9	22.25	21.98	21.68	21.41
C18:1 *cis*-11	0.67	0.61	0.54	0.47
C18:2 n-6	39.59	36.71	33.78	30.89
C18:3 n-3	1.70	1.56	1.41	1.27
C20:0	0.14	0.12	0.09	0.07
C22:0	0.11	0.09	0.08	0.06
C24:0	0.05	0.05	0.04	0.03

DM: Dry matter; FA: fatty acids.
[1]Proportion of 9:1 as fed.
[2]Mineral compounds: Dicalcium phosphate; limestone; salt; sulfur; zinc sulfate; copper sulfate; manganese sulfate; potassium iodate; sodium selenite.
[3]Estimated values based on FA profile of ingredients.

Table 2. Fatty acid (FA) composition of individual feeds.

Fatty acids, g 100 g^{-1} total FA	Sugarcane bagasse	Corn meal	Soybean meal
C10:0	1.75	-	-
C12:0	1.42	0.02	0.13
C14:0	1.50	0.09	0.27
C16:0	30.10	18.18	21.83
C16:1	1.98	-	-
C18:0	5.72	1.42	2.40
C17:0	1.17	-	-
C18:1 *cis*-9	23.69	26.44	12.19
C18:1 *cis*-11	-	0.847	1.493
C18:2 n-6	15.88	50.66	56.97
C18:3 n-3	0.10	1.80	4.48
C20:0	-	0.25	0.80
C22:0	-	0.17	0.11
C24:0	-	0.09	0.05

Technologies), equipped with a CP-Sil 88 for FAME 100 m × 0.25 mm × 0.2 μm (Agilent Technologies) column and a flame ionization detector.

To evaluate the nutritional quality of milk fat, equations were used to calculate the atherogenicity index (AI), thrombogenicity index (TI), omega6:omega3 FA ratio (ω-6:ω-3), and hypo/hyper-cholesterolemic FA ratio (h/H) according to Ribeiro et al. (2014).

The stearoyl-CoA desaturase enzyme (SCD1) activity was calculated for four FA pairs: SCD_{14} = C14:1*cis*-9/14:0 + C14:1*cis*-9; SCD_{16} = C16:1*cis*-9/16:0 + C16:1*cis*-9; SCD_{18} = C18:1*cis*-9/18:0 + C18:1*cis*-9; and SCD_{RA} = conjugated linoleic acid (CLA) *cis*-9 *trans*-11/C18:1*trans*-11 + CLA *cis*-9 *trans*-11 (Kelsey et al., 2003).

The variables studied were analyzed according to the following statistic model:

$$Yijkl = \mu + T_i + S_j + P_k + (A/S)_{ij} + (T\,S)_{ij} + e_{ijkl}$$

where Y_{ijkl} is the observation in cow l, in period k, submitted to treatment j, in Latin square i; μ is overall mean; T_i is fixed effect of treatment; S_j is random effect of Latin square; P_k is random effect of period; $(A/S)_{ij}$ is random effect of cow l in Latin square i; $(T\,S)_{ij}$ is random effect of interaction between treatment (i) and Latin square (j); and e_{ijkl} is random residual error.

The data were statistically analyzed using PROC MIXED procedures from the Statistical Analysis System (SAS version 9.4; SAS Institute, Cary, North Carolina, USA) for variance and regression analysis, respectively, adopting 0.05 as the critical level of probability for type I errors.

RESULTS

The increasing levels of sugarcane bagasse in the diets linearly reduced milk yield ($P < 0.05$; Table 3), which is consistent with the reduction in DM intake (18.30 to 13.90 kg d^{-1}) and, consequently, of nutrients, notably the EE (0.62 to 0.32 kg d^{-1}) and TDN (12.8 to 9.06 kg d^{-1}). Despite the higher proportions of concentrate (70% and 62%) in the diets with 30% and 38% of sugarcane bagasse, there was no effect on the milk fat content ($P > 0.05$; Table 3), as well as the BHBA ($P > 0.05$; Table 3), indicating that there was no body fat mobilization.

There was a linear increase ($P < 0.05$) in the total concentration of saturated C12:0, C14:0, and C16:0 FAs with the inclusion of sugarcane bagasse in the diets (Table 4). Still, relating to Table 4, the total concentration of the odd- and branched-chain FA (OBCFA) increased linearly ($P < 0.05$) with the inclusion of sugarcane bagasse. Also, a linear increase in the concentration of iso-C14:0, iso-C15:0, and iso-C18:0 in milk fat could be observed when the proportion of sugarcane bagasse was increased in the diets (Table 4). The C18:0 presented a quadratic pattern ($P < 0.05$; Table 4) with sugarcane bagasse inclusion levels, as well as the C18:1 *trans*-11 and the CLA *cis*-9, *trans*-11.

Concentrations of C18:1 *cis*-9, C18:2 n-6, and C18:3 n-3 FA in milk fat were linearly reduced ($P < 0.05$) with the inclusion of sugarcane bagasse (Table 4). Also, regarding CLA *trans*-9, *cis*-11 and CLA *trans*-10, *cis*-12, there was a quadratic pattern ($P < 0.05$) for both.

Table 3. Nutrients intake, performance, and BHBA plasma concentration for cows fed diets with different sugarcane bagasse level inclusion.

	Sugarcane bagasse inclusion (%)					P Value	
Item	30	38	46	54	SEM	L	Q
Intake, kg d^{-1}							
Dry matter	18.30	17.77	16.13	13.90	0.505	< 0.01	0.069
Ether extract	0.62	0.54	0.43	0.32	0.014	< 0.01	0.063
Total digestible nutrients	12.8	11.8	10.7	9.6	0.571	< 0.01	0.078
Performance							
Milk yield, kg d^{-1}	22.36	20.58	18.46	16.38	0.741	< 0.01	0.661
FCMY (3.5%), kg d^{-1}	22.70	21.41	19.35	16.48	1.025	< 0.01	0.141
Fatty, g 100 g^{-1}	3.61	3.77	3.78	3.54	0.188	0.707	0.086
BHBA plasma, mmol L^{-1}	0.51	0.55	0.58	0.52	0.037	0.478	0.037

SEM: Standard error of the mean; L: linear effect; Q: quadratic effect; FCMY: 3.5% fat-corrected milk yield; BHBA: β-hydroxybutyrate.

Table 4. Fatty acids (FA) profile of milk from cows fed diets with different sugarcane bagasse level inclusion.

Fatty acids, g 100 g^{-1} total FA	Sugarcane bagasse inclusion (%)					P Value	
	30	38	46	54	SEM	L	Q
4:0	3.300	3.346	3.517	3.378	0.040	0.274	0.264
5:0	0.026	0.019	0.016	0.014	0.001	< 0.01	0.058
6:0	2.179	2.321	2.344	2.228	0.027	0.307	< 0.01
7:0	0.032	0.022	0.017	0.014	0.001	< 0.01	0.065
8:0	1.401	1.433	1.430	1.292	0.027	< 0.01	< 0.01
9:0	0.047	0.031	0.024	0.018	0.002	< 0.01	0.069
10:0	3.436	3.689	3.346	3.043	0.050	<0.01	< 0.01
10:1 *cis*-9	0.282	0.302	0.311	0.302	0.006	0.138	0.185
11:0	0.115	0.089	0.078	0.067	0.004	< 0.01	0.140
12:0	4.269	4.412	4.170	3.697	0.057	< 0.01	< 0.01
12:1 *cis*-9 + 13:0	0.276	0.224	0.209	0.192	0.008	< 0.01	0.063
14:0	12.301	12.468	12.482	12.233	0.121	0.062	0.081
14:0 iso	0.122	0.196	0.214	0.224	0.008	< 0.01	0.192
14:1 *cis*-9	1.011	0.962	1.058	1.120	0.045	0.014	0.134
15:0	1.221	1.182	1.136	1.221	0.025	0.721	0.045
15:0 iso	0.298	0.356	0.458	0.489	0.015	< 0.01	0.371
15:0 *anteiso*	0.535	0.668	0.692	0.779	0.019	< 0.01	0.097
16:0	27.610	28.962	30.835	31.814	0.676	< 0.01	0.723
16:0 iso	0.245	0.340	0.372	0.363	0.013	< 0.01	0.037
16:1 *trans*-12	0.167	0.143	0.140	0.163	0.004	0.674	< 0.01
16:1 *trans*-9 + 17:0 iso	0.440	0.418	0.496	0.528	0.014	< 0.01	0.226
16:1 cis-9 + 17:0 *anteiso*	1.529	1.478	1.664	1.807	0.060	< 0.01	0.038
∑ 12:0+14:0+16:0	43.783	45.171	47.220	47.562	0.762	< 0.01	0.421
17:0	0.630	0.645	0.654	0.710	0.016	< 0.01	< 0.01
17:1 *cis*-9	0.154	0.141	0.150	0.190	0.007	< 0.01	< 0.01
∑ OCLFA[1]	2.618	2.487	2.410	2.561	0.055	0.214	< 0.01
∑ OCFA[2]	5.421	5.408	5.722	6.166	0.126	< 0.01	< 0.01
∑ BCFA[3]	3.211	3.499	3.948	4.244	0.090	< 0.01	0.941
∑ OB∑ *trans*[4]	5.829	5.986	6.358	6.805	0.126	0.041	0.340
18:0	10.259	10.659	9.659	8.841	0.360	< 0.01	0.027
18:0 iso	0.040	0.042	0.049	0.052	0.002	< 0.01	0.840
18:1 *trans*-4	0.029	0.027	0.022	0.021	0.001	< 0.01	0.987
18:1 *trans*-5	0.022	0.022	0.019	0.015	0.001	0.024	0.271
18:1 *trans*-6+7+8	0.226	0.188	0.173	0.155	0.007	< 0.01	0.127
18:1 *trans*-9	0.190	0.167	0.144	0.137	0.006	< 0.01	0.143
18:1 *trans*-10	0.320	0.235	0.215	0.148	0.015	< 0.01	0.705
18:1 *trans*-11	0.795	0.782	0.780	0.961	0.025	< 0.01	< 0.01
18:1 *trans*-12	0.245	0.196	0.161	0.126	0.010	< 0.01	0.528
18:1 *trans*-13 and *trans*-14	0.256	0.210	0.210	0.186	0.008	< 0.01	0.505
18:1 *trans*-16	0.234	0.196	0.152	0.117	0.009	< 0.01	0.853
18:1 *cis*-9	19.570	17.851	16.850	16.810	0.467	< 0.01	0.055
18:1 *cis*-11	1.076	0.904	0.772	0.849	0.026	< 0.01	< 0.01
18:1 *cis*-12	0.249	0.216	0.187	0.159	0.008	< 0.01	0.788
18:1 *cis*-13	0.079	0.061	0.059	0.064	0.002	< 0.01	< 0.01
18:1 *cis*-15 + 19:0	0.095	0.107	0.102	0.107	0.003	0.235	0.613
18:2 *trans*-9 *trans*-12	0.012	0.012	0.012	0.013	0.000	0.483	0.903
18:2 *cis*-9 *trans*-12	0.037	0.025	0.019	0.016	0.002	< 0.01	0.116
18:2 *trans*-9 *cis*-12	0.025	0.021	0.017	0.020	0.001	0.042	0.088
18:2 n-6	2.106	1.748	1.417	1.265	0.058	< 0.01	0.210
CLA *cis*-9 *trans*-11	0.485	0.434	0.447	0.551	0.018	0.036	< 0.01
CLA *trans*-9 *cis*-11	0.025	0.020	0.021	0.023	0.001	0.481	0.020
CLA *trans*-10 *cis*-12	0.008	0.005	0.003	0.004	0.000	< 0.01	0.018
18:3 n-6	0.025	0.025	0.025	0.026	0.001	0.347	0.785
18:3 n-3	0.147	0.130	0.120	0.120	0.004	< 0.01	0.230
20:0	0.146	0.154	0.147	0.145	0.004	0.546	0.221
20:2 n-6	0.033	0.030	0.030	0.032	0.001	0.547	< 0.01

Continued Table 4.

Fatty acids, g 100 g^{-1} total FA	Sugarcane bagasse inclusion (%)					P Value	
	30	38	46	54	SEM	L	Q
20:3 n-6	0.117	0.111	0.106	0.100	0.004	0.014	0.932
20:4 n-6	0.166	0.153	0.147	0.171	0.005	0.695	< 0.01
20:5 n-3	0.012	0.012	0.012	0.013	0.001	0.068	0.182
21:0	0.015	0.018	0.016	0.017	0.000	0.302	0.197
22:0	0.035	0.036	0.040	0.044	0.001	< 0.01	0.311
22:5 n-3	0.051	0.042	0.038	0.045	0.001	0.068	0.182
23:0	0.008	0.007	0.007	0.008	0.000	0.589	0.234
24:0	0.019	0.022	0.025	0.027	0.001	< 0.01	0.855
∑ *trans*, except VA-RU[5]	2.237	1.885	1.808	1.672	0.050	< 0.01	0.156

SEM: Standard error of the mean; L: linear effect; Q: quadratic effect; CLA: conjugated linoleic acid.
[1]Odd-chain linear FA = C5:0 + C7:0 + C9:0 + C11:0 + (C12:1 *cis*-9 + C13:0) + C15:0 + C17:0 + C17:1 *cis*-9 + (C18:1 *cis*-15 + C19:0) + C21:0 + C23:0.
[2]Odd-chain FA = C5:0 + C7:0 + C9:0 + C11:0 + (C12:1 *cis*-9 + C13:0) + C15:0 iso + C15:0 anteiso + C15:0 + (C16:1 *trans*-9 + C17:0iso) + (C16:1 *cis*-9 + C17:0anteiso) + C17:0 + C17:1 *cis*-9 + (C18:1 *cis*-15 + C19:0) + C21:0 + C23:0.
[3]Branched chain FA = C14:0 iso + C15:0 iso + C15:0 anteiso + C16:0 iso + (C16:1 *trans*-9 + C17:0iso) + (C16:1 *cis*-9 + C17:0anteiso) + C18:0 iso.
[4]OB∑ trans = Odd and branched ∑ *trans*, except VA-RU (vaccenic and rumenic) = AGCI + C14:0 iso + C16:0 iso + C18:0 iso.
[5]VA-RU = (C16:1 *trans*-9 + C17:0 iso) + C16:1 *trans*-12 + C18:1 *trans*-4 + C18:1 *trans*-5 + C18:1 *trans*-6 *trans*-8 + C18:1 *trans*-9 + C18:1 *trans*-10 + C18:1 *trans*-12 + C18:1 *trans*-13 *trans*-14 + C18:1 *trans*-16 + C18:2 *trans*-9 *trans*-12 + C18:2 *cis*-9 *trans*-12 + C18:2 *trans*-9 *cis*-12 + CLA *trans*-9 *cis*-11 + CLA *trans*-10 *cis*-12.

The activity of Δ^9-desaturase increased linearly ($P < 0.05$) with the inclusion of sugarcane bagasse for the SCD-14 pair and presented quadratic behavior ($P < 0.05$) for the product:substrate ratio for the SCD-16 and SCD-18 pairs (Table 5). The ω-6:ω-3 ratio was linearly reduced ($P < 0.05$) with the inclusion of sugarcane bagasse in the diets (Table 6). The h/H ratio also decreased linearly ($P < 0.05$) with the inclusion of sugarcane bagasse in the diets (Table 6), due to the lower concentration of C18:1 *cis*-9.

Table 5. Stearoyl-CoA desaturase enzyme (SCD1) activity.

Index	Sugarcane bagasse inclusion (%)				SEM	P Value	
	30	38	46	54		L	Q
SCD_{14}	0.075	0.071	0.078	0.084	0.003	0.019	0.096
SCD_{16}	0.053	0.050	0.051	0.054	0.001	0.473	0.033
SCD_{18}	0.655	0.625	0.634	0.657	0.007	0.607	< 0.01
SCD_{RA}	0.375	0.355	0.364	0.363	0.006	0.440	0.244

SEM: Standard error of the mean; L: linear effect; Q: quadratic effect.

Table 6. Index of nutritional quality of milk fat.

Index	Sugarcane bagasse inclusion (%)				SEM	P Value	
	30	38	46	54		L	Q
Atherogenicity (IA)	2.82	3.10	3.35	3.20	0.094	< 0.01	0.011
Thrombogenicity (IT)	3.39	3.76	4.03	3.84	0.103	< 0.01	< 0.01
ω-6:ω-3	12.44	11.38	10.33	8.80	0.265	< 0.01	0.127
Hypo/hyper-cholesterolemic (h/H)	0.44	0.41	0.37	0.37	0.014	< 0.01	0.108

SEM: Standard error of the mean; L: linear effect; Q: quadratic effect.

DISCUSSION

An excess of the fiber of low degradability from sugarcane bagasse (Table 2) promoted a reduction on DM, EE and TDN intakes. According to Ahmed et al. (2013), the use of sugarcane bagasse for animal feeding is limited due to their low digestibility related to their high content of fiber which contains more than 60% of its DM in the form of cellulose, hemicellulose, and lignin. The reduction in the intake of DM and energy, could explain the decrease in milk yield and 3.5% fat-corrected milk in lactating cows fed increasing levels of sugarcane bagasse.

The milk fat content was not altered once that the contribution of effective fiber from sugarcane bagasse, did not provide the ruminal conditions necessary to induce a drop in the fat content. This pattern is consistent with low concentrations of C18:1 *trans*-10 and CLA *trans*-10, *cis*-12 FA (Table 4).

The C12:0, C14:0, and C16:0 FAs originate mostly from *de novo* synthesis in the mammary gland from acetate and BHBA, rather than direct dietary intake. Although contents of these FAs were higher in the diets with higher ratios of sugarcane bagasse, DM intake was significantly reduced, suggesting a reduction in the intake of these FAs.

The C18:0 is a highly researched FA due to its high concentration in milk and its importance for human health. Its response was due, probably, to the reduction in the proportion of concentrate, notably corn, resulting, consequently, in reducing by half the EE intake (Table 3). Corn was the component of the concentrate that contributed most to the substrate input (C18:2 n-6 and C18:3 n-3) for ruminal biohydrogenation. In addition, much of the polyunsaturated fatty acids (PUFAs) present in the diet may have escaped biohydrogenation or have undergone the process only partially, reducing the content of the C18:0 FA in milk to a specific level of sugarcane bagasse. It is known that biohydrogenation may not be entirely performed for all PUFAs and, for this reason, some FAs, such as C18:2 n-6 and C18:3 n-3, or other intermediate products, such as CLA and C18:1 *trans*-11, can reach the duodenum and be absorbed (Holanda et al., 2011).

According to Shingfield et al. (2013), 70% to 95% of CLA *cis*-9, *trans*-11 secreted in the milk originates from C18:1 *trans*-11, by the action of the enzyme Δ^9-desaturase on the mammary gland, which would explain the quadratic behavior for both. In this study, there was no effect on the activity of this enzyme for the rumenic/vaccenic pair (SCD) (Table 5) among the different levels of bagasse inclusion and for the control diet. This parameter is used to infer substrate selectivity and activity by the Δ^9-desaturase enzyme in the mammary gland, based on the relationship between the levels of *cis*-9-monounsaturated FA and its saturated isomers (Bauman and Lock, 2006). The C18:1 *cis*-9 is recognized for its human health benefits, since it has the ability to reduce the concentration of plasma cholesterol low density lipoprotein (LDL) (FAO, 2010).

The responses observed in CLA *trans*-9, *cis*-11 and CLA *trans*-10, *cis*-12 can be related to their precursor in the partial biohydrogenation – the C18:2 n-6 (Buccioni et al., 2012) – and usually, their concentrations are inversely associated with the milk fat content. However, the concentrations of these FAs in milk fat were apparently very low (Table 4) for inhibition of lipogenesis in the mammary gland, which is corroborated by the high milk fat content (Table 3).

The activity of Δ^9-desaturase and the ratio for the SCD-16 and SCD-18 pairs responded according to the variation in the product quantities formed per unit of time among diets composed of cane bagasse and concentrate.

The atherogenicity index (AI) and thrombogenicity index (TI) values obtained (Table 6) were lower than or similar to those found in the literature (Barros et al., 2013; Aguiar et al., 2015), indicating that the diets tested did not interfere negatively in these qualitative indexes. The indexes obtained are relevant because they indicate the potential for stimulating platelet aggregation, that is, lower indices are related to higher proportions of antiatherogenic FAs and/or a lower proportion of atherogenic FAs in food fat and, therefore, a greater potential for preventing cardiovascular disease (Tonial et al., 2010).

The ω-6:ω-3 ratio linear reduction (Table 6) may be explained by the linear decrease in the concentration of C18:2 n-6 and, in a lesser extent, of C18:3 n-3 (Table 4), which are the main ω-6 and ω-3 FAs found in milk fat. Values below 10.0 indicate desirable proportions between these groups of FA in the human diet to reduce the risk of cardiovascular diseases (FAO, 1994).

Finally, the h/H ratio decreased the response with the inclusion of sugarcane bagasse in the diets (Table 6); unlike AI and TI, higher the h/H ratio, better the nutritional quality of the fat contained in food (Bentes et al., 2009).

One of the bovine milk characteristics is the high saturated FA content with the chains from 4 to 16 C (Vanrobays et al., 2016) that results in the synthesis *de novo*. Some of those FA are LDL cholesterol precursors and are relating to

cardiovascular diseases. The current trend is for the high demand of health food, with low saturated fat content, especially, functional foods that promote physiological benefits to the human body, as the mono and polyunsaturated FA, high density lipoprotein (HDL) cholesterol precursor, that collaborate to reduce coronary disease. As healthcare costs and average life expectancy rise, the public has sought ways to become healthier and develop higher qualities of life (Martirosyan and Singh, 2015).

The adequate milk fat quality is necessary to achieve the consumer market that is demanding low saturated fat production due to their deleterious effects on human health. Thus, these results could be used to the knowledge of milk fatty quality of cows fed common feeds from arid and semiarid regions as sugarcane bagasse, and the knowledge of necessity to manipulating the cow's diet aiming to alter the milk fatty acids profile.

CONCLUSIONS

The different levels of sugarcane bagasse change the fatty acid content in cows' milk when the bagasse is an exclusive roughage source in the diet. It is recommended 30% of sugarcane bagasse inclusion in Girolando cows' diets to promote greater milk yield and healthier milk fat.

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