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# Nutritional, behavioral and performance parameters of F1 Holstein x Zebu cows at different lactation stages

[Parâmetros nutricionais, comportamento ingestivo e desempenho de vacas F1 Holandês x Zebu em diferentes estádios da lactação]

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### **ABSTRACT**

The objective of this study was to evaluate the different lactation stages of F1 Holstein x Zebu cows on intake and digestibility of nutrients, nitrogen use efficiency, feeding behavior and performance. Thirty-six F1 Holstein × Zebu cows with initial body weight (BW) of 482±43kg were used. The early, mid and late lactation stages were characterized after 50±13, 111.5±11.75 and 183.0±17.5 days in milk, respectively. A completely randomized design with three lactation stages and 12 cows in each treatment group was used. Dry matter intake (P=0.01) was higher in late lactation. Milk yield (P<0.01) was 24.17% higher in early lactation than in other stages. Body weight was lowest in mid-lactation cows (465.63kg; P<0.01). The feed efficiency was 23.36% higher in early lactation than in other stages (0.82kg of milk/kg of DM). F1 Holstein x Zebu cows have increased dry matter intake in late lactation. Milk yield and feed efficiency in early lactation were benefited by changes in feeding behavior, such as increased rumination time.

Keywords: crossbreeding, dairy cattle, daily milk yield, feeding behavior, nitrogen balance

### RESUMO

O objetivo deste estudo foi avaliar os diferentes estágios de lactação de vacas F1 Holandês x Zebu quanto ao consumo e à digestibilidade de nutrientes, à eficiência no uso de nitrogênio, ao comportamento ingestivo e ao desempenho. Trinta e seis vacas F1 Holandês × Zebu, com peso corporal inicial (PC) de 482±43kg, foram utilizadas. Os estágios inicial, médio e final da lactação foram caracterizados após 50±13, 111,5±11,75 e 183,0±17,5 dias de lactação, respectivamente. O arranjo experimental adotado foi o delineamento inteiramente ao acaso, com três fases de lactação e 12 vacas em cada grupo de tratamento. O consumo de matéria seca (P=0,01) foi maior no período final da lactação. Na fase inicial da lactação, a produção de leite (P<0,01) foi maior em 24,17% em comparação às demais fases. Na fase intermediária da lactação, as vacas apresentaram menor peso corporal (465,63kg; P<0,01) em relação às demais fases. A eficiência alimentar foi maior em 23,36% na fase inicial da lactação (0,82kg de leite/kg de MS). Vacas F1 Holandês x Zebu aumentam o consumo de matéria seca no período final da lactação. A produção de leite e a eficiência alimentar no início da lactação foram favorecidas por mudanças no comportamento ingestivo, como o aumento do tempo de ruminação.

Palavras-chave: cruzamento, gado leiteiro, produção diária de leite, comportamento ingestivo, balanço de nitrogênio

# INTRODUCTION

Brazil ranks fourth in the world in terms of milk production, with a total of 34.23 billion liters produced in 2016 (Anuário..., 2018). Despite its high production, the national productivity of 1,600 liters per cow per year is lower than the world average (3,500 liters). Most farmers have

chosen to use crossbred Holstein x Zebu cows aiming to alleviate the challenges posed by tropical climate to dairy production. Crossbred cows are responsible for about 80% of the volume of milk produced in the country (Salgado et al., 2016); their characteristics and rusticity (beneficial effects of heterosis) allow flexibility in managing pasture-based milk production systems

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during the summer. On the other hand, in the winter, diets are provided in feed troughs due to shortage of forage caused by seasonality of forage production (Santos *et al.*, 2012; Borges *et al.*, 2019; Monção *et al.*, 2019), especially in semi-arid conditions of Brazil.

The economic results of pasture-based systems can be competitive compared with more intensive systems since feeding represents the most significant costs in dairy production. Moreover, the economic results are dependent on the lactation curve of the animals, characterized by the productivity of cows during lactation (Silva et al., 2019). According to Daltro et al. (2019) and Santos et al. (2014), understanding the dynamics of lactation as well as nutrient intake in crossbred cows at different lactation stages is fundamental in genetic breeding programs because it allows selecting efficient animals. Furthermore, Daltro et al. (2019) emphasize that nutrient intake and lactation curve may vary between and within breeds throughout the lactation cycle due to the effect of heterosis.

Therefore, understanding nutrient intake and digestibility, as well as nutrient use efficiency, feeding behavior and performance of cows at the different lactation stages is an important tool to aid the development of improved management strategies. However, few studies evaluated these dynamics using crossbred F1 Holstein x Zebu cows under semi-arid conditions in Brazil. Based on the above, the objective was to evaluate the

effect of different lactation stages on intake and digestibility of nutrients, nitrogen use efficiency, feeding behavior and performance of F1 Holstein x Zebu cows.

### MATERIAL AND METHODS

All procedures involving animals were approved by the institutional committee on animal use (protocol number 138/2017). The study was carried out at an experimental farm in the municipality of Janaúba, Brazil, at the following geographical coordinates: 43°16′18.2 "W and 15°49′51.5" S.

Thirty-six F1 Holstein × Zebu cows with initial body weight (BW) of 482±43kg (mean±SEM) and mean age of 6 years were used. The early, mid and late lactation stages were characterized after 50±13, 111.5±11.75 and 183.0±17.5 days in milk or postpartum, respectively. A completely randomized design with three lactation stages (early, mid and late) and 12 cows in each treatment group was used. The cows were evaluated for five consecutive days at each lactation stage (early, mid and late). The cows received the same diet throughout the experimental period (Table 1), which was based on the body weight of each cow, maintaining a forage: concentrate ratio of 75:25 (dry matter basis). The diets were offered twice a day, at 8:00 a.m. and at 3:00 p.m. as a Total Mixed Ration (TMR).

Table 1. Chemical composition of ingredients and diet during the experimental period

Item (g/kg DM) <sup>1</sup>	Corn silage	Concentrate	Diet²
Dry matter	502.74	925.92	608.54
Organic matter	962.02	922.31	952.09
Crude protein	68.37	218.25	105.84
NDIN	5.99	12.24	7.55
ADIN	0.79	0.75	0.78
Ether extract	27.77	28.28	27.9
Non-fiber carbohydrates	302.89	371.68	320.09
NDFap	562.98	304.1	498.26
Acid detergent fiber	300.5	72.21	243.43
Lignin	99.58	31.75	82.63
Total digestible nutrients*	561.99	734.16	605.03

<sup>1</sup>Nutrient on dry basis (grams per kilogram) - DM – Dry matter; NDIN - neutral detergent insoluble nitrogen; ADIN – acid detergent insoluble nitrogen; NDFap – neutral detergent fiber corrected for ash and protein; \* National research council - NRC (Nutrient..., 2001).<sup>2</sup> Diet with 75% of roughage and 25% of concentrate.

The TMR contained corn silage as the forage source, which was weighed daily and then mixed to the concentrate. The cows were kept in pens of 16 m²/animal equipped with feeding troughs (1 linear meter) and drinkers (individual capacity of 200 liters). Cows were milked by mechanical milking twice a day, at 7:00 a.m. and 2:00 p.m., in the presence of their calves to stimulate milk letdown. Immediately after milking, calves remained with the cows and were allowed to suckle the residual milk.

The intake was evaluated daily by weighing the feed supplied and refusals. Samples of diets and refusals (5-day sampling) were stored at -20°C for further analysis. Samples of diets, concentrate ingredients, refusals, and feces were analyzed to evaluate feed intake and digestibility. The samples were analyzed for dry matter (DM; method 967.03), ash (method 942.05), CP (CP; method 981.10), and ether extract (EE; method 920.39) according to recommendations of the AOAC (Official..., 1990). The contents of neutral detergent fiber corrected for ash and protein (using heat-stable alpha-amylase without sodium sulfite) (NDFap; Mertens, 2002; Licitra et al., 1996) and acid detergent fiber (ADF) were determined as described by Van Soest et al. (1991), while lignin was determined by treating the acid detergent fiber residue with sulfuric acid at 72%.

Non-fiber carbohydrate (NFC) contents were calculated as proposed by Detmann et al. (2012): NFC (g/kg)=100 – ash - EE - NDFap - CP. The total digestible nutrients (TDN) were estimated using the equation proposed by NRC (Nutrient..., 2001). Samples of diets, concentrate ingredients, refusals, and feces were placed in non-woven fabric bags (100g/m<sup>2</sup>) and incubated in the rumen for 288h for analyzing the indigestible neutral detergent fiber (iNDF) (Detmann et al., 2012; method INCT-CA F-008/1). Bags were incubated in the rumen of two rumen-cannulated crossbred steers weighing 480±30kg, with a mean age of 8 years. The following equation was used to determine the digestibility of each fraction: [(amount of nutrient ingested - amount of nutrient excreted in the feces) x 100]/ amount of nutrient ingested.

Samples of milk from each animal were collected twice a day for five days at each lactation stage. The total amount of milk produced in the morning

and afternoon was also evaluated. Milk quality was evaluated using composite samples formed by samples collected from each cow during morning and afternoon milking, after weighing the milk using a milk meter (Mark V®; Delaval Ltda). The samples were placed in plastic recipients (50.0mL) containing preservative (Bronopol®). The levels of protein, fat, milk urea nitrogen (MUN) and total solids (in g/kg of milk) were determined by infrared absorption spectrophotometry using a milk analyzer (Bentley 2000®; Bentley Instruments Inc.).

On day 3, blood samples were taken from the coccygeal vein using vacuum tubes containing sodium fluoride and potassium oxalate (Glistab® anticoagulant; Labtest Diagnóstica S.A., Lagoa Santa, Brazil) 4 hours after the morning feeding. The samples were centrifuged at 4.000rpm for 20min and the serum obtained was stored in Eppendorf tubes and frozen at -18 °C for further analysis. Plasma urea concentrations were determined by a colorimetric enzymatic method using commercial kits (Ureia 500, Doles® Reagents; Panamá, Brazil). Urine spot samples from all animals at each lactation stage were obtained during spontaneous urination on the 3rd day of sample collection, approximately four hours after feeding. An aliquot of 10mL of the urine sample was filtered and immediately diluted in 40mL of H<sub>2</sub>SO<sub>4</sub> 0.036 N for later analysis of urea and creatinine content, as described by Chizzotti et al. (2007).

The samples were then transferred to Eppendorf tubes and analyzed for urea content using the same methodology used to analyze blood samples. The end-point method was used to determine the creatinine content by means of picrate and acidification with enzymatic methods. Quantification of the daily urinary volume of each animal was calculated by multiplying the individual body weight by the amount of creatinine excreted daily and then dividing the product by the creatinine concentration (mg/L) in the spot sample. The mean value of 24.04 (mg/kg BW) was used to calculate the total daily creatinine excretion (Chizzotti et al., 2007). Total daily urinary volume was estimated to calculate total urinary nitrogen. The amount of nitrogen ingested (N-ingested; g/day) and the amount of N excreted in the feces (N-feces; g/day), urine (Nurine; g/day) and milk (N-milk; g/day) were used to calculate the nitrogen balance (NB). Dietary nitrogen use efficiency (NUE) was calculated by dividing the concentration of nitrogen retained in milk by nitrogen intake inkg/day. Feed efficiency was calculated by dividing the average milk yield (kg/day) by the DM intake (kg/day) (Valadares Filho *et al.*, 2000).

The feeding behavior was assessed on the 4<sup>th</sup> and 5<sup>th</sup> day of sample collection at each lactation stage for 24 hours. At night, the environment was kept under artificial light. A 3-d adaptation period preceded sample collection to allow the animals to adapt to this condition. On the first day, visual observation of each animal was made every 5 minutes for 24 hours to determine the time spent feeding, rumination and resting according to Mezzalira *et al.* (2011). Total chewing time and number of ruminating chews per day were obtained according to Burger *et al.* (2000).

The milk yield (MY) of each cow at each lactation stage was recorded during the five days of sample collection. Milk yield was corrected to 3.5% fat content (FC) using the equation proposed by Sklan *et al.* (1992):  $MY_{3.5} \% = MY \times (0.432 + 0.163 \times FC)$ . A mechanical scale was used to determine the body weight of cows (Valfran, Votuporanga, São Paulo, Brazil).

The initial weight of the animals was set at 10 days post-partum. The cows were weighed on the 5<sup>th</sup> day of sample collection, after evaluation of feeding behavior. Body condition scores (BCS) were assessed weekly by a single technician at the beginning (initial weighing) and the 5<sup>th</sup> day of collection at each lactation stage. A 1 to 5 scale with 0.10-point intervals was used to analyze the BCS, in which 1 represents a very lean cow and 5 an obese cow (adapted from Mishra *et al.*, 2016). The initial BCS of the cows was 3.65±0.10.

The data were analyzed using SAS software version 9.0 (SAS/STAT..., 2008). The PROC UNIVARIATE procedure was used to detect outliers and check the normality of the residuals. Data were analyzed according to the following model:  $Y_{ij}=\mu+LS_i+C_j+IBW+e_{k(ij)}$ ; in which:  $Y_{ij}=the_{ij}=th$  observation on the i-th treatment;

 $\mu$ =overall mean; LC<sub>i</sub>=effect of the i-th lactation stage, with i=1, 2 and 3; C<sub>j</sub>=effect of j-th cow, with j=1, 2, 3, and 36; IBW=Initial body weight as a covariate; e  $_{k(ij)}$ =experimental error associated with all observations (Y  $_{k(ij)}$ ), which is independent, assuming normal distribution with mean zero and variance δ<sub>2</sub>. The means at each lactation stage were compared by the Student-Newman-Keuls (SNK) test. Data are least-square mean values±standard error of the means (SEM) unless indicated otherwise. The mean values were considered different if P<0.10.

### **RESULTS**

Dry matter intake (DMI; P=0.01) was higher in late lactation. The intakes of crude protein (CPI; P=0.93) and neutral detergent fiber corrected for ash and protein (NDFap; P=0.78) and the digestibility of non-fibrous carbohydrate (NFCD; P=0.56) and NDFap (P=0.14) were similar at different stages of lactation (Table 2).

The ether extract intake by cows at mid-lactation was 17.14% lower than the means at early and late lactation (0.41kg/day; P=0.06). The non-fibrous carbohydrate intake (NFCI) in late lactation was 16.03% higher than in early and mid-lactation (4.02kg/day; P=0.04). There was no difference in total digestible nutrient intake (TDNI; P>0.10) and dry matter digestibility (DMD; P=0.02) between early and late lactation (mean of 9.05kg/day and 62.37%, respectively).

The digestibility of crude protein (DCP; P<0.01) and ether extract (DEE; P=0,04) in early lactation was 12.97% and 5.03% higher than in other stages, averaging 54.27% and 82.01%, respectively. There was no difference (P=0.73) between lactation stages on N-ingested (mean of 234.46 g/day). The excretion of N in milk (P=0.01) and urine (P<0.01) was higher in early lactation than in other stages (Table 3). The nitrogen balance in late lactation was 123.5% higher than in early lactation (mean of -4.66 g/day).

Table 2. Nutrient intake and digestibility in F1 Holstein/Zebu cows at different lactation stages

Item <sup>1</sup>	L	actation stage	— SEM <sup>3</sup>	P-value <sup>4</sup>				
Item-	Early	Mid	Late	SEM	r-value			
	Intake							
Dry matter, kg/day	13.72ab	13.27b	14.37a	0.70	0.01			
Dry matter, % BW	2.84ab	2.75b	2.98a	0.12	0.01			
Crude protein, kg/day	1.46	1.46	1.50	0.07	0.93			
Ether extract, kg/day	0.42a	0.35b	0.41a	0.02	0.06			
Non-fibrous carbohydrates, kg/day	4.28b	4.15b	5.02a	0.25	0.04			
NDF <sub>CP</sub> , kg/day	6.97	6.67	6.72	0.31	0.78			
NDF <sub>CP</sub> , % BW	1.44	1.38	1.39	0.05	0.52			
TDN, kg/day	9.00a	7.99b	9.10a	0.36	0.07			
Nutrient digestibility (%)								
Dry matter	63.22a	57.91b	61.53a	1.24	0.02			
Crude protein	62.36a	53.09b	55.45b	1.74	< 0.01			
Ether extract	86.36a	82.16b	81.87b	1.32	0.04			
Non-fibrous carbohydrates	77.37	75.62	78.13	1.67	0.56			
$\mathrm{NDF}_{\mathrm{AP}}$	56.64	53.92	53.71	1.13	0.14			
TDN	65.88a	60.85b	63.43ab	1.12	0.01			

<sup>&</sup>lt;sup>1</sup> NDFap - neutral detergent fiber corrected for ash and protein; TDN - total digestible nutrients; BW - body weight. <sup>2</sup>Lactation stage: Early, 50±12.80 days; Mid, 111.5±11.75 days; and Late lactation, 183±17.25 days. Means followed by equal letters do not differ by Student-Newman-Keuls test (SNK; P>0.05)

Table 3. Balance and nitrogen use efficiency in crossbred F1 Holstein/Zebu cows at different lactation stages

Item <sup>1</sup>	Lactation stage (days) <sup>2</sup>			- SEM <sup>3</sup>	P-value <sup>4</sup>
	Early	Mid	Late	SEM	r-value
N-ingested, g/day	234.42	242.59	226.39	14.37	0.73
N-milk, g/day	69.40 a	60.52 b	55.12 b	3.05	0.01
N-feces, g/day	89.67	114.54	102.64	9.17	0.17
N-urine, g/day	80.02 a	60.43 b	48.80 b	4.70	< 0.01
Nitrogen balance, g/day	-4.66 b	7.09 ab	19.82 a	6.89	0.06
NUE <sup>5</sup>	0.301 a	0.253 b	0.246 b	0.01	< 0.01
UUN, mg/dL	10.55a	9.44a	6.89b	1.09	0.07
PUN, mg/dL	21.82 a	14.52 b	13.09b	0.52	< 0.01
MUN, mg/dL	14.76a	15.15a	12.18b	0.60	< 0.01

<sup>&</sup>lt;sup>1</sup>N- nitrogen; BW – body weight; NUE – Nitrogen use efficiency; UUN – urine urea nitrogen; PUN – plasma urea nitrogen; MUN – milk urea nitrogen.

Cows in early lactation were 17.10% more efficient in terms of nitrogen use than cows in other lactation stages (mean of 0.25; P<0.01). The excretion of plasma nitrogen was higher in early lactation compared with other stages (mean of 18.17mg/dL; P<0.01). The feeding time of cows was not affected (P=0.30) by lactation stage, with

a mean of 5.02 hours/day (Table 4). Cows in early lactation spent an additional 1.31 hours/day ruminating compared with other stages (mean 6.87 hour/day; P<0.01). The resting time in late lactation was 18.37% higher than in early lactation (10.13 hour/day; P=0.05).

<sup>&</sup>lt;sup>3</sup>SEM – standard error of the mean.

<sup>&</sup>lt;sup>4</sup>P – Probability.

<sup>&</sup>lt;sup>2</sup>Lactation stage: Early, 50±12.80 days; Mid, 111.5±11.75 days; and Late lactation, 183±17.25 days. Means followed by equal letters do not differ by Student-Newman-Keuls test (SNK; P>0.05)

<sup>&</sup>lt;sup>3</sup>SEM – standard error of the mean.

<sup>&</sup>lt;sup>4</sup>P – Probability.

Table 4. Feeding behavior of F1 Holstein/Zebu cows at different lactation stages

Itam	Lac	Lactation stage (days) <sup>1</sup>			D13
Item	Early	Mid	Late	– SEM²	P-value <sup>3</sup>
Feeding, hour/day	5.66	4.91	4.50	0.52	0.30
Ruminating, hour/day	8.20 a	6.88 b	6.87 b	0.32	0.01
Resting, hour/day	10.13b	12.20a	12.62a	0.73	0.05
Chewing					
number/bolus	53.21	45.85	52.04	3.46	0.28
number/min	60.05b	66.68a	63.85ab	1.4	< 0.01
hours/day	13.86 a	11.79 b	11.37 b	0.73	0.05
Feeding efficiency DM, g/hour	2569	3125	3278	246	0.11
Ruminating efficiency DM, g/hour	1711b	2008a	1974a	95	0.06

 $<sup>^{1}</sup>$ Lactation stage: Early,  $50\pm12.80$  days; Mid,  $111.5\pm11.75$  days; and Late lactation,  $183\pm17.25$  days. Means followed by equal letters do not differ by Student-Newman-Keuls test (SNK; P>0.05)

Regardless of the lactation stage, the number of chews per bolus was 50.36 (P=0.28). There were more chews in mid-lactation, averaging 66.68 chews/minute (P<0.01). In early lactation, the cows spent an additional 2.28 hours/day chewing compared with other stages (mean of 11.58 hours/day; P=0.05). There was no difference in feeding efficiency of DM (P=0.11). The highest rumination efficiency of DM was observed in mid-lactation (P=0.06).

Milk yield (P<0.01) was 24.17% higher in early lactation than in other stages (mean of 11.02kg/day; Table 5). The lowest 3.5% fat-corrected milk yield was observed in late lactation (P<0.01). The highest fat content and total solids in the milk were observed in mid-lactation, while the milk protein content was lowest in early lactation.

Table 5. Performance and feed efficiency of F1 Holstein/Zebu cows at different lactation stages

Item	Lac	tation stage (	- SEM²	D volvo3	
Item	Early	Mid	Late	- SEM	P-value <sup>3</sup>
Milk yield, kg/day	14.54 a	11.78 b	10.27 b	0.56	< 0.01
3.5% fat-corrected milk yield, kg/day	14.03 a	12.07 b	9.37 c	0.54	< 0.01
Fat, (g/kg milk)	33.1 ab	36.8 a	29.8 b	2.00	0.06
Protein, (g/kg milk)	30.4 a	33.1 b	34.0 b	0.90	0.03
Total solids, (g/kg milk)	120.1ab	127.0a	118.8b	2.70	0.09
Final body weight, kg	492.82 b	465.63 c	504.90 a	4.90	< 0.01
Difference in body weight, kg	6.99 a	-35.19 b	52.57 a	12.50	< 0.01
Final body condition score	3.79	3.83	3.69	0.06	0.32
Difference in body condition score	0.15	-0.02	0.13	0.06	0.16
Feed efficiency, kg of milk/kg of DM	1.07 a	0.87 b	0.77 b	0.05	< 0.01

<sup>&</sup>lt;sup>1</sup>Lactation stage: Early, 50±12.80 days; Mid, 111.5±11.75 days; and Late lactation, 183±17.25 days. Means followed by equal letters do not differ by Student-Newman-Keuls test (SNK; P>0.05)

The body weight of cows was lower in midlactation than in other stages. Cows in midlactation lost (P<0.01) 35.19kg in relation to their initial weight. On the other hand, late-lactation cows gained 52.57kg in relation to their initial body weight. There was no change in the body condition score (mean of 3.7; P=0.32). Cows in early lactation had a feed efficiency 23.36% higher compared with other lactation stages (mean of 0.82kg of milk/kg of DM).

<sup>&</sup>lt;sup>2</sup>SEM – standard error of the mean.

<sup>&</sup>lt;sup>3</sup>P – Probability.

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<sup>&</sup>lt;sup>3</sup>P – Probability

# **DISCUSSION**

Several physical and physiological changes occur during the transition period of dairy cows (from 21 days before to 21 days postpartum), modifying animal behavior. The fetus grows rapidly in the last 21 days of pregnancy, which consequently increases pressure on organs responsible for digestive and metabolic processes (i.e., rumen and liver), reducing the space available for feed (Franzoni et al., 2018). Concomitantly, there is also an increase in blood concentrations of estrogen, corticosteroids and reduction in concentrations of progesterone (Chew et al., 1979; Franzoni et al., 2018). The associations of these factors may reduce dry matter intake by up to 30%, predisposing cows to a negative energy balance in early lactation, with consequent mobilization of energy reserves (Grum et al., 1996).

However, the DMI increases up to 75 days postpartum (characterized as limit for early lactation in pure Holstein cows) but the maximum DM intake occurs at mid-lactation, with reduction in the subsequent lactation stage (Gulay et al., 2003). In crossbred cows, especially F1 Holstein x Zebu, this behavior in the DMI seems to be less marked compared to pure Holstein cows since the DMI was 7.65% higher in late lactation compared with other stages (mean of 13.49kg/day). Santos et al. (2012, 2014) observed a quadratic behavior of lactation stage on DMI in F1 Holstein x Zebu cows kept on pasture, reaching its peak at 40 days post-partum (mean of 13.34kg/day), while in this study the DMI was 13.72kg/day using confined cows (dietary NDF of 498.26g/kg DM).

Even with intermediate DMI values, milk yield was 24.17% higher in early lactation compared with other stages (mean of 11.02kg/day). In Holstein cows, milk yield also increases in early lactation (Gulay et al., 2003; Lopez et al., 2015), but there is a reduction in body weight. This increase in milk yield is linked to peak lactation, which occurs after 60±10 days post-partum in Holstein cows (Keown et al., 1986; Gulay et al., 2003) and 30±8 days in F1 Holstein x Zebu cows (Santos et al., 2014). According to Forbes (1995), the level of intake is among the main factors affecting feed digestibility and milk production. The higher milk yield at early lactation may be related to greater nitrogen use efficiency, longer rumination time (8.20 hours/day) and chewing,

which certainly favored particle size reduction of substrate in the rumen. Cows spent more time resting (12.41 hours) in mid and late lactation than in early lactation (10.13 hours). This response is associated with lactogenesis (level of prolactin), which occurs with greater intensity in early lactation (Lacasse *et al.*, 2011; Ollier *et al.*, 2013, 2014).

In this study, the lowest body weight was observed in cows at mid-lactation. According to Buckley et al. (2000), increased weight loss during the 45-day postpartum period is associated with expulsion of the fetus, placenta and other uterine contents, followed by gradual decrease in body weight due to mobilization of adipose tissues. It explains the lower body weight and beta-hydroxybutyrate (BHB) concentration in mid-lactation and, consequently, concentrations of non-esterified fatty acids (NEFA) (Lacasse et al., 2011). In Holstein cows, Bauman and Currie (1980) reported that energy from body fat stores can contribute to approximately 33% of milk production, explaining the higher productivity during the first 60 days of lactation. The catabolism of adipose tissue can support the production of 120 to 550kg of milk during the first weeks of lactation. However, in F1 Holstein x Zebu cows, the metabolism of adipose tissue was not significantly affected since there was no change in BCS, averaging 3.77.

According to Ferreira et al. (2000), monitoring postpartum body weight in lactating cows is fundamental to ensure that body reserves will be adequate for cows to exhibit estrus and guarantee pregnancy. Furthermore, the authors reported that a BCS of 3.5 is ideal for cows to show estrus within 90 days postpartum. For this condition, it is necessary a body condition score above 3.0 in prepartum, or final third of lactation. In our study, the average BCS of 3.77 indicates that cows had sufficient energy reserves during lactation, which is a positive characteristic of F1 Holstein x Zebu cows. Ferreira et al. (2000) reported that the desirable BCS in late lactation, close to the dry period of the cow, should be approximately 3.5, while at calving it should be 3.5 to 4.0. This assessment in late lactation is essential to prevent some cows from becoming too thin at drying off. Under this condition, there is not enough time to regain the necessary body reserves during the dry

period, aiming for a good body condition at calving (Ferreira et al. (2000).

Feed efficiency was improved in cows at early lactation due to higher milk yield and 3.5%-fat corrected milk yield. The lowest 3.5%-fat corrected milk yield was reported in late lactation, as well as lower amounts of fat and total solids in the milk. Evaluating the lactation curve in F1 Holstein x Zebu cows, Silva *et al.* (2015) and Glória *et al.* (2012) reported a reduction in milk yield in late lactation, which can be explained by the natural reduction in prolactin synthesis. Thus, in F1 Holstein x Zebu cows, Santana *et al.* (2019) recommended a restriction of dietary supply up to 2% of body weight (on dry matter basis) at this stage (late lactation) as a cost-reduction strategy.

### **CONCLUSION**

F1 Holstein x Zebu cows have increased dry matter intake in late lactation. However, milk yield and feed efficiency in early lactation are benefited by changes in feeding behavior, such as increased rumination time.

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