RUMINANT NUTRITION

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# Supplementation strategies for crossbred steers on *Brachiaria brizantha* pasture during the dry season: Effects on performance, intake, digestibility and ingestive behavior

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ABSTRACT. Supplementation strategies have been studied to increase animal performance and the quality of meat. Thus, this study was carried out to evaluate the influence of different supplementation strategies on the performance, feed intake, digestibility and ingestive behaviour of crossbred steers on Brachiaria brizantha pasture, during the dry season. The experiment was performed in Bahia, Brazil, from August 2015 to January 2016, with a dry season of 203 days. Thirty-three crossbred steers with an average weight of 269.5  $\pm$  41.8 kg were distributed in a completely randomised design with three treatments and 11 replicates per treatment. The results were analysed using an ANOVA and a Tukey test, with a 5% probability of error. The treatments were mineral salt with urea and the supplementation of protein based on the animals' body weight (BW), as follows: 0.1 and 0.2% BW. There was no effect of supplementation strategy on the initial and final body weights. Supplementation strategies did not influence (p > 0.05) the variables related to the intake of total dry matter. There was an effect (p < 0.05) on the dry matter and crude protein digestibility coefficients. For ingestive behaviour, feed intake, grazing time and idle time tended to be altered by the supply strategies (p < 0.05), but rumination was not affected (p > 0.05) by the evaluated strategies. In conclusion, the supplementation strategies used during the dry season resulted in a similar performance; therefore, the adopted strategy can be chosen according to the economic and regional conditions of the production system. Thus, the strategy adopted could be chosen according to the economic and regional conditions of the production.

Keywords: crossbred steers; mineral mix; protein supplementation; tropical forage.

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## Introduction

Grass is the main source of feed for beef cattle production systems in Brazil due to the prevailing tropical climatic conditions (Anualpec, 2021). However, climatic condition also a difficulty for grass growth, causing greater accumulation of forage in rainy season and low productivity and poor quality of forage in dry season (Moreira, Prado, Cecato, Wada, & Mizubuti, 2004a; Neves et al., 2018).

During dry seasons, there is a forage decrease, in amount and nutritional value, directly affecting the animal performance raised in pasture system (Moreira et al., 2004a; Silva et al., 2010). Also, cattle could be suffering nutritional constraints that limit pasture intake and digestibility (Detmann, Valente, Batista, & Huhtanen, 2014). Within these limiting factors, crude protein is very important variable, because this nutrient has an essential importance for the adequate microbial growth and degradability fibrous carbohydrates in basal forage diet (Moreira et al., 2004b; Porto et al., 2011; Maeda et al., 2012).

Due to the inverse relation between average daily gain in cattle and pasture productivity during dry season, the additional supplementary strategies appear as an alternative to minimize the effects arising from this period (El-Memari Neto et al., 2003; Silva et al., 2010; Neves et al., 2018; Rocha et al., 2019). This feeding management is common commercial practice in grass-feeding beef cattle systems. However, there are several supplementation techniques that can be implemented and make the production system more attractive.

The objective of this study was to evaluate the animal performance, feed intake, nutrient digestibility, ingestive behavior activities and of crossbred steers subjected to different supplementation strategies during the dry season.

# Material and methods

#### **Ethical considerations**

This study was conducted in strict conformity with the Brazilian legislation on experimentation involving the use of animals, adopted by the National Council of Experimental Control (Concea), and was approved by the Ethics Committee in Animal Use (Ceua) from the State University of Southwest Bahia, located in Itapetinga, Bahia, Brazil, under approval no. 100/2015.

#### Location and animals

The experiment was conducted on the Princesa do Mateiro farm, located in the Ribeirão do Largo, Bahia, northeast Brazil (15° 26' 46" S, 40° 44' 24" W). It lasted 203 days, from August 2015 to January 2016, comprising the dry season in this region (Northeast in Brazil). In this region, the dry season begins in August and ends in January each year.

Thirty-three crossbred steers ( $\frac{1}{2}$  Holstein and  $\frac{1}{2}$  Zebu), with an average body weight (BW) of 269.5 ± 41.8 kg and an average age of 16 months, were distributed across 12 ha of *Brachiaria brizantha* cultivar Marandu. This area was equipped with covered troughs that could be accessed from both sides and water troughs.

The experimental design was completely randomised, with three treatments and eleven replicates. The concentrate supplement was formulated according to the (National Research Council [NRC], 2000), targeting an average daily gain of 600 g animal<sup>-1</sup> day<sup>-1</sup>. The supplements utilised in this phase (Treatments) were composed of mineral salt with urea and supplementation protein based on the animals' BW (Table 1), as follows: 0.1 and 0.2% BW. These strategies were chosen based on previous studies (Silva et al., 2010; Neves et al., 2018; Rocha et al., 2019).

The total area used was 12 hectares subdivided into three areas. In order to avoid the possible effects arising from the pasture, the paddocks were rotated such that they had an occupation period of 7 days for therefore, at the end of the cycle, which corresponded to a complete evaluation period; the steers were transferred back into their starting paddock. In this way, all the animals experienced the same experimental conditions.

## Sampling and chemical analyses

The start of the experiment was preceded by a period of 14 days for the adaptation of the steers to the management and the experimental diets. During the adaptation period, the steers were dewormed with Ivomec<sup>®</sup> long-acting injectable (Ivermectin LA 3.5%, Merial Laboratory, São Paulo, Brazil). The steers were immunologically castrated with the application of Bopriva<sup>®</sup> (Pfizer Animal Health - EUA) according to the manufacturer's recommendations.

The pasture was evaluated at pre-established 28-day intervals, when the entry and exit paddocks for the steers were evaluated. The forage availability was evaluated by the comparative visual yield method proposed by Haydock and Shaw (1975). The weight of the forage mass (natural/green matter) inside the square (0.25  $m^2$ ) was measured using a digital balance, considering three decimal places. After being weighed (total weight), the forage was separated and the morphological components (leaf, stem with sheath and dead material) were weighed, thus giving the individual availability of the components, as well as the leaf:stem ratio for each evaluation period.

Table 1. Ingredients and composition of basal diets (g	kg⁻¹).
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Ingredients (g kg <sup>-1</sup> )	Ground grain sorghum	Soybean meal	Urea	Mineral mix <sup>1</sup>
Mineral salt + urea	-	-	250	750
Supplement 0.1% BW	565.5	193.8	149.3	91.4
Supplement 0.2 % BW	492	313.4	139.4	55.4

<sup>1</sup>Composition (per kilogram): calcium 175 g, phosphorus 60 g, sodium 107 g, sulfur 12 g, magnesium 5 g, cobalt 107 mg, copper 1300 mg, iodine 70 mg, manganese 1000 mg, selenium 18 mg, zinc 4000 mg, iron 1400 mg, fluorine (maximum) 600 mg.

Concomitant to the forage collections, a simulated grazing technique was performed, with the aim of obtaining a representative sample of the height and the quality of the forage intake by the steers. The samples were pre-dried in a forced air circulation oven (55°C) until they reached a constant weight, then ground to 2

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and 1 mm (Willey mill, Thomas<sup>®</sup>), identified in plastic pots, and chemically analysed to identify the percentage of the constituents present in the grazing material.

To estimate the faecal excretion (FE), chromic oxide  $(Cr_2O_3)$  was used as an external marker. The marker was provided daily at 07:00 in a single dose of 10 g animal<sup>-1</sup> day<sup>-1</sup> inside paper cartridges that were administered orally for a period of 11 days; this period consisted of 7 days for the adaptation of the animals to the management procedures and the regulation of chromium excretion in the faeces, and 4 days of sample collection. Subsequently, the faecal output was calculated according to (Smith & Reid, 1955), using the formula below:

FO = OP/CMF

where:

FO = daily faecal output (g day<sup>-1</sup>);

OP = amount of chromic oxide provided (g day<sup>-1</sup>); and

CMF = concentration of chromic oxide in the faeces (g  $g^{-1}$  DM).

The supplement dry matter intake (SDMI) was estimated using a titanium dioxide marker (TiO<sub>2</sub>), which was provided as 15 g animal<sup>-1</sup> day<sup>-1</sup>, mixed with concentrate, for 11 days. It was supplied directly in the trough at 10:00, in accordance with the procedure described by Valadares Filho et al. (2006). Subsequently, SDMI was calculated using the equation below:

 $SDMI = (EF X TIO_2 faeces)/TIO_2 supplement.$ 

where:

 $TiO_2$  faeces and  $TiO_2$  supplement correspond to the concentration of titanium dioxide in the faeces and supplement, respectively.

The estimate of the voluntary intake of roughage was calculated on the basis of the internal indicator indigestible neutral detergent fibre (iNDF), following the methodology proposed by Casali et al. (2008). It was obtained after ruminal incubation for 288 hours, following the methodology and equation described by Detmann et al. (2012).

To determine the performance of the animals, they were weighed at the beginning and end of the rainy season after a 12 hour fast in order to obtain the average body weight, which was used to calculate the total body weight gain and average daily gain (ADG), as well as to adjust the supplementation of the concentrate. The ADG was calculated by the difference between the initial and final body weight divided by the number of days of the experiment. From the DMI per day and ADG, it was possible to calculate the feed conversion of the animals by dividing the DMI by the ADG.

The chemical composition of the forage, concentrate, and supplement were determined at the Laboratório de Misturas e Separações Químicas (Labmesq) at the Uesb, following the methodologies described by Detmann et al. (2012) to determine the dry matter (DM), ash content, ether extract (EE), crude protein (CP), neutral detergent fibre (NDF), and acid detergent fibre (ADF). The chemical composition of forage and concentrate supplements can be seen in Table 2.

Component alarl	Min and Mix	Supplement	level (% BW)
Component <sup>1</sup> , g kg <sup>-1</sup>	Mineral Mix —	0.1	0.2
Dry matter	400.0	895.7	897.7
Crude protein	83.0	465.8	397.6
Organic matter	889.0	894.4	886.6
Ether extract	22.6	32.9	30.9
NDFap	653.5	276.5	336.6
Total carbohydrate	778.0	400.0	458.1
NFCap	129.5	158.4	121.5
Ash	110.0	105.5	113.4
iNDF	250.0	12.2	13.8
ME	1.8	2.2	2.4

Table 2. Composition of supplements and simulated grazing forage samples on a dry matter basis.

<sup>1</sup>NDFap neutral detergent fiber corrected for ash and protein, NFCap non-fibrous carbohydrates corrected for ash and protein, iNDF indigestible neutral detergent fiber, ME metabolizable energy (Mcal kg<sup>-1</sup>).

The total carbohydrate content was estimated following the procedure described to determine the dry matter (DM), ash content, ether extract (EE), crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF). The chemical composition of the forage and concentrate supplements is shown in Table 2.

The total carbohydrate content was estimated following the procedure described by Sniffen, O'Connor, Van Soest, Fox, and Russell (1992). The non-fibrous carbohydrate (NFC) content was determined as the difference between the total carbohydrate and the NDF. The metabolisable energy of the feedstuffs was estimated according to NRC (2000) recommendations. The content of non-fibrous carbohydrates (NFCap) corrected for ash and protein of the forage and faeces was calculated using the equation proposed by Weiss, Pietrzik, Biesalski, Grunert, and Kleinsorge (1999) as follows:

NFCap = 100 - CP - EE - NDFap - MM.

where:

NFCap = non-fibrous carbohydrates corrected for ash and protein;

CP = crude protein content;

EE = ether extract content;

NDFap = neutral detergent fibre corrected for ash and protein; and

MM = mineral matter content. All terms are expressed as % of DM.

The supplements contained urea; for this reason, their NFCap content was obtained using the equation proposed by Hall (2003), as follows:

NFCap = 100 - [(CP% - CP% from urea + urea %) + EE + NDFap + MM].

where:

CP = crude protein content in the concentrate supplement;

CP% from urea = protein equivalent of urea;

urea% = urea content in the concentrate supplement;

EE = ether extract content;

NDFap = neutral detergent fibre corrected for ash and protein; and

MM = mineral matter content. All terms are expressed as % of DM. Faecal samples were processed in the same way as forage, and their chemical composition was determined in the same way as for the supplements.

To measure feeding behaviour, the crossbred steers were subjected to three 24 hour observation periods, at 5 min. intervals, totalling 288 observations of each animal. These observations involved recording the time spent grazing, in rumination, on feed intake and being idle, as proposed by Silva et al. (2006). Observations were performed without interrupting the animal's routine.

#### Statistical analyses

All variables under study were tested for normality and showed a normal distribution. Data were analysed by ANOVA using the general linear model (Statistical Analysis System [SAS], 2004) including diet as a fixed effect. A Tukey's test was used to compare the treatment means; they were considered to be significantly different when p < 0.05.

## Results

In the beginning of the experiment, the highest accumulation of forage was observed, with a sward mass of 4541 kg ha<sup>-1</sup> total dry matter availability, presenting a forage supply of 8.3% BW (Table 3). However, this mass was composed of a more elevated proportion of lignified material (3314 kg ha<sup>-1</sup>) than green forage (819 kg ha<sup>-1</sup>). These forage mass characteristics throughout the experiment had an influence on pasture quality and low animal performance.

 Table 3. Variation of sward components of Marandu grass pasture during the experimental period in the dry season (08/08/2015 to 01/23/2016).

Variables (kg ha <sup>-1</sup> )	8-Aug	12-Sep	17-Oct	21-Nov	26-Dec	23-Jan
Total dry matter	4541	4358	4024	4167	3920	3956
Potentially digestible dry matter	3374	3238	2990	3096	2913	2939
Leaf	819	930	1139	848	853	552
Stem + leaves	2214	1764	2159	1667	1626	2019
Dead material	1100	1229	328	730	712	475

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The initial body weight, final body weight and ADG were similar (p > 0.05) for both supplementation groups (Table 4). Thus, these strategies did not alter the animals' performance. The supplementation strategies did not influence (p > 0.05) the total dry matter, forage dry matter, organic matter and neutral detergent fibre intake. However, the crude protein, ether extract and total digestible nutrient intake were superior (p < 0.05) for the two groups of animals fed with crude protein supplementation (Table 4) compared to the animals supplemented with salt and urea.

The dry matter and crude protein digestibility coefficient were higher (p < 0.05) for the animals fed diets with 0.1 and 0.2% supplementation in comparison with the animals fed the control diet (Table 5). On the other hand, the digestibility coefficients of ethereal extract, neutral detergent fibre corrected for ash and protein and non-fibre carbohydrates corrected for ash and protein were similar (p < 0.05) among the three groups of animals (Table 5).

The time spent in the troughs (min. day<sup>-1</sup>) was higher (p < 0.01) for animals fed a diet with 0.2% supplementation compared to a diet with 0.1% supplementation. However, there was no difference between the control and any supplementation strategy (Table 6). On the other hand, grazing time was different between all groups (p < 0.01), in which the control diet animals spent more time grazing, followed by the animals who received 0.1% supplementation, with the animals receiving 0.2% supplementation grazing the least. In contrast to grazing time, the time spent being idle was lower for the control diet animals, intermediate for 0.1% supplementation animals and higher for the animals who received 0.2% supplementation (p < 0.01; Table 6). Rumination time was not affected (p > 0.05) by the diets.

Itom	Min anal Mix	Supplement Level (% BW)		OTM <sup>*</sup>	n (Malaa
Item	Mineral Mix –	0.1	0.2	— SEM <sup>*</sup>	p < Value
Initial body weight, kg	269.72	268.36	270.91	7.41	0.99
Final body weight, kg	332.64	331.91	341.09	7.58	0.87
Average daily gain, kg day <sup>-1</sup>	0.31	0.31	0.35	0.01	0.44
Feed efficiency, kg gain DM <sup>-1</sup> intake	0.05	0.05	0.05	0.00	0.59
Total dry matter intake, kg day <sup>-1</sup>	5.96	6.58	6.57	0.14	0.13
Forage dry matter intake, kg day <sup>-1</sup>	5.96	6.23	5.79	0.13	0.39
Organic matter intake, kg day <sup>-1</sup>	5.29	5.84	5.84	0.01	0.13
NDF intake, kg day <sup>-1</sup>	3.89	4.17	4.05	0.09	0.45
Crude protein intake, kg day <sup>-1</sup>	0.49 <sup>b</sup>	0.68ª	0.79ª	0.03	0.00
Ether extract intake, kg day-1	0.13 <sup>b</sup>	0.15 <sup>ab</sup>	0.16ª	0.00	0.04
NFCap intake, kg day-2	0.77	0.85	0.84	0.02	0.16
TDN intake, kg day <sup>-3</sup>	2.92 <sup>b</sup>	3.45ª	3.43ª	0.08	0.01

Table 4. Animal performance and feed intake of crossbred steers with different strategies of supplementation.

<sup>ab</sup>Values with different letters in the same row were different by Tukey's test (p < 0,05). "SEM, Standard error of means. !NDF, neutral detergent fiber. 2NFCap, non-fibrous carbohydrates corrected for ash and protein. 3TDN, total digestible nutrients.

<b>Table 5.</b> Total apparent digestibility coefficients (g kg <sup>-1</sup> ) of dry matter and nutrients of crossbred steers with different strategies of
supplementation.

	Minoral Mix	Supplement		1	
Digestibility, g kg <sup>-1</sup>	igestibility, g kg <sup>-1</sup> Mineral Mix -	0.1	0.2	SEM	p-value
Dry matter	482.41 <sup>b</sup>	518.82ª	532.06ª	0.54	0.00
Crude protein	423.10 <sup>b</sup>	474.48 <sup>a</sup>	499.05ª	0.79	0.00
Ether extract	760.00	774.87	782.57	0.61	0.31
$NDFap^1$	551.21	584.62	588.74	0.74	0.07
NFCap <sup>2</sup>	431.02	486.11	463.89	1.14	0.14

<sup>ab</sup>Values with different letters in the same row were different by Tukey's test (p < 0,05). \*SEM, Standard error of means. \*NDFap, neutral detergent fiber corrected for ash and protein. 2NFCap, non-fibrous carbohydrates corrected for ash and protein.

Table 6. Effect of different strategies of supplementation on ingestive behaviour activities of crossbred steers in g	razing	g.

Activities	Mineral Mix -	Supplement I	— SEM <sup>*</sup>		
Activities	Milleral Mix	0.1	0.2	3EIVI	p-value
Feed intake (min. day <sup>-1</sup> )	9.09 <sup>ab</sup>	$7.27^{\mathrm{b}}$	12.58ª	0.69	0.00
Grazing time (min. day <sup>-1</sup> )	558.03ª	546.21 <sup>ab</sup>	516.36 <sup>b</sup>	6.03	0.01
Idleness time (min. day <sup>-1</sup> )	420.76 <sup>b</sup>	450.76 <sup>ab</sup>	472.58ª	7.19	0.01
Rumination time (min. day <sup>-1</sup> )	452.12	435.76	438.48	6.48	0.55

<sup>ab</sup>Values with different letters in the same row were different by Tukey's test (p < 0,05). 'SEM, Standard error of means.

#### Discussion

During the year, tropical forages are affected by variations on climate conditions and thus, the pasture quality during the dry season is compromised by the elevated stem + leaves and residue of dead material. Thus, it is a decrease in the availability of total dry matter and potentially digestible dry matter (Kammes & Allen, 2012).

In this work the mean crude protein content of the pasture was 8.3%; mean value above to the 5.6% (Silva et al., 2009) in the similar condition. Thus, all animals had access to this minimum level of protein (NRC, 2000). Classical studies claim that crude protein total in the diet content above 7.5% provides a microbiota adequate to ruminal fermentation during the dry season in tropical regions (Van Soest, 1994; Hobson & Stewart, 2012).

The hypothesis of this study was that the additional supplementation of crude protein would increase forage intake and, consequently, animal performance, as observed in some studies in the literature (El-Memari Neto et al., 2003; Neves et al., 2018; Rocha et al., 2019). However, the utilisation of supplementary strategies during the dry season did not influence the dry matter intake of the forage. The results showed that there were no associative effects of forage with the supplement. Thus, the use of supplementary strategies as sources of non-structural carbohydrates associated with high nitrogen degradability (urea) foods provided in catalytic doses allowed the animals to better use the forage, without interfering with the total dry matter intake.

The differences in the intake of crude protein, ether extract and total digestible nutrients may be associated with the additional supply of protein sources and sorghum as an energy source via supplementation. In this context, the use of low-quality nitrogen compounds allows for an increase in the rate of degradation and synthesis of the microbial protein, improving energy extraction and increasing the amount of nutrients destined for the intestine and in the production of volatile fatty acids for energy metabolism (Detmann et al., 2004; Sampaio et al., 2010).

Overall, additional protein supplementation did not alter animal performance in terms of final weight, average daily gain or feed efficiency. The average daily gain was low ( $0.33 \pm 0.01$  kg). The level of average daily gain during the dry season can be considered the minimum threshold to avoid animal performance the following summer.

The results observed with the supplementation of protein-based additives for cattle on pasture during the winter are contradictory. Studies conducted in tropical and sub-tropic regions have shown that a low level (below 0.5% of body weight) of protein-based additives has no positive effect on animal performance (Moreira et al., 2004a; Silva et al., 2010). However, when dietary supplementation levels are above 0.6%, there is a slight increase in animal performance (El-Memari Neto et al., 2003; Silva et al., 2010; Neves et al., 2018). Supplementation levels above 0.6% of the live weight may induce an effect of replacing pasture by the supplement added to the diet.

The supply of concentrate supplement (0.1 and 0.2% BW) improved the apparent digestibility coefficients of dry matter and crude protein during the dry season. This result can be attributed to the combination of nutrients with higher digestibility of the concentrate in relation to pasture, which provided an improvement of the nutrient digestibility of pasture due to better rumen conditions (Askar et al., 2013).

The different grazing time values are most likely due to all animals receiving a basal diet in which there were differences in the dietary ingredients, such that the animals that received a greater supply of nutrients or supplementation required less time to conduct this activity. Thus, the animals that received only the mineral mix tended to increase the grazing time to meet their nutritional requirements.

The animals fed with the higher concentrate level  $(0.2\% \text{ BW}^{-1})$  and the mineral mix tended to spend more time on feed intake due to the greater availability of nutrients and the composition of the diet as observe by Mendes et al. (2015) and Souza et al. (2019). On the other hand, grazing time was shorter for animals fed with 0.2% BW<sup>-1</sup> supplementation. This means more concentrate intake time and less grazing time. Likewise, animals that ingested a higher level of concentrate spent more time idle. However, there was no difference in rumination time. The highest level of concentrate supplementation (0.2% BW<sup>-1</sup>) can be considered low and has no effect on rumination.

## Conclusion

The results of this experiment suggest that low supplementation (0.1 or 0.2% of live weight) can be adopted during the dry period in northeastern Brazil (Bahia) without significantly altering animal behavior and

promoting satisfactory weight gains in the production system. of beef cattle used in the region (low earnings). Thus, the strategy adopted could be chosen according to the economic and regional conditions of the production (low animal performance).

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