Are the effects of dietary lipid content in grazing beef cattle independent of the amount of concentrate supplement?

Os efeitos do teor dietético de lipídeo sobre as características nutricionais e de desempenho são independentes da quantidade de suplemento concentrado?

Matheus Leonardi Damasceno¹*; Eriton Egidio Lisboa Valente²; Mariana Barbizan¹; Sidnei Antônio Lopes³; Silvana Teixeira Carvalho²

Highlights _____

Dietary lipid content affects nutrient intake in beef cattle.

There is no information on about dietary lipid content and supplementation level.

Dietary lipid content and supplementation level have independent effects.

Abstract __

This study aimed to evaluate the effects of levels of concentrate supplement and dietary lipids on the nutritional characteristics, metabolic parameters, and performance of Nellore bulls grazing tropical pasture. Twenty-seven Nellore bulls were allotted to 2 × 2 completely randomized factorial design with two levels of supplementation and two levels of dietary lipids. The concentrate supplement was fed at 4 g kg⁻¹ BW (low supplementation) and 8 g kg⁻¹ BW (high supplementation), whereas the dietary lipid content was 28 g of ether extract (EE) g⁻¹ on a dry matter basis (DM) (low-lipid diet) and 42 g EE kg⁻¹ DM (high-lipid diet). There was no interaction between the level of concentrate supplement and dietary lipid content on all studied variables. High levels of supplementation led to shorter grazing time and reduced forage dry matter intake (DMF). On the other hand, the DM intake increased with increasing levels of concentrate supplement, with no significant effects on DM digestibility. The dietary lipid content did not affect forage and DM intakes. However, the digestibility of DM and neutral detergent fiber (NDF) were lowest in high-lipid diets. Bulls receiving high levels of supplementation had higher average daily gains than low-supplemented animals.

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¹ Students of the Doctoral Course of the Postgraduate Program in Animal Science, Department of Animal Science, Western Paraná State University, UNIOESTE, Marechal Cândido Rondon, PR, Brazil. E-mail: matheusld31@gmail.com; maribarbizan94@gmail.com

² Researchers, Department of Animal Science, Western Paraná State University, UNIOESTE, Marechal Cândido Rondon, PR, Brazil. E-mail: eritonvalente@yahoo.com.br; silteixeira@gmail.com

³ Researchers, Department of Animal Science, National Institute of Science and Technology in Animal Science, INCT, CA, Viçosa, MG, Brasil. E-mail: sidneyufv@hotmail.com

^{*} Author for correspondence

Neither the amount of concentrate supplement nor the dietary lipid content affected the hot carcass yield and carcass weight. Carcass fat deposition was greater in bulls fed high-lipid diets than in animals receiving low-lipid diets. The effects of the level of concentrate supplement and dietary lipid content on the nutritional characteristics and performance of grazing beef cattle are independent. Concentrate supplementation at 8 g kg⁻¹ BW increases cattle performance but decreases forage utilization. Diets containing 42 g EE kg⁻¹ DM reduces the digestibility of dry matter and fiber fractions, but increases backfat thickness. **Key words:** Grain. Pasture. Whole soybean. Supplementation.

Resumo __

Este estudo teve como objetivo avaliar o efeito da guantidade de suplemento concentrado e da concentração de lipídios na dieta, sobre as características nutricionais e metabólicas e o desempenho de touros Nelore em pastagem tropical. Vinte e sete touros Nelore foram distribuídos em esquema fatorial 2 x 2 com dois níveis de suplemento e dois níveis de lipídios alimentares). A quantidade de suplemento concentrado foi de 4 g kg⁻¹ de PC (baixo suplemento) e 8 g kg⁻¹ de PC (alto suplemento), enquanto os teores de lipídios dietéticos foram de 28 g de extrato etéreo (EE) g-1 de matéria seca (MS) (dieta com baixo lipídios) e 42 g de EE kg⁻¹ de MS (dieta com alto lipídios). Não houve interação entre a quantidade de suplemento concentrado e o teor dietético de lipídios para todas as variáveis analisadas. Os altos níveis de suplemento reduziram o tempo de pastejo e consumo de matéria seca de forragem (MSF). Por outro lado, o consumo MS aumentou com as maiores quantidades de suplemento concentrado, sem qualquer efeito sobre a digestibilidade da MS. O teor de lipídios não afetou o consumo de forragem e MS. No entanto, a digestibilidade da MS e da fibra detergente neutro (FDN) foram mais baixas para as dietas com elevado teor de lipídios. Os touros que receberam níveis elevados de suplementos obtiveram maior ganho médio diário. Nem a quantidade de suplemento concentrado, nem o teor dietético de lipídeo foram influenciaram no rendimento e no peso da carcaça quente. Os touros que receberam alto teor dietético de lipídio, apresentaram maior deposição de gordura na carcaça. Os efeitos do nível de suplemento concentrado e do teor de lipídios da dieta sobre as características nutricionais e desempenho de bovinos de corte em pastejo são independentes. A oferta de 8 g kg⁻¹ de PC de suplemento de concentrado aumenta o desempenho dos bovinos, mas diminui a utilização de forragens. O conteúdo lipídico de 42 g de EE kg⁻¹ de MS reduz a digestibilidade da matéria seca e fibras, mas aumenta a deposição de gordura subcutânea.

Palavras-chave: Grãos. Pastagem. Soja. Suplementação.

Introduction ____

Beef cattle cannot express their full genetic potential on tropical pastures. Therefore, supplements are an essential strategy for enhancing animal performance and increasing backfat deposition in the carcasses (Poppi, Quigley, Silva, & McLennan, 2018). However, using grains as a source of starch or lipids may decrease forage utilization due to the negative associative effects between pasture and concentrate supplements (Aldrich, Merchen, & Drackley, 1995; M. A. Souza et al., 2010).

Cattle performance can be improved if lipid supplementation increases energy intake (Rosa et al., 2013). However, supplementation with excessively high levels of lipids has been associated with reduced dry matter and energy intakes and low body weight gain in cattle



(Jordan et al., 2006). Although lipids have been extensity studied in feedlot trials, there is not enough data from experiments with grazing cattle. The extrapolation of information from feedlot to grazing production systems can be biased due to differences between diets. primarily because of the fiber content and synchrony of nutrient supply. Both the amount of concentrate supplement and dietary lipid content can affect rumen microorganisms (Carvalho et al., 2011; Kairenius et al., 2018), with adverse effects on ruminal digestion. High-lipid diets are associated with decreased DM intake (Fiorentini et al., 2012) and reduced fiber digestibility in the rumen (Hall, Goetsch, Landis, Forster, & Brake, 1990). The DM intake is not usually reduced in feedlot cattle fed diets with EE levels up to 5-6% (Felton & Kerley, 2004; Messana et al., 2014). Although there are few studies evaluating the dietary lipid content for grazing cattle, low lipid levels (3-4%) usually do not change the DM and forage intake (Almeida et al., 2018; Brokaw, Hess, & Rule, 2001).

Total mixed rations are widely used in feedlots due to their high degree of nutrient synchrony in the rumen. However, shortterm changes in nutrient synchrony occur in grazing cattle receiving concentrate supplementation. Although rumen microbiota is inherently adaptable to asynchronous nutrient supply (Reynolds & Kristensen, 2008), diet modifications may also change the microbial population due to an increased supply of non-fiber carbohydrates (Carvalho et al., 2011). Therefore, short-term changes in the ruminal environment due to the high amount of starch and fat after supplement intake may differ from synchronous diets (feedlot) in terms of nutritional characteristics. Although dietary fat is not significantly detrimental to the digestibility of non-fiber

carbohydrates (Jenkins & Bridges, 2007), there is no information on the interaction between lipid content and concentrate levels in asynchronous diets. However, changes in the rumen microbial population of grazing cattle caused by increased grain inclusion potentially affect cattle's response to dietary lipid levels. Therefore, we hypothesize that there is a relationship between the amount of concentrate supplement and dietary lipid levels on intake, digestibility, and performance of grazing beef cattle.

This study aimed to evaluate the effects of levels of concentrate supplement and dietary lipids on the nutritional characteristics, metabolic parameters, and performance of Nellore bulls grazing tropical pasture.

Material and Methods _____

Animals, diet, and experimental design

This study was approved by the Ethics Committee on Animal Use (CEUA/ Universidade Estadual do Oeste do Paraná - under the process nº 27/16), following the ethical principles of animal experimentation established by the Brazilian Council of Animal Experimentation Control (CONCEA).

The study was carried out in the municipality of Entre Rios do Oeste, Paraná, Brazil (24°40'34" S, 54°16'38" W) from January to April 2017. Twenty-seven Nellore bulls averaging 22 months and 439.3 \pm 27.8 kg of initial body weight (BW) were randomly assigned to four treatments (three treatments with seven replicates each and one treatment with six replicates). The bulls were allowed 10 days of adaptation to diets and facilities, and data were collected for 84 days.

A 2 × 2 completely randomized factorial design with four treatments (two levels of supplementation and two levels of dietary lipids) was used. The concentrate supplement was fed at 4 g kg⁻¹ BW (low supplementation) and 8 g kg⁻¹ BW (high supplementation),

whereas the dietary lipid content was 28 g of ether extract (EE) g^{-1} on a dry matter basis (DM) (low-lipid diet) and 42 g EE k g^{-1} DM (high-lipid diet) (Table 1). The mineral supplement was offered ad libitum.

Table 1Ingredients and chemical composition of the diet

	Forago	Low supplementation ¹		High supplementation ¹	
	Forage	Low-lipid	High-lipid	Low-lipid	High-lipid
Ingredient (g kg ⁻¹ DM)					
Corn		330	200	685	620
Soybean meal		350	0	275	100
Raw whole soybeans		320	800	40	280
Chemical composition (g kg ⁻¹ DM)					
Organic matter	913	950	950	970	970
Crude protein	101	330	330	210	220
Ether extract	21	80	160	40	80
Non-fiber carbohydrates	150	438	416	584	573
Neutral detergent fiber	642	190	200	170	180

¹Low supplementation = 4 g of supplement kg⁻¹ BW; High supplementation = 8 g of supplement kg⁻¹ BW; Low-lipid diet = 28 g EE kg⁻¹ DM; High-lipid diet = 42 g EE kg⁻¹ DM.

Each group was kept on 2-ha paddocks planted with *Urochloa brizantha* cv. Xaraés grass and managed under continuous stocking. The bulls were supplemented daily at 11 a.m. and were rotated among paddocks every seven days to minimize the possible effects of the paddock on experimental treatments (Table 1).

Measurements and sampling

The forage mass was evaluated every 28d by cutting three samples of 0.25 $m^2 \mbox{ at } 1$

cm above the ground level in each paddock. The forage accumulation rate was evaluated using three 1-m² exclusion cages randomly placed in each paddock. Forage accumulation was calculated by subtracting the forage mass (kg DM ha⁻¹) at the beginning of the evaluation period from the forage mass inside the cages after 28 d. The forage accumulation rate (kg DM ha⁻¹ d⁻¹) was calculated by dividing the forage accumulation by the period of evaluation (28 d). Forage samples were collected using the hand-plucking method at the beginning of the experiment and every 14 d to estimate the chemical composition of ingested forage. The grazing behavior of bulls was evaluated by visual observation during twelve consecutive hours (7 a.m. to 7 p.m.) on days 17 and 18, 52 and 53, 73 and 74. Trained observers were positioned at strategic locations outside the paddocks (at 10 m of distance) without interfering with the normal behavior of bulls. The time spent grazing was recorded.

From the 33rd and 40th day of the experimental period, a digestibility trial was carried out to evaluate the voluntary intake, nutrient digestibility, and fecal excretion of whole soybeans. The fecal collection was carried out at 4:30 p.m. on day 38, at 11 a.m. on day 39, and at 6 a.m. on day 40. Fecal samples (200g) were taken immediately after spontaneous defecation, stored in plastic bags, and frozen (-20°C) for further analysis. A total of 130 g of feces from each sampling day (3 d) was collected to evaluate the relationship between soybean intake and excretion. The samples were washed through a 4-mm mesh sieve to estimate the fecal excretion of whole soybeans. The fecal output (kg) was estimated using titanium dioxide (TiO₂) as an external marker (Myers, Ludden, Nayigihugu, & Hess, 2004). A total of 15 g d⁻¹ of titanium dioxide was wrapped in paper cartridges and introduced (11 a.m.) into the esophagus of each animal by a flexible rubber tube. The fecal output was determined by dividing the amount of titanium dioxide supplied daily (kg) by the marker concentration in the feces (kg kg⁻¹).

The individual supplement intake (kg d⁻¹) was estimated using chromium oxide (Cr_2O_3) as an external marker (Williams, David, & lismaa, 1962). A total of 15 g animal⁻¹ d⁻¹ was mixed with the concentrate supplement. The individual intake of the supplement was estimated according to the following equation:

$$SI = \frac{FO \times MCF}{MCS}$$

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Where: SI is the supplement intake (kg d⁻¹), FO is the fecal output (kg d⁻¹), MCF is the marker concentration in the feces (kg kg⁻¹), and MCS is the marker concentration in the supplement (kg kg⁻¹).

The dry matter intake was calculated by the following equation:

$$DMI = \frac{\left[(FO \times iNDF_{feces}) - iNDF_{supplement}\right]}{iNDF_{forage}} + SI$$

Where DMI is the dry matter intake (kg d⁻¹), FO is the fecal output (kg d⁻¹), iNDF_{feces} is the neutral detergent fiber (iNDF) concentration in the feces (kg kg⁻¹), iNDF_{supplement} is the iNDF concentration in the supplement (kg), iNDF_{forage} is the iNDF concentration in the forage (kg kg⁻¹), and SI is the supplement intake.

Urine, ruminal fluid, and blood samples were collected 4 hours before and 4 hours after supplementation from half of the animals per day (days 42 and 43). Urine samples were collected after spontaneous urination, diluted in H_2SO_4 (0.036N) at a ratio of 1: 4 (urine: H_2SO_4) and frozen at -20°C. Subsequently, urine samples were analyzed for concentrations of creatinine, nitrogen, allantoin, and uric acid. Aliquots of 25 mL of ruminal fluid were sampled via esophageal tubing with the aid of a vacuum pump, and the pH was immediately measured. A total of 0.5 mL of H_2SO_4 (50%) was added to the sampled material, and the mixture was frozen at -20° C. Subsequently, the rumen ammonia nitrogen concentration was determined. Blood samples were collected by jugular venipuncture into vacuum tubes and centrifuged at 3,000 g for 15 minutes to obtain the serum, which was frozen at -20° C. Then, the urea nitrogen concentration was determined.

Animal performance was evaluated by weighing the bulls at the beginning and end of the experiment after 14 hours of fasting from solids. The bulls were weighed unfasted (8 a.m.) at the beginning of the experiment and every 28 days to adjust the amount of concentrate supplement to be supplied to each group.

Chemical analysis

Samples forage, feces, of and supplement ingredients were oven-dried at 55°C for 72 hours and ground to pass a 1-mm screen, except for indigestible neutral detergent fiber (iNDF) determinations, in which samples were ground to 2-mm. Samples were analyzed for DM (method no. 920.39), crude protein (CP) (method no. 954.01), organic matter (OM) (method no. 942.05), and ether extract (EE) (method no. 920.39) as described by Association of Official Analytical Chemists [AOAC] (1990). Samples were treated with thermostable α -amylase without sodium sulfite (Mertens, 2002) to analyze the neutral detergent fiber (NDF) content.

The iNDF content in feed and fecal samples was evaluated using F57 filter bags (Ankom, Macedon, NY, USA) incubated in the rumen for 288 hours (Valente et al., 2011). Fecal samples were analyzed for chromium and titanium dioxide concentrations using atomic absorption and colorimetric methods, respectively. The methodologies used to determine the concentration of external and internal markers have been described above. The total digestible nutrients (TDN) and nonfibrous carbohydrates (NFC) were calculated according to Sniffen, O'Connor, Van Soest, Fox and Russell (1992). The whole soybeans were collected manually, weighed, and analyzed for DM content as previously described (Rennó et al., 2015). The total soybean excretion in feces was calculated as the concentration of grains in feces multiplied by the daily fecal output.

The urinary nitrogen compounds were analyzed by the Kjeldahl method as previously described. Urinary creatinine and uric acid concentrations were analyzed using commercially available test kits (Analisa® Belo Horizonte, MG, BR). Urinary allantoin concentration was analyzed using highperformance liquid chromatography (George et al., 2006). Rumen fluid was analyzed for rumen ammonia nitrogen (RAN) concentration using the colorimetric method (Chaney & Marbach, 1962) by replacing phenol with sodium salicylate (Felix & Cardoso, 2004). Urea concentration was analyzed in serum samples using a commercial kit (Gold Analisa, Belo Horizonte, MG, Brazil).

Urine volume and purine derivatives

The daily urine volume was calculated by the relationship between the daily creatinine excretion and its concentration in spot samples (Silva et al., 2012), using the shrunk body weight estimated according to Valadares et al. (2016):

$$CE = 0.0345 \times SBW^{0.9491}$$

Where: CE is the creatinine excretion (g/d), and SBW is the shrunk body weight (kg).

The total excretion of purine derivatives was calculated as the sum of allantoin and uric acid excreted in the urine. Absorbed purines were calculated from the excretion of purine derivatives, according to Barbosa et al. (2011):

$$AP = \frac{PD - 0.301 \text{ x BW}^{0.75}}{0.80}$$

Where: AP is the absorbed purines (mmol d^{-1}); PD is the excretion of the purine derivatives

(mmol d⁻¹); 0.301 is the endogenous excretion of purine derivatives in the urine (mmol) per unit metabolic weight (BW0.75), and 0.80 is the recovery of absorbed purine as purine derivatives in urine (mmol mmol⁻¹).

The synthesis of microbial nitrogen compounds in the rumen was calculated as a function of AP according to the equation of (Barbosa et al., 2011):

$$Nmic = \frac{70 \text{ x AP}}{0.93 \text{ x R x 1000}}$$

Where: Nmic is the intestinal flow of microbial nitrogen compounds (g d⁻¹); 70 is the N content in purines (mg of N mol⁻¹); 0.93 is the digestibility of microbial purines; R is the purine-N: total N in bacteria (0.134).

Carcass quality

The backfat thickness was evaluated at the beginning and at the end of the experimental period by ultrasound. The fat deposition was calculated by subtracting the final backfat thickness from the initial backfat thickness. Ultrasound images were collected using a Pie Medical-Aquila device (Esaote Europe B.V.) with a linear transducer of 18 cm and 3.5 MHz frequency. The animals were scanned across the 13th thoracic vertebrae, and the images were analyzed with the SketchUp Pro 2016 software.

On the day after the end of the experimental period, the animals were fasted from solids for 8 hours, weighed to obtain the final BW, and transported to the slaughterhouse. The slaughter procedures followed the Brazilian inspection standards, and the animals were stunned by concussion, bled by cutting the jugular vein, skinned and eviscerated. The carcasses were then identified, washed, and weighed to obtain the hot carcass weight (HCW). The hot carcass yield (%) was calculated as the ratio between HCW and final BW.

Statistical analysis

The experiment was conducted as a 2×2 completely randomized factorial design with four treatments (two levels of supplementation and two levels of dietary lipids), according to the model:

$$Yij = \mu + Si + Li + (S \times L)ij + eij$$

where Yij is the dependent variable; μ = the overall mean; Si is the fixed effect of supplementation level; Lj is the fixed effect of lipid level; (SxL)ij is the interaction between the main effects; and eij is the random error associated with Yij, distributed as eijk~N (0, σe^2).

Means were compared by orthogonal contrasts. The initial body weight was used as a covariate in the analyses of final body weight. Data were analyzed using the MIXED procedure of SAS (SAS University Edition) with α = 0.10 for reducing type II error.

Results and Discussion _____

Forage allowance is critical to ruminants and interferes with the effects of supplementation on cattle performance. The minimum recommended forage allowance is dependent on forage species; however, the forage allowance in this study was considerably high (Oliveira et al., 2016), thereby ensuring that the effects of supplementation were not limited by lack of forage. The mean forage mass was 6,536 kg DM ha⁻¹, while the stocking rate was 1,696 kg BW ha⁻¹. The forage allowance was 3.85 kg DM kg BW⁻¹, and the mean forage accumulation rate was 117 kg DM ha⁻¹ d⁻¹.

There was no interaction between the level of concentrate supplement and lipid content on the studied variables (Table 2). Bulls receiving high levels of supplementation spent less time grazing than those receiving low levels of concentrate supplement (Table 3). Grazing activities decreased with increasing supplementintake as a result of the substitution effect (Mendes et al., 2015). Although the DM intake increased with increasing levels of concentrate supplementation, the forage intake and the time spent grazing decreased in high-supplemented bulls. The reduced time spent grazing may be associated with lower physical activity levels and, consequently, the energy expenditure is reduced (Valente et al., 2019).

Table 2

Probabilities for fixed effects of level of concentrate supplement, dietary lipid content and their interactions on intake, digestibility, ruminal parameters, and performance of Nellore bulls grazing tropical pastures

Variable1	<i>P</i> -value ²						
	S	L	S × L				
Intake of nutrients							
Grazing time	0.009	0.944	0.153				
Dry matter of forage	<0.001	0.980	0.937				
Dry matter of supplement	0.074	0.960	0.246				
Dry matter	0.039	0.961	0.255				
Organic matter	0.240	0.835	0.687				
Crude protein	0.573	<0.001	0.795				
Ether extract	0.125	0.962	0.171				
Neutral detergent fiber	<0.001	0.172	0.445				
Non-fiber carbohydrates	0.031	0.232	0.327				
Digestible organic matter	0.039	0.415	0.357				
Total digestible nutrients	<0.001	<0.001	0.714				
Intake of whole soybeans	<0.001	0.005	0.759				
Fecal excretion of whole soybeans	0.014	0.745	0.922				
	Digestibility of nutrients						
Dry matter	0.150	0.013	0.554				
Organic matter	0.124	0.002	0.734				
Crude protein	0.296	0.757	0.213				
Ether extract	0.634	0.010	0.712				
Neutral detergent fiber	0.648	0.008	0.140				
Non-fiber carbohydrates	0.185	0.108	0.127				

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	Ruminal parameters					
pH _m	0.016	0.005	0.458			
pH _a	0.020	0.037	0.928			
Rumen ammonia nitrogen	0.001	0.174	0.325			
Urinary nitrogen excretion	0.002	0.091	0.716			
Serum urea nitrogen	0.079	0.866	0.922			
Microbial nitrogen	0.558	0.874	0.232			
Performance and carcass traits						
Average daily gain	0.065	0.706	0.957			
Body weight gain	0.065	0.706	0.957			
Final weight	0.065	0.706	0.957			
Hot carcass weight	0.162	0.507	0.469			
Carcass yield	0.880	0.259	0.714			
BFTi	0.342	0.712	0.376			
BFTf	0.126	0.029	0.505			
FD	0.997	0.028	0.958			

¹pHm = pH in the morning; pHa = pH in the afternoon; BFTi = initial backfat thickness; BFTf = final backfat thickness; FD = fat deposition

 2 S = effect of level of concentrate supplementation; L = effect of dietary lipid content; S × L = interaction between levels of concentrate supplement and dietary lipid content.

Table 3

Diurnal grazing time (h), intake (kg d⁻¹), and fecal excretion of whole soybeans (kg d-1) in bulls fed different levels of concentrate supplement and lipids in the diets

Variable ¹	Low supplementation ²		High supplementation ²		Mean
	Low-lipid	High-lipid	Low-lipid	High-lipid	Iviean
Grazing time	4.03±0.14	4.07±0.10	3.66±0.15	3.77±0.10	3.87±0.07
Dry matter of forage	7.99±0.47	8.53±0.28	7.44±0.32	6.82±0.48	7.69±0.22
Dry matter of supplement	1.97±0.27	1.93±0.20	3.96±0.33	3.98±0.32	3.00±0.23
Dry matter	9.96±0.60	10.46±0.35	11.40±0.50	10.80±0.40	10.69±0.25
Organic matter	9.17±0.61	9.63±0.30	10.64±0.52	10.09±0.35	9.91±0.23
Crude protein	1.45±0.12	1.49±0.06	1.58±0.08	1.56±0.05	1.53±0.04
Ether extract	0.28±0.02	0.44±0.02	0.27±0.01	0.42±0.02	0.36±0.02
Neutral detergent fiber	5.50±0.33	5.86±0.18	5.45±0.24	5.09±0.27	5.48±0.13
Non-fiber carbohydrates	1.93±0.14	1.82±0.05	3.33±0.21	3.00±0.13	2.55±0.14
Digestible organic matter	6.36±0.47	6.25±0.23	7.56±0.43	6.73±0.30	13.03±0.20
Total digestible nutrients	6.56±0.45	6.60±0.30	7.74±0.49	7.06±0.25	6.99±0.20
Intake of whole soybeans	0.50±0.04	1.54±0.10	0.15±0.01	1.11±0.09	0.87±0.10
Fecal excretion of whole soybeans	0.02±0.18	0.03±0.10	0.01±0.01	0.01±0.06	0.02±0.03

¹Values expressed as mean ± standard error

²Low supplementation = 4 g of supplement kg⁻¹ BW; High supplementation = 8 g of supplement kg⁻¹ BW; Low-lipid diet = 28 g EE kg⁻¹ DM; High-lipid diet = 42 g EE kg⁻¹ DM.

High levels of supplementation (8 g kg⁻¹ BW) decreased forage intake, but increased NFC intake (Table 3). Decreases in forage intake with starch supplementation are frequently observed in grazing beef cattle (Valente et al., 2014) and are associated with changes in the rumen microbial ecosystem, which impairs the rumen dynamics of NDF (Souza et al., 2010). However, the highest NFC intake did not alter the dietary NDF intake. Other authors (Figueiras et al., 2010; Lazzarini et al., 2016) reported that this effect is commonly observed and that NFC intake has a lesser adverse effect on NDF intake in cattle grazing medium-quality forage than in animals on low-quality forage. Thus, the lack of effect of supplementation on NDF intake in our study may indicate that physical regulation also played an important role in feed intake (Mertens, 2002).

On the one hand, the CP intake was similar between bulls fed low and high levels of concentrate supplement. On the other hand, the DM, OM, NFC, DOM (digestible organic matter), and TDN intakes increased with increasing levels of concentrate supplement. The dietary lipid content did not affect the intakes of forage, DM, OM, CP, NDF, NFC, DOM and TDN. In forage-concentrate diets, there are complex interactions between dietary components that may result in different associative effects. The decrease in forage intake was not proportional to the increase in supplement intake. Therefore, although forage intake decreased with increasing supplementation, the increase in the amount of supplement improved the total DM and energy intake (TDN).

The lack of interaction between supplementation level and dietary lipid content on feed intake indicates that the effect of lipids on rumen metabolism is not necessarily dependent on the predominance of amylolytic or fibrolytic microbial populations. The adverse effects of lipid supplementation on rumen metabolism are mainly related to impaired fiber utilization because dietary lipids are less harmful to NFC digestibility than to fiber digestibility (Jenkins, 1993).

The fecal excretion of whole soybeans was higher in bulls receiving low levels of concentrate supplement (Table 3) because this group consumed more soybeans to adjust the dietary lipid content. However, fecal soybean excretion was low (lower than 5% of grain intake) despite this difference between treatments. Although the increase of lipid content also increased the excretion of whole soybeans in feces, the intakes of total DM, forage, NDF, and TDN were not affected by the dietary lipid content. Most of the dietary fat escape from the rumen to the lower gastrointestinal tract (Jenkin & Bridges, 2007). However, the natural protection of the whole soybean grain can hinder the access of enzymes to lipids. Although there was an increase in the fecal excretion of soybean grains with increasing soybean inclusion, the amount excreted was not significant compared to the intake (6 to 44 g kg⁻¹ grain). Low fecal excretion of whole soybeans (50 g kg⁻¹ grain) was also reported in feedlots even at high inclusion levels (250 g kg⁻¹ DM) (Rennó et al., 2015), which indicates that cattle have high efficiency in degrading whole soybeans.

High levels of concentrate supplementation did not change the digestibility of DM, CP, EE, NDF, and NFC, but improved OM digestibility. However, high lipid contents reduced the digestibility of DM, NDF, and OM, but increased EE digestibility (Table 4).

The inclusion of concentrate in diets for cattle grazing tropical pastures is usually associated with competition between

microorganisms for substrates and decreased fiber digestibility (Lazzarini et al., 2016). Nevertheless, the amount of concentrate supplement did not decrease fiber digestibility (Table 4). The adverse effects of starch on NDF digestibility is dependent on CP intake (Souza et al., 2010). In all treatments, the dietary CP content was similar and sufficient to meet both the microbial and animal requirements (Valadares et al., 2016). Therefore, microbial competition for substrate utilization and their deleterious effects on fiber degradation were probably reduced by an adequate protein supply.

Table 4

Diurnal grazing time (h), intake (kg d⁻¹), and fecal excretion of whole soybeans (kg d-1) in bulls fed different levels of concentrate supplement and lipids in the diets

Variable ¹	Low supplementation ²		High supplementation ²		Mean
	Low-lipid	High-lipid	Low-lipid	High-lipid	IVICALI
Dry matter	64.67±1.41	62.28±0.74	67.40±0.43	63.31±1.71	61.15±0.63
Organic matter	69.05±0.59	65.72±1.15	71.57±0.74	67.06±1.48	68.38±0.63
Crude protein	72.62±0.64	73.56±0.74	73.90±1.01	73.35±1.08	73.35±0.43
Ether extract	50.00±9.20	61.96±1.81	54.88±3.23	61.44±2.87	57.07±2.38
Neutral detergent fiber	62.43±0.82	59.99±1.57	66.13±0.85	57.98±2.69	61.63±0.98
Non-fiber carbohydrates	82.30±4.61	78.68±1.71	82.33±1.81	79.16±1.49	80.61±1.38

¹Values expressed as mean ± standard error

²Low supplementation = 4 g of supplement kg⁻¹ BW; High supplementation = 8 g of supplement kg⁻¹ BW; Low-lipid diet = 28 g EE kg⁻¹ DM; High-lipid diet = 42 g EE kg⁻¹ DM.

Although the maximum dietary EE content was 42 g kg⁻¹ DM, which is not enough to affect the nutritional characteristics in feedlot studies (Messana et al., 2014), the NDF and OM digestibilities were lower in high-lipid than in low-lipid diets. The high proportion of forage in diets helped to reduce the digestibility of OM and NDF with increasing lipid levels because forage-based diets are more susceptible to disrupt rumen fermentation with lipid inclusion (Jenkins, 1993). Other studies have also observed this same effect (Carvalho et al., 2016, 2017). The EE digestibility increased with increasing lipid levels due to the lower relative contribution of metabolic fecal metabolic fraction to EE excretion.

Bullsreceivinghighlevelsofconcentrate supplement had lower concentrations of rumen ammonia nitrogen (RAN), serum urea nitrogen (SUN) and urinary nitrogen excretion (UNE) than low-supplemented animals. The dietary lipid content did not affect the RAN, SUN, and UNE concentrations (Table 5). An optimum protein: starch ratio in supplements improves nitrogen assimilation and minimizes N excretion (Souza et al., 2010). Although CP intake was not affected by the concentrate supplement or lipid content, the NFC intake increased in bulls receiving a high supplementation level compared with lowsupplemented animals. Thus, the increase of NFC intake increased the nitrogen assimilation

by rumen microorganisms and, consequently, reduced urea levels in the blood and the urinary nitrogen excretion.

Neither the amount of concentrate supplement nor the dietary lipid content affected the N_{mic} production and efficiency of microbial protein synthesis (Table 4). Although dietary lipids cannot supply energy to rumen microorganisms (Jenkins, 1993) and have potentially adverse effects on microbial growth, the dietary lipid content was not sufficiently high to modify the Nmic production and efficiency of microbial protein synthesis. Bulls receiving high levels of concentrate supplement had lower ruminal pH after supplementation than low-supplemented animals. Moreover, bulls fed high-lipid diets had lower ruminal pH before supplementation than animals fed low-lipid diets. Rumen pH was lower after supplementation due to the increase of NFC fermentation. The higher rumen pH in bulls fed high-lipid diets may be attributed to an indirect effect of lipid inclusion. The increase of lipid levels can modify the ruminal environment, thereby resulting in greater variability in pH (Messana et al., 2013; Santana et al., 2017). However, this variation was not observed in our study. According to Demeyer and Van Nevel (1995), protected lipid sources such as soybean grains are less harmful to ruminal microorganisms. Therefore, no reduction in rumen pH was observed.

Table 5

Variable ¹	Low supplementation ²		High supplementation ²		Mean
	Low-lipid	High-lipid	Low-lipid	High-lipid	wear
pHm	7.12±0.40	6.89±0.08	7.48±0.11	7.08±0.09	7.14±0.05
рНа	6.96±0.16	7.21±0.13	6.69±0.03	6.92±0.08	6.94±0.05
RAN (mg dL ⁻¹)	12.46±0.56	10.80±0.89	8.27±0.45	8.02±1.01	9.88±0.49
UNE (mg dL ⁻¹)	102.56±10.73	112.56±5.89	75.14±4.93	90.62±6.79	95.22±4.21
SUN (mg dL ⁻¹)	16.72±0.73	16.57±0.78	14.68±1.02	14.94±1.19	15.72±0.50
Microbial nitrogen (g d-1)	117.67±5.45	125.43±4.55	123.01±10.57	110.82±6.70	119.23±2.82

Ruminal parameters in bulls fed different levels of concentrate supplement and lipids in the diet

¹Values expressed as mean ± standard error; pHm = pH in the morning; pHa = pH in the afternoon; RAN = rumen ammonia nitrogen; UNE = urinary nitrogen excretion; SUN: serum urea nitrogen

²Low supplementation = 4 g of supplement kg⁻¹ BW; High supplementation = 8 g of supplement kg⁻¹ BW; Low-lipid diet = 28 g EE kg⁻¹ DM; High-lipid diet = 42 g EE kg⁻¹ DM.

Bulls receiving high levels of supplement had higher average daily gain (ADG), BW gain, and final BW (Table 6) than low-supplemented animals. This improved performance is explained by the supplementation per se, which increased energy intake. Many authors have reported similar results in beef cattle on tropical forages receiving protein supplementation (Valente, Paulino, Detmann, Valadares, & Lopes, 2013; Detmann, Valente, Batista, & Huhtanen, 2014; Poppi et al., 2018). The lack of effect of lipid content on animal performance was due to the low dietary lipid content. Although the whole soybean was added to supplements at high levels, the proportion of forage in the total diet was larger. Therefore, none of the treatments reached considerably high levels of total lipids due to the low lipid content of the forage. On the one hand, bulls fed high-lipid diets (80 to 100g of EE kg⁻¹ DM) have lower DM intake and performance (Felton & Kerley, 2004; Jordan et al., 2006). On the other hand, some feedlot studies have shown that cattle performance is not affected by moderate lipid levels (40 to 60 g of EE⁻¹ kg), even though some authors report a decrease in DM intake (Bassi et al., 2012; Messana et al., 2014). Neither the amount of concentrate supplement nor the dietary lipid content was high enough to influence the HCW and carcass yield (%) (Table 6). However, bulls fed high lipid-diets had greater backfat deposition. The improved performance of bulls receiving high levels of concentrate supplement was not enough to influence the final weight and carcass yield. Many studies (Pompei et al., 2018; Menezes et al., 2019) observed this same effect, and the main explanation involves the use of contemporary animals of the same breed and diets with similar energy content.

Table 6

Variable ¹ -	Low supplementation ²		High supplementation ²		Mean
	Low-lipid	High-lipid	Low-lipid	High-lipid	IVIEdIT
Average daily gain (kg d ⁻¹)	0.95±0.06	0.97±0.03	1.05±0.05	1.08±0.05	1.01±0.02
Body weight gain (kg)	79.8±5.8	81.7±2.9	88.6±4.9	90.7±4.8	85.2±2.3
Final weight (kg)	517±11	523±17	527±11	531±13	525±6
Hot carcass weight (kg)	282.1±7.2	284.3±10.4	285.7±6.9	291.9±8.6	286.0±4.1
Hot carcass yield (%)	52.71±0.45	53.11±0.22	52.47±0.63	53.21±0.48	52.87±0.23
BFTi (mm)	2.80±0.10	3.15±0.28	3.31±0.17	3.18±0.33	3.11±0.43
BFTf (mm)	3.65±0.40	4.64±0.31	4.34±0.16	4.86±0.32	4.37±0.43
FD (mm)	0.85±0.49	1.60±0.15	0.86±0.30	1.60±0.20	1.22±0.37

Performance of bulls fed different levels of concentrate supplement and lipids in the diet

¹Values expressed as mean ± standard error; BFTi = initial backfat thickness; BFTf = final backfat thickness; FD = fat deposition

²Low supplementation = 4 g of supplement kg⁻¹ BW; High supplementation = 8 g of supplement kg⁻¹ BW; Low-lipid diet = 28 g EE kg⁻¹ DM; High-lipid diet = 42 g EE kg⁻¹ DM.

All treatments stimulated backfat deposition in the carcasses. However, the backfat deposition was greater in bulls fed high-lipid diets than in animals receiving lowlipid diets. Lipid sources with some degree of ruminal protection can stimulate the passage of unsaturated fatty acids through the rumen, thereby increasing intestinal absorption (Doreau & Ferlay, 1994; Lima et al., 2017). Consequently, the metabolic efficiency in adipose tissue anabolism reactions increases (Souza, Medeiros, Morais, Oshiro & Torres, 2009), which results in greater fat deposition.

Conclusions _____

The effects of the level of concentrate supplement and dietary lipid content on the nutritional characteristics and performance of grazing beef cattle are independent. Concentrate supplementation at 8 g kg⁻¹ BW increases cattle performance but decreases forage utilization. Although diets containing 42 g EE kg⁻¹ DM reduced the digestibility of dry matter and fiber fractions, backfat deposition increased without affecting feed intake and animal performance. Supplementation of lipids above 42 g kg⁻¹ DM should be carried out with prudence, regardless of the amount of concentrate supplement to grazing cattle.

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