## RESEARCH



# Ingestive behavior of grazing heifers receiving crude glycerin supplementation during the dry-rainy season transition

Vitor Visintin Silva de Almeida<sup>1,2\*</sup>, Robério Rodrigues Silva<sup>1</sup>, Aline Cardoso Oliveira Visintin<sup>3</sup>, Augusto Cesar de Queiroz<sup>4</sup>, Fabiano Ferreira da Silva<sup>1</sup>, Cláudia Batista Sampaio<sup>4</sup>, Mateus de Melo Lisboa<sup>1</sup>, Fabrício Bacelar Lima Mendes<sup>1</sup>, and Tulio Otávio Jardim D'Almeida Lins<sup>1</sup>

The search to find food alternatives for corn and feeding alternatives that reduce the cost of production has been constant. The use of oleaginous grains to produce ethanol and biodiesel has produced an excess of byproducts, especially biodiesel which produces crude glycerin. The objective of this study was to evaluate the effect of including crude glycerin on the ingestive behavior of crossbred heifers supplemented with pasture. Thirty-six crossbred heifers with an initial mean weight of  $301.5 \pm 31$  kg were distributed in a completely randomized design with four treatments and nine replicates per treatment. The animals were kept in a *Brachiaria brizantha* (Hochst. ex A. Rich.) Stapf 'Marandu' pasture in a rotational-grazing system. The treatments tested were 0.0%, 3.33%, 6.66%, and 9.99% crude glycerin included in total DM to replace corn (*Zea mays* L.) Including crude glycerin reduced the time for grazing and eating at the trough and increased idle time. Rumination was quadratically influenced. Eating time (min kg<sup>-1</sup> DM and neutral detergent fiber [NDF]) was reduced, while rumination time (min kg<sup>-1</sup> DM and NDF) was quadratically affected by adding glycerin. The variables, time spent per ruminated bolus and the number of chews per day, were not affected. The number of boluses per day showed a quadratic effect. The number of grazing, idle, ruminating, and eating at the trough periods were not affected by including crude glycerin; means were 15.1, 24.9, and 13.3 and 3.71 periods d<sup>-1</sup>, respectively. Feed and rumination efficiency (kg h<sup>-1</sup> DM and NDF) increased when crude glycerin was included in the diet. Including crude glycerin promotes feed and rumination efficiency in grazing heifers.

Key words: Brachiaria brizantha, chewing, eating, feed efficiency, rumination efficiency.

## INTRODUCTION

The cattle production system in Brazil is based on pasture, which is the basal resource with the lowest cost. However, production is not constant due to climatic variations which change forage availability throughout the year with quantitative and qualitative alterations in forage, especially during the dry period. Over the dry period, forage presents lower quality as regards its crude protein contents and high fiber levels on the cell wall, which impairs the performance of animals on pasture. Thus, strategic supplementation is an alternative for increasing the potentially digestible

<sup>4</sup>Universidade Federal de Viçosa – UFV, Departamento de Zootecnia, Viçosa, MG 36570.000, Brasil. *Received: 26 November 2013. Accepted: 20 May 2014.* doi:10.4067/S0718-58392014000300006 DM and digestibility of available forage, which results in better animal productive performance. The main effects of supplementation are observed on forage intake and digestibility as a result of changes in the ruminal environment and microbial population, which affect the determinant factors of ruminal digestion, digesta flow through the rumen, and availability of nutrients for absorption in the intestine (Ospina et al., 2003). Ingestion of potentially digestible DM (pdDM) plays an important role in animal performance because it is responsible for nutrient uptake to meet the nutritional requirements for maintenance and production. Using supplementation to improve intake and consequently animal performance is an interesting strategy, but it can significantly increase production costs.

Alternative feeds can be used in the supplementation of ruminants to substitute part of the concentrate to reduce production costs without affecting feed intake or animal performance. In Brazil, crude glycerin has been used for this purpose. Glycerin is a co-product of the biodiesel industry (Abdalla et al., 2008) and every 100 m<sup>3</sup> of biodiesel that is produced generates 10 m<sup>3</sup> of glycerin (Dasari et al., 2005). Biodiesel production, a renewable energy source, has gained worldwide prominence due

<sup>&</sup>lt;sup>1</sup>Universidade Estadual do Sudoeste da Bahia UESB, Praça Primavera, 40, Bairro Primavera, Itapetinga-BA, CEP 45700-000. <sup>\*</sup>Corresponding author (rrsilva.uesb@hotmail.com).

<sup>&</sup>lt;sup>2</sup>Instituto Federal de Educação, Ciência e Tecnologia Baiano – IFBAIANO, Campus Santa Inês, Santa Inês, BA, 45320-000, Brasil.
<sup>3</sup>Universidade Federal de Alagoas, Departamento de Zootecnia, Arapiraca, AL 57310540, Brasil.

to the concern about environmental damage caused by burning fossil fuels. The chemical composition of glycerin is highly variable since it depends on the raw material used for biodiesel production (Behr et al., 2008). It is hygroscopic in nature, which can increase water-holding capacity in low-moisture environments and improve palatability of the concentrate due to its mild aroma and sweet flavor, as well as increasing concentrate intake (Elam et al., 2008). Therefore, it can be considered as an excellent alternative in the composition of supplements for animals raised on pasture.

Providing concentrate usually changes ingestive behavior (grazing, rumination, and idle times) of grazing animals (Krysl and Hess, 1993). Animals under supplementation travel longer daily distances and choose forage better; they are therefore more selective compared with animals that only graze (Adams, 1985). According to Silva et al. (2010), results found in the literature are controversial regarding alterations caused by supplementation of grazing ruminants on their ingestive behavior.

Thus, the objective of this study was to evaluate the ingestive behavior of crossbred heifers on *Brachiaria brizantha* (Hochst. ex A. Rich.) Stapf pasture supplemented with glycerin levels in the concentrate during the dry-rainy season transition.

### MATERIAL AND METHODS

The experiment was conducted on the Princesa do Mateiro Farm located in Ribeirão do Lago (15°25' S, 40°40' W, 782 m a.s.l.), Bahia, Brazil. Thirty-six 11-mo-old crossbred heifers with an initial mean weight of  $301.5 \pm$ 31 kg were included in the experiment. The experimental period was 84 d; animals spent 14 d adapting to the management system and experimental diets. Animals were kept on a rotational-grazing pasture production system in *Brachiaria brizantha* (Hochst. ex A. Rich.) Stapf 'Marandú' over an area of 14 ha divided into eight same-sized paddocks.

The tested treatments were G0, G3.33, G6.66, and G9.99, which corresponded to diets with 0%, 3.33%, 6.66%, and 9.99%, respectively, of crude glycerin included in total DM to substitute corn. Diets were formulated according to NRC (2001) and contained equal amounts of energy and protein with a 60%:40% roughage:concentrate ratio. The concentrate was supplied daily at 10:00 h (Table 1). Pasture was evaluated every 28 d to estimate DM availability (Figure 1) by collecting 12 samples at soil level with a 0.25 m<sup>2</sup> frame according to the methodology described by McMeniman (1997). Eight paddocks, deferred in May, were used. To reduce the influence of biomass variation among paddocks, heifers remained in each paddock for 7 d; after this period, they were transferred to another paddock in a randomly preestablished order. Before samples were collected, DM

Table 1. Centesimal composition of the supplements (as fed basis).

	Glycerin inclusion (% DM)							
Ingredients (%)	0.0	3.33	6.66	9.99				
Corn grain	90.7	78.8	66.5	55.0				
Soybean meal	2.9	5.0	7.2	9.3				
Glycerin	0.0	9.7	19.6	28.9				
Urea	2.9	2.9	2.9	3.0				
Mineral <sup>1</sup>	1.5	1.5	1.6	1.6				
Limestone	1.4	1.4	1.3	1.3				
Phosphate	0.6	0.7	0.9	1.0				

<sup>1</sup>Centesimal composition: NaCl: 47.15; dicalcium phosphate: 50; zinc sulfate: 1.5; copper sulfate: 0.75; cobalt sulfate: 0.05; potassium iodate: 0.05; magnesium sulfate: 0.5.

biomass from the sample was visually estimated. With the values of the cut and visually estimated samples after the frame was cast 50 times, forage biomass was calculated (kg ha<sup>-1</sup>) by the equation proposed by Gardner (1986). The triple-pairing technique (Moraes et al., 1990) was used to study the accumulation of biomass over time and with the four paddocks that remained sealed for 28 d functioning as controls. The accumulation of DM in the different experimental periods was calculated by multiplying the daily accumulation rate (DAR) by the number of days in the period.

The DM daily accumulation rate (DAR) was estimated by the equation proposed by Campbell (1966):

$$DARj = (G_i - F_{i-1})/n$$

where  $DAR_j$  is the daily accumulation rate of DM in period *j* (kg DM ha<sup>-1</sup> d<sup>-1</sup>); *G<sub>i</sub>* is the mean final DM of the four empty paddocks at instant *i* (kg DM ha<sup>-1</sup>); *F<sub>i</sub>* is initial DM in the empty paddocks at instant *i* - *I* (kg DM ha<sup>-1</sup>); and *n* is the number of days in period *j*.

The potentially digestible DM (pdDM) of the pasture was estimated as described by Paulino et al. (2006):

## pdDM = 0.98 (100 - %NDF) + (%NDF - %iNDF)

where NDF is neutral detergent fiber and iNDF is indigestible neutral detergent fiber.

Chromic oxide was used as an external marker to estimate fecal production. The marker was supplied daily at 09:00 h in a single 10 g dose conditioned in curlpaper for 12 d, that is, 7 d for the adaptation and regulation of the marker excretion flow and 5 d to collect the feces. Feces



Figure 1. Availability of total dry matter (TDM), potentially digestible dry matter (pdDM), leaf, stem, and dead material of *Brachiaria* brizantha.

were collected directly from the rectal ampulla once a day for 5 d when the marker was administered. They were then stored in a cold chamber at -10 °C and later analyzed by atomic absorption spectrophotometry for chromium concentration. Fecal excretion (FE) was calculated based on the ratio between the amount of marker supplied and its concentration in the feces according to the equation:

## $FE = (CrSupplied/CrFeces) \times 100$

where CrSupplied is the quantity of chromium supplied (g) and CrFeces is the concentration of the marker in the feces (%).

Concentrate (supplement) DM intake (SDMI) was estimated with the titanium dioxide marker, which was supplied at 10 g per animal mixed in the concentrate for 12 d according to the procedure described by Valadares Filho et al. (2006), the same feces-collection scheme described for the chromic oxide was followed and expressed by the equation:

## $SDMI = (FE \times TiOFeces)/TiOSupplement$

where FE is fecal excretion and TiOFeces and TiOSupplement are the concentration of titanium dioxide in the feces and supplement, respectively.

The titanium concentration was determined by acid digestion with sulfuric acid at 400 °C followed by adding hydrogen peroxide 30%, transferring to a volumetric flask, completing the volume to 100 mL, and filtering to obtain the solution. The reading was taken in an atomic absorption spectrometer at the Animal Nutrition Laboratory, Departamento de Zootecnia, Universidade Federal de Viçosa (UFV). Indigestible NDF (iNDF) was used as an internal marker to estimate voluntary roughage DM intake. The marker was obtained after ruminal incubation of 0.5 g samples of feed, orts, and feces in nonwoven textile (TNT) bags (grammage 100 [100 g  $m^2$ ], 5 × 5 cm) for 240 h (Casali, 2006). The remaining incubation material was subjected to extraction with neutral detergent to determine iNDF. Roughage DM intake (DMI) was calculated as follows:

DMI (kg d<sup>-1</sup>) = [(FE × CMF) - SI/CMR] + SDMI where FE is fecal excretion (kg d<sup>-1</sup>) obtained with chromic oxide, CMF is the concentration of the marker in the feces (kg kg<sup>-1</sup>), and CMR is the concentration of the marker in the roughage (kg kg<sup>-1</sup>).

The DM, ash, crude protein (CP), and ether extract (EE) contents in the samples of feed, orts, and feces were analyzed according to Silva and Queiroz (2002). Organic matter (OM) content was estimated by subtracting ash from DM content. Total carbohydrates (TC) were estimated according to Sniffen et al. (1992) as follows:

TC = 100 - (%CP + %EE + %ash)

The non-fibrous carbohydrates corrected for the residual ash and protein (NFCap) were calculated as proposed by Hall (2003):

NFCap = (100 - %NDFap - %CP - %EE - %ash)

Total digestible nutrients (TDN) were calculated according to Weiss (1999) with NDF and NFC corrected

for the residual ash and protein contents by the equation:

TDN (%) = DCP + DNDFap + DNFCap + 2.25DEE where DCP is digestible crude protein, DNDFap is digestible NDFap, DNFCap is digestible NFCap, and DEE is digestible EE. Estimated total digestible nutrients (estTDN) of feeds and total diets were calculated according to equations described by the NRC (2001).

Observations regarding animal behavior were done visually during two 24-h periods at 5-min intervals. The observed and recorded behavioral variables were: idle, rumination, grazing, and eating at the trough (hereafter referred to as trough); eating and rumination times were calculated as a function of DM and NDF intakes (min kg<sup>-1</sup> DM or NDF). The number chew per bolus and time spent ruminating each bolus was counted for each animal with a stopwatch. To obtain the mean number of chews and time, three boluses were observed during three different periods of the day (09:00-12:00 h, 15:00-18:00 h, and 19:00-21:00 h) according to Burger et al. (2000). As previously described, total rumination time was divided by mean time spent on ruminating each bolus to determine the number of daily boluses. The discretization of time series was performed on the datacollection spreadsheets by counting the discrete periods eating, ruminating, and idle. The mean duration of each of the discrete periods was obtained by dividing daily times spent on each of the activities by the number of discrete periods of the same activity as described by Silva et al. (2006). The g DM and NDF/meal variables were obtained by dividing mean individual intake of each fraction by the number of eating periods per day (in 24 h). Feed and rumination efficiency, expressed as g DM h<sup>-1</sup> and g NDF h<sup>-1</sup>, was obtained by dividing mean daily DM and NDF intake by total time spent eating and/or ruminating in 24 h, respectively. The g DM and NDF per bolus variables were obtained by dividing mean individual intake of each fraction by the number of ruminated boluses per day (in 24 h).

Bite rate (BITR) of the animals on each type of supplementation was estimated as the time spent by the animal to perform 20 bites (Hodgson, 1982). To calculate bite mass (BITM), daily intake was divided by total daily bites (Jamieson and Hodgson, 1979). Results of the observations of bites and swallowed bolus were recorded six times during the day according to Baggio et al. (2009), that is, three in the morning and three in the afternoon; these were also used to determine the number of bites per day (BITDAY), which is the product of the bite rate and grazing time.

Data were evaluated by variance and regression analyses with the SAEG software (Sistema de Análises Estatísticas e Genéticas) (UFV, 2000). The statistical models were chosen according to the significance of the regression coefficients by using the t-test at 5% probability and determination coefficient ( $r^2$ ) and the studied biological phenomenon.

## **RESULTS AND DISCUSSION**

The DM and NDF intakes and time spent on behavioral activities are shown in Table 3. Including crude glycerin in the diet did not change (P > 0.05) DM or NDF intakes, which had means of 7.68 and 3.03 kg, respectively. Grazing time decreased (P < 0.05) from 507.2 to 432.2 min when glycerin was included. Despite the small reduction (75 min), this behavior indicates that animals that were fed control treatments grazed more time to ingest the same amount of DM than animals that were fed glycerin. In the dry-rainy season transition, pasture had lower leaf availability and high amounts of stem and dead material (Figure 1). In this situation, animals tend to graze longer, seeking the more digestible parts of the plants to meet their energy requirements. The intake regulation mechanism is complex and includes physical limitations, physiological control, and psychogenic factors. Physiological factors include controlling hunger and satiety by the hypothalamic region of the brain (Doughterty and Collins, 2003).

Crude glycerin used in this study consisted of approximately 80% glycerol, a gluconeogenic substrate metabolized to propionate in the rumen (Trabue et al., 2007). According to Benson et al. (2002), propionate is probably the first to signal the end of meals because its flow toward the liver significantly increases during meals and increases ATP production because it produces glucose, which indicates satiety. This indicates that animals that were fed crude glycerin exhibited momentary satiety caused by rapid energy uptake; this extended their idle time, which was compensated by more intense grazing in which the animals were able to increase forage mass per bite when grazing (Table 4) and maintaining the same DM intake. Time spent eating at the trough (Table 3) increased linearly (P < 0.05) as glycerin was included in the diet. This behavior can be justified by the fact that crude glycerin is too viscous; when it was mixed with the diet, this facilitated biting and swallowing the supplement and promoted ingesting the same amount of concentrate in less time. Rumination time (Table 3) had a quadratic response (P < 0.01) with an estimated maximum value of 379.29 min for the 3.51% level of glycerin inclusion.

When glycerin was included in the concentrate, there was a small decrease in dietary NDF contents (Table 2) because glycerin, added to replace corn, is devoid of fiber. Thus, higher levels of glycerin cause small reductions in rumination time since NDF content is positively correlated with time spent on this activity. Including crude glycerin increased (P < 0.05) idle time, which can be explained by the exclusive nature of each of the activities, that is, an animal cannot be engaged in more than one activity at the same time; this could generate competition between the animal's ingestive activities in time distribution (Fischer et al., 1998). This behavior is in line with results shown by the grazing and trough variables.

Eating (grazing + trough) times (min kg<sup>-1</sup> DM and NDF) decreased linearly (P < 0.05) as glycerin was included in the diet. This indicates that, as glycerin is included, animals took longer to ingest the same amount of DM or NDF. This decrease is largely due to the 53% decrease in trough time when glycerin was included in the diet.

Glycerin inclusion had a quadratic effect (P < 0.05) on rumination time in min kg<sup>-1</sup> DM and NDF, whose peaks were at 2.76% and 3.39% crude glycerin

Table 2. Chemical composition of *Brachiaria brizantha*, concentrates and total diets.

		Gly	cerin incl	usion (%	DM)
Ingredients (%)	B.brizantha	0.0	3.33	6.66	9.99
Dry matter	27.94	92.30	93.20	93.00	92.15
Crude protein	7.78	17.68	17.94	18.30	18.55
Ether extract	2.15	2.53	5.36	8.62	11.08
Total carbohydrates	80.54	74.84	71.41	67.29	63.94
Non-fibrous carbohydrates	22.09	63.82	62.51	60.50	57.74
NDFap <sup>1</sup>	62.92	11.02	8.90	6.79	6.20
ADF <sup>2</sup>	33.45	4.72	4.63	4.57	3.65
Ash	9.37	4.95	5.29	5.79	6.43
TDN <sup>3</sup>	64.89	84.60	88.37	92.61	95.19
Total diets					
Crude protein		12.17	12.25	12.23	12.50
Ether extract		2.28	3.27	4.42	5.28
Total carbohydrates		79.51	78.52	77.36	76.34
Non-fibrous carbohydrates		39.57	38.87	37.38	37.59
NDFap <sup>1</sup>		39.94	39.65	39.98	38.75
ADF <sup>2</sup>		23.31	22.74	22.46	21.78
Ash		6.04	5.96	5.99	5.88
TDN <sup>3</sup>		72.94	74.35	75.73	77.20

<sup>1</sup>NDFap: Neutral detergent fiber corrected for ash and protein.

<sup>2</sup>ADF: Acid detergent fiber.

<sup>3</sup>Estimated according to the NRC (2001).

Table 3. Dry matter (DMI) and neutral detergent fiber (NDFI) intakes and time spent grazing, ruminating, idle, and eating at the trough by heifers fed supplements with increasing glycerin levels.

	Glycerin inclusion (% DM)					CV		
Item	0.0	3.33	6.66	9.99	Regression equation	(%)	$r^2$	$p^1$
DMI, kg d <sup>-1</sup>	7.48	7.35	8.16	7.71	$\bar{Y} = 7.68$	10.6	-	NS
NDFI, kg d <sup>-1</sup>	3.04	2.82	3.26	3.02	$\bar{Y} = 3.03$	12.5	-	NS
Grazing, min d-1	507.2	512.5	428.0	432.2	$\hat{\mathbf{Y}} = 516.417 - 9.29263 \mathbf{x}$	8.3	0.75	***
Idle, min d-1	533.8	510.2	621.3	683.3	$\hat{\mathbf{Y}} = 503.306 + 16.8001 \mathbf{x}$	9.5	0.82	***
Rumination, min d-1	358.8	375.5	365.5	304.4	$\hat{\mathbf{Y}} = 357.667 + 12.3123 \mathrm{x} - 1.75351 \mathrm{x}^2$	11.3	0.99	***
Trough, min d-1	42.5	44.1	26.9	22.7	$\hat{\mathbf{Y}} = 45.5556 - 2.2939 \mathbf{x}$	36.7	0.85	***
Eating, min kg-1 DM	73.9	76.9	56.0	59.4	$\hat{\mathbf{Y}} = 76.2655 - 1.93084 \mathbf{x}$	13.4	0.64	***
Rumination, min kg-1 DM	47.9	52.1	45.0	39.9	$\hat{\mathbf{Y}} = 48.5785 + 1.15369x - 0.208918x^2$	15.3	0.89	**
Eating, min kg-1 NDF	182.8	202.5	140.1	151.8	$\hat{\mathbf{Y}} = 192.626 - 4.66179 \mathbf{x}$	15.1	0.49	***
Rumination, min kg-1 NDF	118.0	137.3	112.9	102.0	$\hat{Y} = 120.925 + 4.61023x - 0.67958x^2$	17.0	0.75	***

<sup>1</sup>NS: Non significant; <sup>\*</sup>P < 0.1; <sup>\*\*</sup>P < 0.05; <sup>\*\*\*</sup>P < 0.01.

inclusion, respectively (Table 3); this demonstrates that lower glycerin levels in the concentrate promote longer rumination times per kg DM or NDF because NDF contents are higher in these diets. Glycerin is basically composed of glycerol (80%) and has no fibrous component that contributes to reducing time spent eating and ruminating. Thus, the change in dietary composition and the lesser need to chew in diets with glycerin resulted in shorter time spent (min d<sup>-1</sup>) on eating and rumination activities, and consequently lower eating and rumination rates in min kg<sup>-1</sup> DM and NDF.

Bite mass is the most relevant variable in the ingestive behavior of grazing animals, and it explains the highest percentage of variation in daily forage intake since bite rate and grazing time play secondary roles (Chacon and Stobbs, 1976). Bite mass increased (P < 0.05) and bite rate decreased (P < 0.05) as glycerin was added to the diet (Table 4); in general, when bite rate decreases, bite mass increases due to longer chewing times (Galli et al., 1996), which make it possible to maintain forage intake rate when there are variations in the pasture.

The number of bites (BITN) and time spent (BITT) per swallowed bolus variables were not affected (P > 0.05) by including glycerin in the diet and means were 39.81 bites and 55.42 s, respectively. The reduction in the number of bites per day (BITD) by including glycerin in the diet was probably caused by reduced grazing time since there was no difference between BITN (39.81) and BITT (55.42), which contributed a greater number of bites per day. The variables of time spent per ruminated bolus and number of chews per bolus and per day were not influenced (P < 0.05) by glycerin inclusion levels (Table 5). The number of boluses per day was quadratically affected (P > 0.05). The number of ruminated boluses per day was quadratically affected (P < 0.05) by including glycerin in the diet with 473 boluses d<sup>-1</sup> at 2.65% crude glycerin. This behavior can be explained by the reduction in dietary fiber contents as glycerin was included because this reduces the need

to form boluses that need to be re-chewed. Once formed, boluses showed the same rumination time (P > 0.05) with a mean of 49.9 s for each one.

The number of chews per bolus and per day did not change (P < 0.05) by including glycerin in the diet and had estimated values of 49.35 and 20.562 chews, respectively. The number of chews per bolus obtained in this research study was similar to the 43 chews observed by Silva et al. (2005), who used supplementation at 0.75% BW in crossbred heifers at pasture. The number of periods and time spent in each period of the behavioral activities are displayed in Table 6. The number of periods grazing, idle, ruminating, and eating at the trough were not affected (P > 0.05) by including glycerin and means were 15.1, 24.9, 13.3, and 3.71 periods per day, respectively. Times for idle and rumination periods were not influenced by glycerin inclusion in the diet (P> 0.05) with mean values of 23.9 and 27.3 min per period. Pereira et al. (2005) supplied different supplements at 0.75% BW and did not report any effect on the duration of rumination periods with a mean of 27.56 min. The mean times per period of grazing and eating at the trough decreased linearly (P < 0.05) as glycerin was included. This behavior was expected since times spent grazing and at the trough decreased as this ingredient was added; this did not change the number of periods which were reflected in the time spent per period.

Ingestion of DM and NDF (g per meal) was not affected (P > 0.05) by including crude glycerin (Table 7) with respective means of 420.6 and 166.7 g per meal, which were expected given that the animals showed no difference in the number of meals (grazing and trough) with a mean of 18.71 meals, or in DM and NDF intake with glycerin inclusion. Feed and rumination efficiency (kg DM and NDF h<sup>-1</sup>) increased (P < 0.05) by including crude glycerin in the diet. The DM intake per hour increased from 0.828 kg in the control treatment to 1.035 kg in the animals fed at the 9.99% inclusion level, which corresponds to raising

Table 4. Mean values of bite rate (BITR), bite mass (BITM), number of bites per swallowed bolus (BITN), time per swallowed bolus (BITT), and number of bites per day (BITD) of heifers fed supplements with increasing glycerin levels.

		Glycerin incl	usion (% DM)			CV		
Item	0.0	3.33	6.66	9.99	Regression equation	(%)	$r^2$	$\mathbf{p}^1$
BITR, bites s-1	0.82	0.73	0.69	0.62	$\hat{Y} = 0.809751 - 0.0192228x$	23.4	0.97	***
BITM, g DM per bite	0.21	0.23	0.32	0.36	$\hat{\mathbf{Y}} = 0.192673 + 0.0165122x$	26.3	0.96	***
BITN, nr	41.63	45.18	34.56	37.87	$\hat{Y} = 39.81$	22.6	-	NS
BITT, s	51.11	59.98	49.78	60.80	$\hat{Y} = 55.42$	9.5	-	NS
BITD, bites d-1	25103	21948	18201	15603	$\hat{Y} = 25056.9 - 967.069x$	20.7	0.99	***

<sup>1</sup>NS: Non significant; <sup>\*</sup>P < 0.1; <sup>\*\*</sup>P < 0.05; <sup>\*\*\*</sup>P < 0.01.

Table 5. Number of ruminated boluses per day, time spent per bolus, number of chews per bolus, and number of chews per day of heifers fed supplements with increasing glycerin levels.

		Glycerin incl	usion (% DM)			CV		
Activity	0.0	3.33	6.66	9.99	Regression equation	(%)	$r^2$	$\mathbf{p}^1$
Boluses per day	461.9	458.9	450.8	350.4	$\hat{\mathbf{Y}} = 457.625 + 11.619x - 2.1930x^2$	17.1	0.96	**
Time per bolus, s	47.3	50.7	49.1	52.6	$\bar{Y} = 49.9$	11.6	-	NS
Chews per bolus	44.5	53.1	49.3	50.5	$\bar{Y} = 49.3$	11.7	-	NS
Chews per day	20 272	22 857	20 354	18 766	$\bar{Y} = 20562$	13.2	-	NS

<sup>1</sup>NS: Non significant; <sup>\*</sup>P < 0.1; <sup>\*\*</sup>P < 0.05; <sup>\*\*\*</sup>P < 0.01.

Table 6. Mean values of the number of grazing (NGP), idle (NIP), rumination (NRP), and trough (NTP) periods, and the duration of grazing (TGP), idle (TIP), rumination (TRP), and trough (TTP) periods of heifers fed supplements with increasing glycerin levels.

		Glycerin inclu	sion (% DM)			CV		
Item	0.0	3.33	6.66	9.99	Regression equation	(%)	$r^2$	$\mathbf{p}^1$
NGP	15.56	15.44	12.67	16.78	$\bar{Y} = 15.1$	16.72	-	NS
NIP	23.06	27.28	22.56	26.94	$\bar{Y} = 24.9$	10.49	-	NS
NRP	12.44	14.67	12.94	13.22	$\bar{Y} = 13.3$	11.35	-	NS
NTP	3.50	4.39	3.78	3.17	$\bar{Y} = 3.71$	24.08	-	NS
TGP, min	35.28	33.74	32.84	28.00	$\hat{\mathbf{Y}} = 35.8868 - 0.684381 \mathbf{x}$	19.23	0.87	***
TIP, min	23.39	19.09	27.23	26.18	$\bar{Y} = 23.9$	13.16	-	NS
TRP, min	29.07	26.34	28.72	25.19	$\bar{Y} = 27.3$	16.44	-	NS
TTP, min	10.25	8.20	7.14	6.51	$\hat{Y} = 9.86680 - 0.368337 x$	18.92	0.94	***

<sup>1</sup>NS: Non significant; <sup>\*</sup>P < 0.1; <sup>\*\*</sup>P < 0.05; <sup>\*\*\*</sup>P < 0.01.

Table 7. Dry matter (DM) and neutral detergent fiber (NDF) intakes, feed and rumination efficiency, and rumination of heifers fed supplements with increasing glycerin levels.

		Glycerin inclu	sion (% DM)			CV		
Item	0.0	3.33	6.66	9.99	Regression equation	(%)	$r^2$	$\mathbf{p}^1$
Intake								
g DM per meal <sup>1</sup>	400.8	359.0	516.8	405.7	$\bar{Y} = 420.6$	18.7	-	NS
g NDF per meal <sup>1</sup>	162.8	138.3	206.4	159.1	$\bar{Y} = 166.7$	20.0	-	NS
Feed efficiency								
kg DM h-1	0.828	0.797	1.091	1.035	$\hat{\mathbf{Y}} = 0.801339 + 0.027.4522x$	11.9	0.65	888
kg NDF h-1	0.336	0.306	0.435	0.405	$\hat{\mathbf{Y}} = 0.320516 + 0.0101176 \mathbf{x}$	12.7	0.53	***
Rumination efficiency								
kg DM h-1	1.236	1.277	1.365	1.599	$\hat{\mathbf{Y}} = 1.19297 + 0.0354097 \mathbf{x}$	16.9	0.97	***
kg NDF h-1	0.489	0.512	0.546	0.627	$\hat{\mathbf{Y}} = 0.476506 + 0.0135016 \mathbf{x}$	18.3	0.91	***
Rumination								
g DM per bolus	16.27	17.73	18.62	23.23	$\hat{\mathbf{Y}} = 15.6964 + 0.654063 \mathbf{x}$	19.7	0.88	***
g NDF per bolus	6.46	6.80	7.47	8.74	$\hat{\mathbf{Y}} = 6.24033 + 0.225283 \mathbf{x}$	20.8	0.93	***

<sup>1</sup>NS: Non significant; <sup>\*</sup>P < 0.1; <sup>\*\*</sup>P < 0.05; <sup>\*\*\*</sup>P < 0.01.

by 27 g each percentage unit of crude glycerin added to the diet. These results are similar to the 1.293 kg DM h<sup>-1</sup> found by Miranda et al. (1999), who worked with dairy heifers that were fed sugarcane-based diets. According to Welch et al. (1982), rumination efficiency is important to control the use of low-digestibility feeds because the animal can ruminate a larger amount of this type of feed during the regular 8 or 9 h of rumination; this means higher feed intake and better productive performance.

Rumination efficiency increased as crude glycerin was added with means of 1.236 and 1.599 kg NDF for each hour spent ruminating, respectively, for animals fed the control diet and the diet with 9.99% inclusion. The possible improvement in rumination efficiency can be related to the reduction in dietary NDF levels, which led the animal to be more efficient in using fiber per unit of time. This result was already expected since, according to Dulphy et al. (1980), rumination efficiency in kg h<sup>-1</sup> can be increased for low-fiber diets because of the greater ability to reduce the size of fibrous material particles. The increase in feed and rumination efficiency (kg DM or NDF h<sup>-1</sup>) by including glycerin in the diet demonstrates that, even though glycerin is devoid of NDF in its composition, this ingredient is effective in maintaining eating and rumination activities when used in the supplementation of grazing animals. This fact is important to reduce variations in the ruminal environment, especially in animals aimed at high production, usually supplemented with large amounts. Thus, 9.99% of glycerin in the diet can be used to improve feed and rumination efficiency of heifers raised

on pasture. The amount of DM per bolus increased (P < 0.05) as crude glycerin was included and increased from 16.27 to 23.23 g at levels of 0% and 9.99%. For both DM and NDF values, this reflects the behavior observed in the number of ruminated boluses (Table 4), which increased as crude glycerin was included since neither DM nor NDF intakes were changed.

#### CONCLUSIONS

Including crude glycerin in diets reduces time spent grazing and eating at the trough, increases idle time, and promotes improved feed and rumination efficiency in grazing heifers. Adding glycerin decreases bite rate and increases mass per bite in heifers supplemented during the dry-rainy season transition.

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