



# Milk production and composition, food consumption, and energy balance of postpartum crossbred Holstein-Gir dairy cows fed two diets of different energy levels

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## Abstract

The objective of this study was to evaluate the production, consumption, and energy balance parameters of primiparous 3/4 and 7/8 Holstein × Gir (HG) dairy cows fed two diets of differing energy levels during the postpartum period. At the beginning of the study, 28 days prepartum, the average weight of both genetic groups was  $498 \pm 12$  kg and body condition score (BCS) was  $3.5 \pm 0.05$ . At the end of the study, 61 days postpartum, the 3/4 HG cows had higher weight and body condition scores than the 7/8 HG ( $456 \pm 8$  and  $429 \pm 8$  kg and  $3.13 \pm 0.03$  and  $2.94 \pm 0.03$  BCS for 3/4 HG and 7/8 HG, respectively). Milk from cows fed the high-energy diet had higher percentages of fat, protein, lactose, and total dry extract than cows fed the low-energy diet. Cows fed the high-energy diet had higher net energy intake ( $95.3 \pm 1.9$  vs.  $88.1 \pm 2.1$  MJ/day) and higher energy balance ( $3.64 \pm 2.13$  vs.  $-6.02 \pm 2.30$  MJ/day). The 3/4 HG cows displayed higher energy for maintenance ( $33.1 \pm 0.4$  MJ/day) than the 7/8 HG ( $31.5 \pm 0.5$  MJ/day). In conclusion, although the primiparous 3/4 HG were heavier than the 7/8 HG and had a higher body condition score, no differences in milk produced up to 60 days postpartum were observed. The higher energy diet during the postpartum period increased energy balance, resulting in higher production of milk fat, protein, and lactose.

**Keywords** Dairy cattle · Girolando · Energy consumption · Milk composition · Primiparous

## Introduction

Given the higher average temperatures experienced by tropical countries throughout the year, tropical nations face distinct challenges in milk production when using European cattle breeds as these animals suffer from heat stress and metabolic changes in such conditions. As a result, it is important to study animals that are better adapted to tropical climates. The production of F1 crossbred females for dairy farms in this environment normally involves breeding zebu (*Bos indicus*) females with a Holstein bull to exploit the beneficial effects of heterosis (Santos et al. 2014).

At the beginning of lactation, there is an incompatibility between dry matter intake (DMI) and the energy required for milk production, and thus, negative energy balance (NEB) occurs. We have seen remarkable advances in animal productivity in the last 75 years, with annual milk yield per cow increasing over fourfold and no evidence about this increase is nearing a plateau. Thanks to these gains in productive efficiency, there have been dramatic reductions in resource inputs and the carbon footprint per unit of milk produced. The primary source for these historic gains relates to animal variation in nutrient partitioning (Baumgard et al. 2017). Animals with NEB are more likely to develop metabolic diseases, such as ketosis, mastitis, and placental retention, thereby impairing milk production and fertility (Daibert et al. 2018).

The production and composition of milk from crossbred Holstein × Gir cows are affected by various factors, such as the level of farm management and the ratio of crossbreed used. However, the body condition score (BCS) at calving, the energy balance in which the animal finds itself, and the diet offered are important factors in determining the composition of milk.

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Research shows that 3/4 HG and 7/8 HG cows produce higher quantities of milk when compared with animals of other genetic groups, but this production is decreased when the ratio of Gir genes is increased (Canaza-Cayo et al. 2017). These animals are responsible for most of the milk produced in the tropics. Thus, it is necessary to study the milk production and the energy balance that these animals experience in the early postpartum period according to the level of energy present in their diet in order to understand the differences between these cows and specialized breeds and to adapt the management of these animals in observance of their productive potential and requirements.

Therefore, this study aimed to evaluate the different production, consumption, and energy balance parameters of primiparous 3/4 HG and 7/8 HG dairy cows fed two different diets of differing energy levels during the postpartum period.

## Material and methods

Twenty-eight dairy crossbred primiparous females with a mean age of  $35.64 \pm 5.72$  months were used, 15 of which had a genetic composition of 3/4 HG and 13 of which were 7/8 HG animals. At 28 days prepartum, females from the genetic group 3/4 HG weighed  $498 \pm 11$  kg, and the 7/8 HG weighed  $498 \pm 12$  kg, and the BCS was  $3.5 \pm 0.05$  (scale from 1 to 5; Edmonson et al. 1989).

From 28 days of the expected calving, the animals were housed in a free-stall system with individual feed bins (Calan Gate, Calan Inc., New Hampshire, USA) for adaptation. The diet at this stage was provided ad libitum and consisted of corn silage plus 2 kg of concentrate per animal (21% crude protein, 8.45 MJ/kg of dry matter, 0.98% Ca and 0.5% P) based on corn, soybean bran, urea, common salt, and a mineral nucleus. The feeding was provided assuming a minimum leftover of 5%. Ten days before their expected calving date, the animals were transferred and kept in maternity pens with access to pasture (*Cynodon dactylon*) until parturition while continuing to be fed the same diet. A mineral mixture and water were provided ad libitum during the entire prepartum period.

After calving, every week, the primiparous was weighed and evaluated for BCS (Edmonson et al. 1989) using a scale of 0.25. They were again housed in the free stall where they returned to the previous feeding system with individual feed bins until 61 days postpartum. At this time, the females were distributed homogeneously among the diets, in a  $2 \times 2$  factorial arrangement (energy in the diet and genetic composition). The diets offered were either high energy (HE) or low energy (LE) (Table 1), with eight 3/4 HG animals and seven 7/8 HG animals fed the HE diet and seven 3/4 HG animals and six 7/8 HG animals fed with the LE diet. The diets were formulated using SuperCrac software (TD Software Ltda) and according to the nutritional requirements for growing Holstein cows

producing 16 l of milk/day as determined by the National Research Council –NRC Nutrient requirements of dairy cattle. 7 ed. Washington, D. C (2001).

The cows were milked twice daily at 06:00 and 15:00 using a mechanized milking system equipped with a closed circuit with central middle line and digital milk meter. Milk control was performed daily, and milk yield (MY) was recorded. MY was corrected to 3.5% fat (FCM) as per Sklan et al. (1992).

Samples were collected weekly to determine the composition of milk, with an equal number of samples taken from morning and afternoon milk production. The analytical principle applied was the differential absorption of infrared waves by the different components of the milk as per the International Dairy Federation (IDF 2000).

As per Silva and Queiroz (2002), samples of the total diet offered at each treatment phase were analyzed for dry matter, ash, neutral detergent fiber (NDF), corrected neutral detergent fiber (NDFc), acid detergent fiber (ADF), crude energy (CE), etheral extract (EE), and crude protein (CP).

The dry matter intake (DMI) was determined by calculating the difference between the offered diet and daily scraps in dry matter. The energy balance (EB) was estimated by calculating the difference between net energy intake (NEI), the sum of net energy of maintenance (NEm), and net energy of lactation (NElac), expressed in megajoules per day. The NEI, NEm, and NElac were calculated according to the procedures laid out by the NRC (2001).

Data was analyzed using the SAS MIXED procedure, version 9.1.3 (SAS Institute, Cary, NC, USA), according to the following model:

$$Y_{ijkl} = \mu + G_i + N_j + \delta_{ik} + W_l + (G \times N)_{ij} + (G \times W)_{ik} + (N \times W)_{jk} + \varepsilon_{ijkl},$$

where:

$\mu$	Overall mean
$G_i$	Fixed effect of the genetic group
$N_j$	Fixed effect of the food plan (HE or LE)
$\delta_{jk}$	Covariance between repeated measures within animals
$W_l$	Effect of first week
$(G \times N)_{ij}$	Effect of interaction between genetic group and food plan
$(G \times W)_{il}$	Effect of interaction between genetic group and first week
$(N \times W)_{jl}$	Effect of interaction between food plan and first week
$\varepsilon_{ijkl}$	Effect of random error associated with $Y_{ijkl}$ observation

The experimental week was included as a repeated statement, and the cow was nested within treatment as random effect. An autoregressive covariance structure was used to

**Table 1** Composition of total diet provided to primiparous crossbred dairy cows during the early postpartum period

Diet composition (dry matter basis)	Energy diet	
	High energy (HE)	Low energy (LE)
Corn silage (%)	60.1	80.3
Soybean meal (%)	11.6	11.8
Ground corn (%)	25.1	4.7
Nucleus <sup>a</sup> (%)	2.1	2.14
Urea (%)	1.1	1.09
Dry matter (%)	33.2	27.4
Crude protein (kg/dry matter)	0.2	0.2
Neutral detergent fiber (%)	44	55
Calcium (g/kg)	7.2	7.4
Phosphorus (g/kg)	3.8	3.6
Net energy (MJ/kg)	8.1	7.1

<sup>a</sup> Mineral and vitamin composition: calcium (190 g/kg), phosphorus (60 g/kg), sulfur (20 g/kg), magnesium (20 g/kg), potassium (35 g/kg), sodium (70 g/kg), cobalt (15 mg/kg), copper (700 mg/kg), chromium (10 mg/kg), iron (700 mg/kg), iodine (40 mg/kg), manganese (1.60 mg/kg), selenium (19 mg/kg), zinc (2.5 mg/kg), fluorine (600 mg/kg), vitamin A (400 IU/kg), vitamin D3 (100 IU/kg), vitamin E (2.4 IU/kg)

model the association among the repeated measurements. The comparison between means was achieved using the least squares means (LSMeans) test, where the significance level was set at 5% for all tests.

## Results

The genetic composition of the female cows was found to have an effect on weight and BCS ( $P = 0.024$  and  $P = 0.010$ , respectively), with 3/4 HG and 7/8 HG cows having a mean weight of  $456.62 \pm 7.90$  and  $429 \pm 8$  kg and a BCS of  $3.13 \pm$

$0.03$  and  $2.94 \pm 0.03$ , respectively (Table 2). For MY and FCM, an effect of the postpartum week ( $P = 0.03$  and  $P = 0.03$ , respectively) was observed (Table 2). The energy corrected milk (ECM) was not affected by any of the analyzed variables ( $P > 0.05$ ) (Table 2).

MY was lower in the first week postpartum, with an increase in MY achieved from the fourth week (Fig. 1). FCM followed the same pattern, with lower results being recorded in the first 2 weeks postpartum and an increase observed from the third week (Fig. 1).

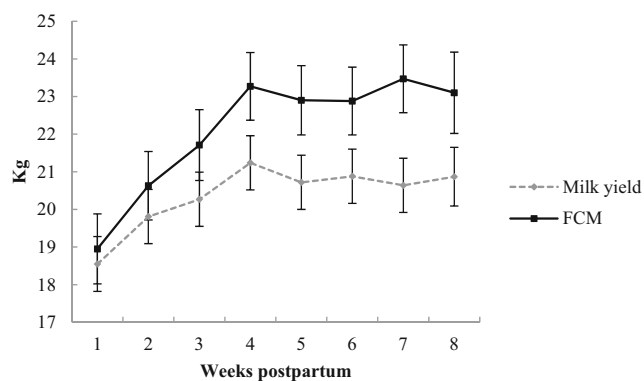
There was a diet effect on the percentages of fat, protein, lactose, and total dry extract in milk ( $P = 0.01$ ,  $P = 0.001$ ,  $P =$

**Table 2** Effect of diet, genetic group, postpartum week on, and their interactions with the weight, body condition, yield, and milk composition of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

	Energy diet		Genetic group		SEM	P value					
	HE	LE	3/4 HG	7/8 HG		Diet	GG	Week	Diet × week	GG × week	GG × diet
Weight (kg)	439	447	457	429	8.5	0.50	<i>0.02</i>	0.05	0.12	0.21	0.77
BCS	3.04	3.03	3.13	2.94	0.03	0.12	<i>0.01</i>	0.12	0.98	0.21	0.16
MY (kg)	19.5	21.2	20.7	20.1	0.8	0.14	0.58	<i>0.03</i>	0.63	0.17	0.49
FCM (kg)	21.7	22.6	22.7	21.5	0.7	0.38	0.25	<i>0.03</i>	0.23	0.80	0.39
ECM (kg)	19.6	20.3	20.5	19.3	0.7	0.46	0.19	0.12	0.29	0.78	0.38
Fat (%)	4.25	3.80	4.05	4.00	0.1	<i>0.01</i>	0.78	0.21	0.33	0.65	0.84
Protein (%)	3.09	2.83	3.06	2.86	0.05	<i>0.001</i>	<i>0.01</i>	<i>&lt;0.0001</i>	0.28	0.36	0.80
Lactose (%)	4.65	4.53	4.59	4.60	0.03	<i>0.01</i>	0.87	0.08	0.72	0.87	0.24
TDE (%)	12.9	12.0	12.5	12.3	0.1	<i>&lt;0.0001</i>	0.34	0.93	0.60	0.83	0.95
F/P	1.37	1.37	1.35	1.38	0.40	0.96	0.53	<i>0.0008</i>	0.19	0.68	0.76

P values in italics indicate statistical significance ( $P < 0.05$ )

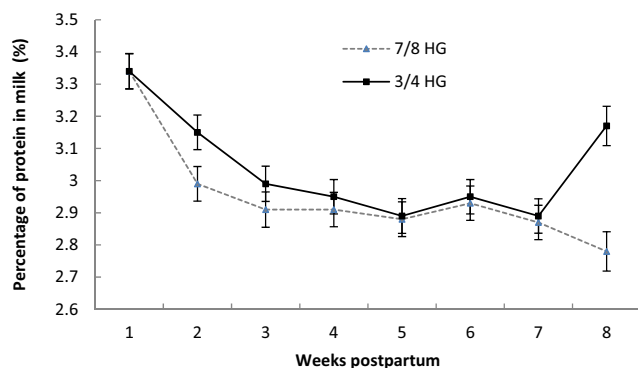
SEM standard error of mean, BCS body condition score, MY milk yield, FCM fat corrected milk, ECM energy corrected milk, TDE total dry extract, F/P fat/protein ratio, GG genetic group effect, Week postpartum week effect, GG × diet genetic group and diet interaction, Diet × week diet and postpartum week interaction, GG × week genetic group and postpartum week interaction



**Fig. 1** Milk yield (MY) and fat corrected milk (FCM) of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

0.01, and  $P < 0.0001$ , respectively) (Table 2). Cows fed the HE diet produced milk with  $4.25 \pm 0.1\%$  fat,  $3.1 \pm 0.05\%$  protein,  $4.65 \pm 0.03\%$  lactose, and  $12.9 \pm 0.1\%$  total dry extract (TDE) in their milk, while cows fed the LE diet produced milk with  $3.8 \pm 0.1\%$  fat,  $2.8 \pm 0.05\%$  protein,  $4.5 \pm 0.03\%$  lactose, and  $11.97 \pm 0.16\%$  TDE (Table 2). In addition, there was a genetic group effect for protein ( $P = 0.01$ ). The 3/4 HG cows had a higher percentage of protein in their milk than the 7/8 HG cows ( $3.1 \pm 0.05$  and  $2.9 \pm 0.05\%$ , respectively). Moreover, there was a postpartum week effect for the percentage of protein in milk ( $P < 0.0001$ ) (Table 2). The fat/protein ratio (F/P) was also affected by the postpartum week ( $P = 0.0008$ ) (Table 2). The highest milk protein production (Fig. 2) was recorded in the first week, and the milk from the primiparous HG exhibited a lower F/P ratio in the first 2 weeks, increasing from the third week (Fig. 3).

Both NEI and EB were found to have dietary effects ( $P = 0.01$  and  $P = 0.005$ , respectively). Cows fed the HE diet had higher NEI ( $95.3 \pm 1.9$  vs.  $88.09 \pm 2.05$  MJ/day) and higher EB ( $3.6 \pm 2.1$  vs.  $-6.0 \pm 2.3$  MJ/day) (Table 3) than others. In addition, genetic group had a significant effect on NEm ( $P = 0.02$ ). The 3/4 HG cows had higher energy for maintenance ( $33.1 \pm 0.4$  MJ/day) than the 7/8 HG ( $33.1 \pm 0.5$  MJ/day)



**Fig. 2** Percentage of protein in milk of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

(Table 3). Moreover, weeks postpartum had an effect on DMI, NEI, and NEIac ( $P = 0.0009$ ,  $P = 0.02$ , and  $P = 0.001$ , respectively) (Table 3). Finally, the CG  $\times$  diet interaction had a significant effect on DMI, NEI, and EB ( $P = 0.02$ ,  $P = 0.02$ , and  $P = 0.004$ , respectively) (Table 3).

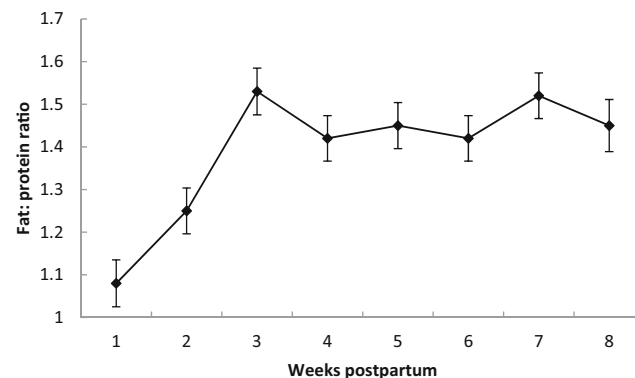
NEI exhibited low levels in the first 2 weeks, followed by an increase and then remaining stable from the fourth week postpartum (Fig. 2). Similarly, DMI showed low levels in the first 2 weeks postpartum and then increased and remained stable from week 4 (Fig. 4). NEIac was lower in the first week of lactation, followed by an increase before reaching a peak in the fourth week (Fig. 5).

The 7/8 HG cows fed the LE diet displayed higher DMI than the 7/8 HG cows fed the HE diet; however, no difference was observed among the 3/4 HG fed the HE or LE diet (Fig. 6). Primiparous 3/4 HG fed the HE diet show the highest NEI compared with the other combinations (Fig. 7). Regarding BE, the 3/4 HG cows fed the LE diet demonstrated the lowest BE in the study (Fig. 8).

## Discussion

The primiparous animals fed the HE diet produced milk with higher levels of protein, fat, lactose, and TDE, which is explained by the higher ruminal availability of propionate, originating from the fermentation of non-fibrous carbohydrates (Delahoy et al. 2003). According to the NRC (2001), the protein content of the diet of cows does not affect the protein content in their milk, unlike non-fibrous carbohydrates as its concentration will lead to the higher production of ruminal propionate. The HE diet possibly provided higher ruminal propionate content since it contained greater amounts of concentrate and therefore of non-fibrous carbohydrates.

Energy deficit leads to increased lipolysis and absorption of mobilized fatty acids from body stores, which results in an increase in fat synthesis in the mammary gland (NRC 2001). In addition, the flow of amino acids to the mammary gland is



**Fig. 3** Fat/protein ratio (F/P) in milk of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

**Table 3** Effect of diet, genetic group, postpartum week on, and their interactions with food consumption and energy balance of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

	Diet		Genetic group		SEM	P value					
	HE	LE	3/4 HG	7/8 HG		Diet	GG	Week	Diet × week	GG × week	GG × diet
DMI (Kg)	11.8	12.4	12.2	12.1	0.27	0.10	0.78	<i>0.0009</i>	0.53	1.0	<i>0.02</i>
NEI (MJ/day)	95.3	88.1	92.3	91.1	2.00	<i>0.01</i>	0.65	<i>0.001</i>	0.56	1.0	<i>0.02</i>
NE <sub>m</sub> (MJ/day)	32.1	32.5	33.1	31.5	1.92	0.49	<i>0.02</i>	0.05	0.12	0.23	0.77
NE <sub>lac</sub> (MJ/day)	59.6	61.6	62.2	59.0	2.13	0.50	0.28	<i>0.02</i>	0.56	0.43	0.35
EB (MJ/day)	3.64	−6.03	−2.97	0.54	2.26	<i>0.004</i>	0.27	0.06	0.76	0.82	<i>0.004</i>

Values of *P* in italics indicate statistical difference ( $P < 0.05$ )

SEM standard error of mean, DMI dry matter intake, NEI net energy intake, NE<sub>m</sub> net energy of maintenance, NE<sub>lac</sub> net energy of lactation, EB energy balance, GG genetic group effect, Week postpartum week effect, GG × diet genetic group and diet interaction, Diet × week diet and postpartum week interaction, GG × week genetic group and postpartum week interaction

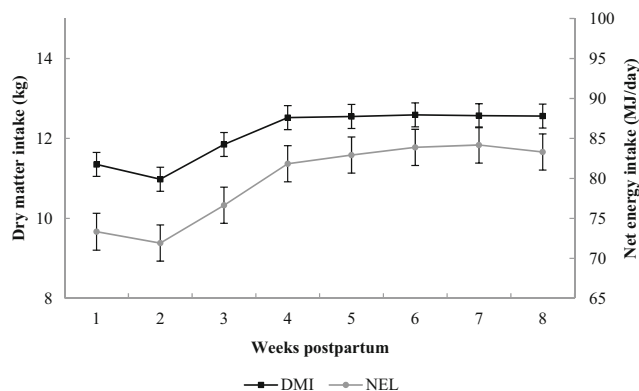
compromised, reducing the protein content in milk (revised by Baumgard et al. 2017) and resulting in a higher F/P ratio. A F/P > 1.5 ratio may indicate high lipolysis and may be a sign of the occurrence of ketosis, abomasal displacement, ovarian cysts, mastitis, and laminitis (Daibert et al. 2018). A lower value may indicate acidosis, and a higher value may indicate ketosis. In Gruber et. al. (1995) study, the F/P ratio was higher in cows that received a LE diet at the beginning of lactation. The authors attributed this fact to the mobilization of adipose tissue caused by insufficient dietary energy and increased milk fat and decreased milk protein content caused by a decrease in the synthesis of microbial proteins in the rumen. In this study, there was no difference in the F/P ratio between the diets, but there was a postpartum week effect, with low results recorded in the first weeks postpartum with a subsequent increase. It is possible that with increased milk production, the primiparous animals need to mobilize greater body reserves to support the production of milk fat. However, there was a reduction in protein percentage, which is indicative of NEB.

Regarding crossbreeding, the 3/4 HG cows produced milk with a higher percentage of protein than the 7/8 HG. These higher percentages agree with the characteristics of the

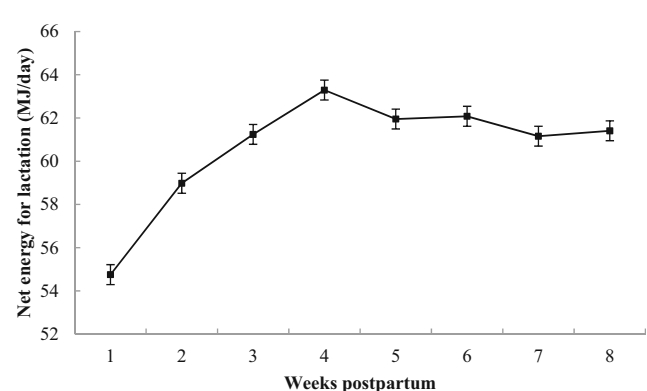
crossbreeding, as identified by Madalena et al. (1990) who observed higher protein and fat yield in milk from 3/4 HG cows as compared with 7/8 HG during the first lactation.

The nutritional needs of cows change abruptly at calving and early lactation. At these times, cows are not able to compensate for burgeoning energy demands through food consumption, resulting in NEB. This NEB, which can start before calving and reach its zenith 2 weeks later, leads to the mobilization of body fat and skeletal muscle (revised by Baumgard et al. 2017). It was found that the more abundant the energy in the diet, the higher the NEI and BE of postpartum primiparous crossbreds. That is, the HE diet was more efficient than the LE diet in providing energy to the cows to meet the increase in energy demand in the early postpartum period. Thus, the BE of the cows fed the HE diet was positive, whereas the BE of the cows fed the LE diet was negative, indicating that they had to mobilize body fat to supply the increased demand for energy.

In this study, there was no difference in MY, FCM, or ECM between the two genetic groups. However, there were differences between the groups regarding weight and BCS. NE<sub>m</sub> is associated with live weight. The primiparous 3/4 HG

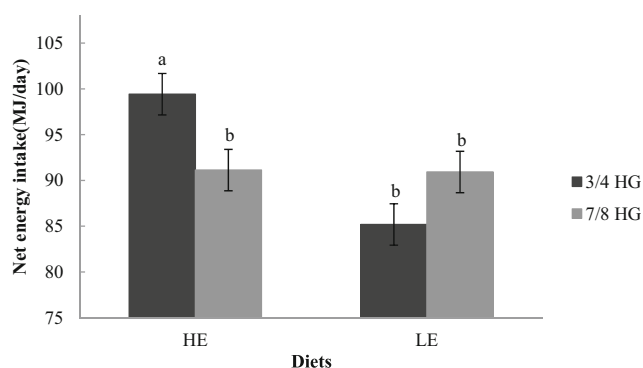


**Fig. 4** Dry matter intake (DMI) and net energy intake (NEI) of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period



**Fig. 5** Net energy of lactation (NE<sub>lac</sub>) of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

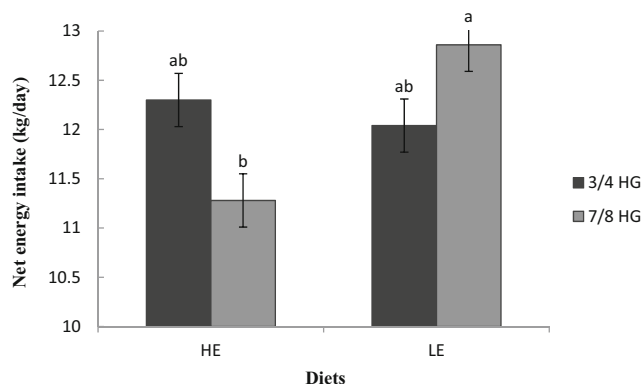




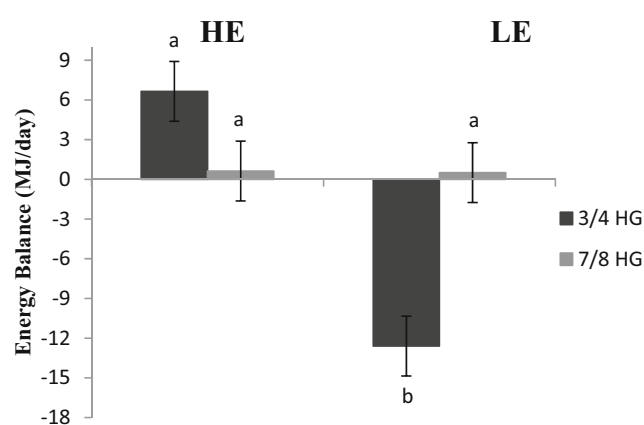
**Fig. 6** Dry matter intake (DMI) of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

presented a greater live weight than the 7/8 HG and therefore higher NEM. Moraes et al. (2015) find that the increase in energy required to maintain cows in modern milk production systems is consistent with the literature describing the increase of heat production during the fasting of cows with the highest genetic merit. In this way, cows with greater productive potential would present higher NEM. Considering that the 7/8 HG cows have a higher proportion of Holstein genes (*Bos taurus*), this result disagrees with that of Oliveira (2015). The Oliveira (2015) study supports the hypothesis that *Bos taurus* dairy cows have higher maintenance energy requirements and greater energy efficiency for milk production than crossbred *Bos taurus* × *Bos indicus* dairy cows. These opposing findings reinforce the need for further studies comparing the energy requirements of different crossbreeds.

DMI and NEI followed the same pattern during the postpartum weeks, with a significant increase in DMI and NEI from the fourth week. When leaving the dry period and after calving, the animal begins lactation and experiences a sudden increase in energy demand for milk production (Grummer 1995). In this early postpartum period, DMI increases gradually to fulfill this demand, as observed in this study. The digested products are assimilated, and the nutrients were allocated to physiological steps that prioritize maintenance requirements and give



**Fig. 7** Net energy intake (NEI) of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period



**Fig. 8** Energy balance of primiparous crossbred cows (3/4 and 7/8 HG) fed low- and high-energy diets during the early postpartum period

secondary importance to productive functions, such as milk synthesis or fetal development. Subsequently, body reserves can be replenished or mobilized to meet the hierarchical objectives of nutrient traffic (Baumgard et al. 2017). Along with the increase in MY, there is also an increase in NE<sub>lac</sub>, as observed in this study. The increase in NE<sub>lac</sub> is accompanied by an increase in PL across the weeks.

The DMI of 7/8 HG cows differed according to the diet they received. Those fed the HE diet exhibited lower DMI than those fed the LE diet. Possibly, the 7/8 HG cows had their energy demands fulfilled faster with the HE diet, reducing daily consumption. Regarding BE, the 3/4 HG cows fed the LE diet showed significant negative BE. Considering the higher live weight and NEM of this group, this poor-energy diet was not able to meet the energy demands of the animals, and as a result, the cows mobilized body fat despite having a BCS than the 7/8 HG.

Under the current conditions of this study, we concluded that the primiparous 3/4 HG were heavier than the 7/8 HG and displayed a higher BCS yet no differences in liters of milk produced up to 60 days postpartum were identified. Moreover, a diet with higher energy content benefits energy balance, resulting in greater production of fat, protein, and lactose when primiparous cows are fed HE diets.

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## Compliance with ethical standards

**Statement of animal rights** The Animal Care Committee of Embrapa Gado de Leite approved the study design (protocol number #03/2012), and it was conducted according to the principles of the Sociedade Brasileira de Ciência em Animais de Laboratório, which regulates conditions for experiments involving animals.

**Conflict of interest** The authors declare that they have no conflict of interest.

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