SEMINA
Ciências Agrárias

DOI: 10.5433/1679-0359.2025v46n2p459

Replacing corn and soybean meal with a combination of wheat bran and urea on performance of grazing suckling beef calves

Substituição de grão de milho moído e farelo de soja por uma combinação de farelo de trigo e ureia sobre o desempenho de bezerros de corte lactentes em pastejo

Ághata Elins Moreira da Silva[1]; Deilen Paff Sotelo Moreno[2]*; Camila de Paula[3]; Naycker Patricia Verissimo dos Santos[4]; Danilo Henrique Souza da Silva[5]; Gutierrez José de Freitas Assis[6]; Arturo Macias Franco[7]; Edenio Detmann[8]; Luciana Navajas Rennó[8]; Mário Fonseca Paulino[8]

Highlights

Wheat bran and urea were fed to suckling beef calves in a protein-energy supplement.
Calves fed wheat bran and urea had equal performance as other supplemented calves.
Replacing corn and soybean meal with wheat bran and urea for calves is recommended.

Abstract

The objective of this study was to evaluate the effects of replacing corn and soybean meal with a combination of wheat bran and urea in a protein-energy supplement on productive, nutritional, and metabolic characteristics of grazing suckling beef calves. Fifty-two Nellore calves (87±4.95 days of age; 111.3±5.98 kg of initial body weight [BW]) were allocated in a completely randomized design with 4 treatments and 13 replicates. The 140-d trial evaluated treatments consisting of progressive

[1] Dr., Postdoctoral Fellow, Department of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, AB, Canada. E-mail: aghatael@ualberta.ca
[2] Prof. Dr., Department of Animal Production, Faculty of Agricultural and Animal Sciences and Natural Resources, University of the Llanos, UNILLANOS, Villavicencio, MT, Colombia. E-mail: desotelo@unillanos.edu.co
[3] Doctoral Student of the Graduate Program in Animal Science, Universidade Federal de Viçosa, UFV, Department of Animal Science, Viçosa, MG, Brazil. E-mail: camila.p.paula@ufv.br
[4] Veterinary, Department of Animal Science, UFV, Viçosa, MG, Brazil. E-mail: naycker@outlook.com
[5] Agronomist, Department of Agronomy, UFV, Viçosa, MG, Brazil. E-mail: danilo.henrique@ufv.br
[6] Dr., Instituto Tecnológico de Agropecuária de Pitangui, ITAP, Pitangui, MG, Brazil. E-mail: gutierrez.assis@epamig.br
[7] Dr., Postdoctoral Fellow, Department of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, AB, Canada. E-mail: maciasfr@ualberta.ca
[8] Profs. Drs., Department of Animal Science, UFV, Viçosa, MG, Brazil. E-mail: lucianarenno@ufv.br; detmann@ufv.br; mariofonsecapaulino@gmail.com
* Author for correspondence

Received: Oct. 03, 2024 Approved: Feb. 26, 2025

replacing corn and soybean meal with wheat bran and urea (0, 50, and 100%) and a control group receiving only mineral mixture. Supplements were formulated to contain 20% of crude protein (CP), provided in the amount of 7 g kg^{-1} BW. Dry matter, organic matter, CP, total digestible nutrients and digested organic matter (DOM) intake were higher for animals receiving supplementation than those without supplementation (P<0.01). Among the animals that receiving protein-energy supplementation, the non-fiber carbohydrates (NFC) intake displayed a linear decreasing effect, while for the CP-to-DOM ratio a linear increasing effect was observed (P=0.006). The NFC digestibility was higher for the animals that received supplement (P=0.035), and among these animals a linear decreasing response for DOM digestibility was observed (P=0.046). Additionally, the neutral detergent fiber digestibility of these animals showed a quadratic response (P=0.023). Overall, the supplemented animals exhibiting a higher BW at weaning (P=0.047). However, no differences were observed in the performance among the supplemented calves (P>0.05). Concentrations of insulin-like growth factor-1 and serum urea nitrogen were higher for animals receiving supplementation (P<0.041). In conclusion, replacing corn and soybean meal with a combination of wheat bran and urea as a source of protein-energy supplement at 7 g kg^{-1} BW is recommended for suckling beef calves on tropical pastures.
Key words: Alternative feed. *Bos indicus.* Creep feeding. IGF-1.

Resumo

O objetivo deste estudo foi avaliar os efeitos da substituição do milho e farelo de soja por uma combinação de farelo de trigo e ureia no suplemento nas características produtivas, nutricionais e metabólicas de bezerros de corte lactentes em pastejo. Cinquenta e dois bezerros Nelore com idade e peso corporal (PC) inicial de 87±4,95 dias e 111,3±5,98 kg, respectivamente, foram distribuídos em um delineamento inteiramente casualizado com quatro tratamentos e treze repetições. O experimento durou 140 dias e os tratamentos testados consistiram na substituição de milho e farelo de soja por farelo de trigo e ureia (0, 50 e 100%) e um grupo controle que recebeu apenas mistura mineral. Os suplementos foram formulados para conter 20% de proteína bruta (PB), fornecidos na quantidade de 7 g kg^{-1} de PC. A ingestão de matéria seca, matéria orgânica, PB, nutrientes digestíveis totais e matéria orgânica digestível (MOD) foi maior para os animais recebendo suplementação em comparação aos não suplementados (P<0,01). Entre os animais recebendo suplementação proteica-energética, a ingestão de carboidratos não fibrosos (CNF) mostrou um efeito linear decrescente, enquanto que para a relação PB:MOD foi observado um efeito linear crescente (P<0,006). A digestibilidade de CNF foi maior para os animais suplementados (P=0,035), e entre esses animais uma resposta linear decrescente para a digestibilidade da MOD (P=0,046) foi observada. Além disso, a digestibilidade da fibra em detergente neutro desses animais apresentou uma resposta quadrática (P=0,023). No geral, os animais suplementados apresentaram maior PC ao desmame (P=0,047). No entanto, não foram observadas diferenças no desempenho entre os bezerros suplementados (P>0,05). As concentrações de IGF-1 (fator de crescimento insulínico 1) e nitrogênio ureico sérico foram maiores para os animais suplementados (P<0,041). Em conclusão, recomenda-se a substituição do milho e farelo de soja por farelo de trigo e ureia como fonte de suplemento proteico-energético na quantidade de 7 g kg^{-1} de PC para bezerros de corte em pastagens tropicais.
Palavras-chave: Alimento alternativo. *Bos indicus.* creep-feeding. IGF-1.

Introduction

Supplementation of grass-fed calves generally increases nutrient availability, resulting in an elevated weaning weight and a shorter production period. Data gathered from 11 grass-fed calf trials evaluating a control (mineral mixture only) against three to four treatments showed that supplemented calves in a creep-feeding system had a higher weaning weight than calves that received only the mineral mixture (Lopes et al., 2016; Moreno et al., 2022).

Thus, offering protein-energy supplements in the creep-feeding system for suckling calves under grazing conditions is a strategy used to increase nutrient input and, maximize growth performance 200 g d^{-1} more (Carvalho et al., 2019), and can lead to adequacy of indicators of anabolic statuses, such as insulin-like growth factor-1 (IGF-1), as well as protein (urea, total proteins, and albumin) and energetic (insulin, glucose, cholesterol, and triglycerides) under tropical conditions (Ortega et al., 2020; Moreno et al., 2022).

However, the supplementation of calves in a creep-feeding system can also dramatically increase production costs. Therefore, cost-reduction strategies must be implemented to maintain favorable cost-benefit ratios in supplementation programs. Wheat bran could be used as an effective feed with good nutritional characteristics and a lower cost, compared to the expensive commodities used to formulate commercial and non-commercial supplements for calves. Thus, wheat bran can be an affordable alternative to lower supplementation cost. Wheat bran's protein exhibits high ruminal degradability (Goes et al., 2004; Pan et al., 2021) and it serves as a good source of phosphorus and other minerals, with a lower starch content than to grains (Humer & Zebeli, 2017). These benefits are critical for animals that receive mainly low-quality forage because a high starch content reduces digestibility and forage intake (Franco et al., 2017; Pan et al., 2021). Despite its benefits, wheat bran has a lower protein concentration that common commodities, and may need to be combined with high-protein supplements.

Urea is commonly used in the diet of ruminants as a source of non-protein nitrogen, but its usage in young animals' diets is questioned because of its toxicity and its requirement for an active ruminal function (Signoretti et al., 2011). Calves trend to transition from partial rumen utilization to almost-full rumen function at around 3-4 months of age (Porto et al., 2009). Therefore, using urea as an alternative protein supplementation for older calves could be a viable and economically feasible practice in cow-calf operations.

Studies have consistently shown that supplementation of suckling beef calves fed in a creep-feeding system have higher daily gain rates resulting in better performance and greater body weight (BW) at weaning (Marquez et al., 2014; Cardenas et al., 2015; Lima et al., 2016; Lopes et al., 2016; Moreno et al., 2022). However, few studies have been carried out on replacing corn and soybean meal with wheat bran and urea in supplements for suckling beef calves grazed on pastures, evaluating the effect on productive and nutritional performance and metabolic status associated with animal anabolism. Hence, this study could bring new

evidences which may influence the biological response of beef calves and provide proper development in the preweaning period under grazing in tropical conditions.

This study was conducted to evaluate the effects of replacing commonly used commodities (corn and soybean meal) with wheat bran and urea in a protein-energy supplement for suckling beef calves reared on tropical pasture. We assessed the supplement's effect on the calves' performance, nutritional intake, and metabolic characteristics.

Material and Methods

This study was approved by the Brazilian Ethics Committee on Animal Use (CEUAP/UFV – process no. 19/2018), according to ethical principles of animal experimentation established by the National Council of Animal Experimentation Control (CONCEA).

Location and weather conditions

The experiment was conducted at the Beef Cattle Farm of the Animal Science Department of the Universidade Federal de Viçosa (UFV), Viçosa-MG, Brazil (20°45' S, 42°52' W), between January to June 2017, corresponding to the transition from the rainy season to the dry season (140 experimental days). The experimental area was located in a mountainous region at an altitude of 670 m. The average rainfall during the experiment was 58.1 mm (66.6, 84.0, 81.4, 44.8, 51.4, and 18.6 mm), with an average temperature of 20.7 °C (23.3, 22.7, 21.8, 20.5, 18.2, and 17.7 °C), and a medium relative humidity of 80.3% (75.8, 79.1, 79.8, 81.4, 83.5, and 82.2%) for January, February, March, April, May, and June, respectively (Universidade Federal de Viçosa [UFV], 2017).

Animal management, experimental design, and treatments

Fifty-two Nellore calves with an initial age and body weight of 87±4.95 days and 111.3±5.98 kg, respectively, fed by their respective mothers with average body weight of 496.6±18.75 kg, were used in this experiment. After a 15-day adaptation period to the area and experimental conditions, the calves were randomly allocated into eight 7.5-ha paddocks each (two paddocks for each treatment), evenly covered with Brachiaria decumbens Stapf, and equipped with drinkers and feeders.

The experimental design was completely randomized and involved a progressive replacing corn and soybean meal with wheat bran and urea at levels of 0%, 50%, and 100%, along with a control group receiving only a mineral mix (Table 1). All supplements were formulated to contain 20% crude protein (CP; Table 1), and provided in the amount of 7 g kg^{-1} of BW. All the animals had free access to the mineral mix and water throughout the trial.

Table 1
Ingredient and chemical composition of supplements and *Brachiaria decumbes* used in the diets of suckling beef calves

Item[1]	Supplements[2]			*B. decumbens*[3]	*B. decumbens*[4]
	0%	50%	100%		
Ingredients (%; as-fed basis)					
Mineral mix[5]	Ad libitum			-	-
Ground corn	70	35	-	-	--
Soybean meal	30	15	-	-	-
Wheat bran	-	49	98	-	-
Urea/A.S. (9:1)	-	1	2	-	-
Molasses	3	3	3	-	-
Chemical Composition (g kg^{-1} on dry matter basis)					
Dry matter	893.5	894.0	894.5	348.1±1.23	308.5±1.33
Organic matter	970.9	961.7	952.5	926.1±0.16	921.9±0.14
Crude protein	202.0	202.4	202.8	60.9±0.25	66.7±0.21
Ether extract	41.3	67.7	94.2	13.8±0.12	15.8±0.08
NFC	640.6	502.5	364.3	240.2±0.56	228.6±0.53
apNDF	86.9	204.9	323.0	611.0±0.71	610.7±0.70
iNDF	18.5	60.1	101.7	213.3±0.64	227.5±0.21
Milk yield and composition [6]					
	$Milk_{4\%}$[7]	Protein[8]	Fat[8]	Lactose [8]	Total solids[8]
Mean	6.6±0.48	35.2±0.13	47.2±0.29	44.5±0.06	138.5±0.45

[1] A.S.: Ammonium sulfate; NFC: non fiber carbohydrate; apNDF: neutral detergent fiber corrected for ash and protein residue; iNDF: indigestible neutral detergent fiber.
[2] Replacement levels of corn and soybean meal for wheat bran and urea.
[3] Samples collected by hand plucked method during the digestibility trial (mean ± standard error of the mean).
[4] Samples collected by hand plucked method during the experimental period (mean ± standard error of the mean).
[5] Dicalcium phosphate (500 g kg^{-1}), sodium chloride (472 g kg^{-1}), zinc sulfate (15 g kg^{-1}), copper sulfate (7g g kg^{-1}), manganese sulfate (5 g kg^{-1}), cobalt sulfate (0.5 g kg^{-1}), sodium selenite (0.06 g kg^{-1}), potassium iodate (0.5 g kg^{-1}).
[6] Mean ± standard error of the mean.
[7] $Milk_{4\%}$: Milk yield corrected for 4% of fat (kg day^{-1}).
[8] g kg^{-1}.

The supplementation level of 7 g kg^{-1} BW corresponded to approximately 51% and 34% of the dietary requirements of CP and energy, respectively, for beef calves Zebu (5 to 6 mo of age) under grazing conditions with BW of 200 kg and expected gain of 0.8 kg day^{-1} (Valadares et al., 2016).

Supplements were provided daily at 11h00 to minimize any potential interference with the animals' grazing behavior and they were fed in a collective feeder in each paddock, closely mimicking the handling practices typical in beef-production systems, due to gregarious behaviour of the cattle, especially in pastoral environments

that influences feeding patterns. As the evaluations were focused on individual performance and these measurements were collected individually (voluntary intake, total digestibility, nitrogen levels, growth performance, and metabolic profile), the animal was considered the experimental unit (thirteen replicates by treatment), as recommended by Detmann et al. (2016).

In order to minimize potential effects of the paddocks on experimental treatments, animals were rotated across the eight pastures every 7 days with each group staying on each plot the same length of time.

Sample collections and processing

Representative samples of supplements were collected monthly for chemical analysis. Pasture samples were collected every 14 days to assess chemical composition using the hand-plucking method. This approach aimed to closely simulate the grazing behavior of the calves by identifying the specific grazing sites and plant parts selected by the animals (Wallis de Vries, 1995). A second pasture sample was collected every 14 days to estimate the forage's potentially digestible dry matter (pdDM), consisting of five random biomass subsamples collected in each paddock by cutting approximately 1 cm above the ground using a metal square (0.5 m × 0.5 m). Samples were then then prepared by drying in a forced air circulation oven and partially dried at 60 °C for 72 hours, followed by grinding in a knife Willye mill (model 3; Arthur G. Thomas, Philadelphia, PA, USA) to pass through a 2 mm screen. After that, half of each ground sample was ground again to pass through a 1 mm screen.

To evaluate the intake and digestibility of nutrients in all calves, a ten-day digestibility trial was conducted from the 70th experimental day. Individual supplement intake was estimated using titanium dioxide (TiO_2), which was homogenized in the supplement in a plastic bag at a rate of 10 g kg^{-1} of supplement for 9 days (Detmann et al., 2016). Chromium oxide (Cr_2O_3) was utilized as an external marker to estimate fecal excretion with dosage of 10 g per animal (Detmann et al., 2016). The Cr_2O_3 was packed in paper cartridges and delivered via the esophagus with a metal probe, once daily at 11h00 over 9 days. Additionally, indigestible neutral detergent fiber (iNDF) served as an internal marker to estimate forage dry matter (DM) intake (Detmann et al., 2016). The first six days were used for the stabilization of the excretion of the markers, and fecal samples were collected immediately after defecation or directly from the rectum of the animals in amounts of approximately 200 g at 6h00, 10h00, 15h00, and 18h00 on days 6, 7, 8, and 9 of the digestion trial, respectively. All the fecal samples were identified, partially oven-dried at 60 °C for 72 hours, and ground as described for forage samples. Grounded samples were pooled over the sampling time points by calves and stored in plastic pots before analysis.

Samples of supplements, forage, and feces ground to 2 mm were analyzed for iNDF (after 288 h of ruminal in situ incubation; INCT-CA F-009/1). Samples ground to 1 mm were analyzed according to the procedures suggested by Detmann et al. (2012), for DM (dried overnight at 105 °C; method INCT-CA G-003/1), CP (Kjeldahl procedure; method INCT-CA N-001/1), ash (complete combustion in a muffle furnace at 600 °C for

4 h; method INCT-CA M-001/1), ether extract (EE- Randall procedure; method INCT-CA G-005/1), and neutral detergent fiber corrected for ash and protein (apNDF: using a heat-stable α-amylase, omitting sodium sulfite, and correcting for residual ash and protein; methods INCT-CA F-002/1; INCT-CA M-002/1; INCT-CA N-004/1). Additionally, fecal samples were also analyzed for Cr_2O_3 concentration using nitroperchloric digestion and atomic absorption spectrophotometry (GBC Avanta Σ, Scientific Equipment, Braeside, Victoria, Australia; method INCT-CA M-005/1) and TiO_2 (method INCT-CA M-007/1) according to Detmann et al. (2012). The fecal DM excretion was estimated using the Cr_2O_3 marker, based on the ratio between the amount of chromium supplied and its concentration in the feces, as recommended by Detmann et al. (2016):

$$\text{Fecal DM (kg/day)} = \text{AII/ICF}$$

Where: AII = amount of indicator ingested (g) and ICF = concentration of indicator in fecal DM (g kg^{-1} of fecal DM).

Individual supplement intake (ISI) was estimated by the ratio of excretion of TiO_2 in feces and marker concentration in the supplement, as recommended by Detmann et al. (2016):

$$\text{ISI (kg/day)} = [(\text{FE} \times \text{ICaF})/\text{IOG}] \times \text{SOG}$$

Where: FE = fecal DM excretion (kg day^{-1}); ICaF = indicator concentration in animal feces (kg kg^{-1}); IOG = indicator present in the supplement offered to each group (kg day^{-1}); and SOG = supplement amount offered to the group of animals or treatment (kg day^{-1}).

Individual DM intake (DMI) was estimated by using iNDF as an internal marker and calculated by the following equation described by Detmann et al. (2016):

$$\text{DMI (kg day}^{-1}) = [(\text{FE} \times \text{iNDF} - \text{iNDFS})/\text{iNDFP}] + \text{ISI}$$

Where: iNDFF = concentration of iNDF in the feces (kg kg^{-1}); iNDFS = concentration of iNDF in the supplement (kg kg^{-1}); and iNDFP = concentration of iNDF in the pasture (kg).

Digested organic matter (DOM) was calculated according to Detmann et al. (2016):

$$\text{DOM (kg)} = (\text{IOMS} + \text{IFOM}) \times \text{DOOM}$$

Where: IOMS = intake organic matter from the supplement; IFOM = intake forage organic matter; and DOOM = digestibility of organic matter.

On the 80th day, urine spot samples were collected four hours before and four hours after the supplementation to evaluate the microbial protein production. Urine samples were collected during spontaneous urination, proportionally sampled, diluted in 40 mL H_2SO_4 (0.036 N), and frozen at -20 °C for before analysis of creatinine (K067) and uric acid (K0139, Bioclin® Quibasa, Belo Horizonte, Brazil) concentrations. Allantoin levels were analyzed according to the colorimetric method described by Chen and Gomes (1992).

Productive performance and carcass characteristics

For productive performance evaluation, all calves were weighed at 06h00 at the beginning and end of the 140-day experimental phase after a 14-hour fast from solids and milk, allowing only water intake. Additionally, every 30 days, all calves were

weighed to monitor performance and animal welfare.

Carcass characteristics were evaluated by ultrasound (Aloka SSD 500; 3.5-MHz linear probe; Aloka Co. Ltd., Wallingford, CT, USA). Carcass images were obtained between the 12th and 13th ribs over the *longissimus* muscle to measure the ribeye area and backfat-thickness on *longissimus* muscle (SFTL), and between the ischium and pubis to measure the backfat-thickness at the rump (SFTR). Vegetable oil was used to ensure adequate acoustic contact. Images were analyzed in the BioSoft Toolbox II for Beef software (Biotronics Inc., Ames, IA, USA).

On day 85 of the experiment, milk samples were collected to estimate the cows' milk yield and composition. Milking procedures were made as described by Almeida et al. (2018), which included a controlled suckling period prior to calf separation. In order to deplete the milk produced by cows, calves were separated from dams at 15h00. The cows were then returned to the paddock, while the calves remained in the cattle shed. At 17h30, the calves were reunited with their dams and allowed to suckle for 30 min. At 18h00, calves were once again separated from mothers until the next morning. On the next day, at 06h00, cows were then milked mechanically immediately after an injection of 2 mL of oxytocin (10 IU/mL; Ocitovet®, Vet&Cia Animal Health, São Paulo, Brazil) in the mammary artery, and the produced milk was weighed immediately after milking. The exact time when the milking of each cow ended was recorded, and the milk yield was converted to a 24-h production.

Individual samples of 50 mL of milk were taken for analyses of protein, fat, lactose, and total solids. Samples were stored at 4 °C in a refrigerator using a bronopol tablet per sample as a preservative. Milk samples were analyzed using spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark). Milk production was corrected to 4% of fat ($Milk_{4\%}$) according to the National Research Council [NRC] (1996):

$$Milk_{4\%}\ (kg) = 0.4 \times (milk\ production) + [15 \times (fat\ production \times milk\ production/100)]$$

Blood samples and measurements

On days 69 and 139, blood was collected to quantify the serum concentration of urea nitrogen, total proteins, albumin, and triglycerides, as well as the plasma concentration of glucose. On day 139, a blood sample was also collected to quantify the serum insulin-like growth factor-1 (IGF-1) concentration. All samples were collected at 07h00 via jugular venipuncture in vacuum tubes with clot activator and gel for serum separation (BD Vacutainer SST II Advance, São Paulo, Brazil) and vacuum tubes containing sodium fluoride and EDTA (BD Vacutainer Fluoreto/EDTA, São Paulo, Brazil) as glycolytic inhibitor and anticoagulant, respectively, for plasma preparation. Immediately after collection, samples were centrifuged (3,600 × g for 15 min) and serum and plasma were frozen at -20 °C for later analysis.

Creatinine (K067) and uric acid (K0139) concentrations were measured in urine, while serum samples were used to determine urea (K056), total protein (K031), albumin (K040), and triglyceride (K117) levels. Plasma samples were analyzed for glucose concentrations (K082). All analyses were

performed using Bioclin® kits (Belo Horizonte, Brazil) on an automated biochemical analyzer (Mindray BS200E, Shenzhen, China). Serum urea nitrogen (SUN) was calculated as 46.67% of the total serum urea. Insulin-like growth factor 1 (IGF-1) levels were quantified using the chemiluminescence method with the IGF1-Somatomedin C kit (117) on an Immulite 2000 XPI system (Siemens, Eschborn, Germany).

Statistical analysis

Response variables were analyzed using the MIXED procedure in SAS 9.4 (Statistical Analysis System, Inc., Cary, NC, USA). Comparisons between the means of the treatments were carried out using orthogonal contrasts constructed in order to evaluate the effects of supplementation (control vs. supplemented), and linear and quadratic order in function the level of replacing corn and soybean meal with wheat bran and urea. For the variables that did not present a supplementation effect but the effect of the level of supplementation was significant, a Dunnett's test was performed to identify whether a supplemented treatment differed from the control.

The effect of treatment on all variables measured was evaluated by ANOVA, adopting the initial BW as the covariate when significant, according to the following mathematical model:

$$Y_{ijk} = \mu + T_i + e_{(i)j} + \varepsilon_{(ij)k}$$

Where, Y_{ijk} = observations of individual k on paddock j under treatment i; μ = overall mean; T_i = fixed effect of treatment; $e_{(i)j}$ = random error, unobservable, associate to each j paddock under treatment i, assumed to be normally and independently distributed (NID; 0, σe^2); and $\varepsilon_{(ij)k}$: random error, unobservable, associate to each k observation on j paddock under treatment i, assumed to be NID (0, σe^2).

The blood concentrations, with exception of IGF-1, were analyzed as time-repeated measures where day of collection was considered the repeated variable. The choice of the most appropriate covariance structure was based on the lowest value of the corrected Akaike information criterion. The degrees of freedom were estimated according to the Kenward–Roger method. The data showed normality by the Shapiro–Wilk test and homoscedasticity through the Bartlett test. Differences were considered significant at $P \leq 0.05$.

Results and Discussion

The average availability of DM and pdDM was 3.9- and 2.8-ton ha^{-1}, respectively, representimg a 71.4% potential of available forage mass utilization. The forage's CP mean content was 66.7 g kg^{-1} DM (Table 1), lower than the critical limit of 7% of CP (Lazzarini et al., 2009). Thus, the consumption of only forage could lead to a lower contribution of CP reaching the rumen, limiting microbial growth and hapmpering the digestion of fibrous carbohydrates.

Overall, animals receiving supplementation showed a significantly higher intake of DM, organic matter, CP, total digestible nutrients (TDN), non-fibrous carbohydrates (NFC), and digested organic matter (DOM) than those without supplementation ($P<0.01$; Table 2). This increased intake can be attributed to the enhanced nutrient availability from the

supplements. On the other hand, the similar forage DM intake across all groups (P=0.555; Table 2) suggests that supplementation did not replace forage consumption, but rather provided additional nutrients to the diet of the supplemented animals.

The diet's protein-to-energy ratio can be a powerful tool for understanding the metabolic effects of protein on intake and is a reliable indicator of diet adequacy to the animal's requirements (Detmann et al., 2014). According to Detmann et al. (2014), the maximum forage intake is observed when the CP-to-DOM ratio is approximately 288 g kg^{-1}. In this regard, our results showed a higher CP-to-DOM ratio in supplemented calves than calves in the control group (263 *vs* 249 g CP kg^{-1} DOM; Table 2). Thus, supplemented calves exhibited values closer to those recommended to favor a higher forage intake and digestion, indicating a better equilibrium of protein-to-energy ratio. Nonetheless, this metabolic adequacy was not enough to promote a higher intake of forage DM, differing from the findings of Silva et al. (2017), Ortega et al. (2020), and Moreno et al. (2022).

Table 2
Voluntary intake of suckling beef calves fed with protein-energy supplements in tropical pasture

Item[1]	Treatments[2]				SEM[3]	*P*-value[4]		
	Control	0%	50%	100%		C vs S	L	Q
				(kg day^{-1})				
Dry matter	2.34	3.32	3.13	3.20	0.167	0.010	0.652	0.551
FDM	1.32	1.24	1.13	1.24	0.157	0.555	0.983	0.593
SDM	-	1.15	1.14	1.08	-	-	-	-
MDM	1.027	0.935	0.857	0.871	0.0694	0.156	0.555	0.615
Organic matter	2.16	3.12	2.93	2.99	0.156	0.009	0.598	0.547
Crude protein	0.342	0.545	0.511	0.522	0.0207	0.001	0.482	0.431
Ether extract	0.372	0.403	0.371	0.418	0.0305	0.512	0.734	0.353
TDN	1.78	2.69	2.30	2.40	0.121	0.008	0.169	0.185
apNDF	0.79	0.85	0.94	1.09	0.089	0.180	0.126	0.810
NFC	0.650	1.321	1.120	0.990	0.0443	<0.001	0.006	0.547
DOM	1.38	2.24	1.89	1.90	0.092	0.004	0.063	0.179
iNDF	0.288	0.263	0.328	0.368	0.0281	0.384	0.057	0.783
DNDF	0.381	0.429	0.371	0.493	0.0524	0.453	0.437	0.233
CP:DOM (g kg^{-1})	249	243	271	276	11.9	0.060	0.006	0.110

[1]FFM: forage dry matter; SDM: supplement dry matter; MDM: milk dry matter; TDN: total digestible nutrients; apNDF: neutral detergent fiber corrected for ash and protein residue; NFC: non fibrous carbohydrate; DOM: digested organic matter; iNDF: indigestible neutral detergent fiber; DNDF: digested neutral detergent fiber.
[2]Control: Mineral mix only; 0%. 50%. 100%: Replacement levels of corn and soybean meal for wheat bran and urea.
[3]Means standard error of the mean.
[4]Contrast between animals fed with mineral mix only or protein-energy supplement (C vs S) and linear (L) and quadratic (Q) effects for the levels of substitution.

Among animals that received supplements, a linear decrease in protein-to-energy ratio was observed when replacing corn and soybean meal for wheat bran and urea for NFC (P=0.006; Table 2). By contrast, a linear increase in the CP-to-DOM ratio was observed with a higher amount of wheat bran and urea in the supplement (P=0.006; Table 2). The decrease in NFC in the supplemented groups can be attributed to the composition of the diet (Table 1), because wheat bran contains a higher fiber content and lower amounts of NFC than to corn and soybean meal.

Total nutrient digestibility, except for NFC digestibility (P=0.035; Table 3), was not influenced by the supplements. This finding may be due to the lower NFC intake of animals in the control group. The apparent digestibility of NFC is proportional to its intake due to the dilution of the metabolic fecal fraction. On the other hand, a linear decrease in DOM digestibility when replacing corn and soybean meal with wheat bran and urea was observed among the animals that received supplementation (P=0.046; Table 3). Wheat bran contains higher amounts of iNDF than corn and soybean meal, and thus likely reduces overall feed digestibility. This shift in feed composition (hardly digested) may explain the observed decrease in DOM digestibility.

The apNDF digestibility exhibited a quadratic response for the supplemented animals, with the highest value in the treatment with 0% corn and soybean meal substitution for wheat bran and urea (P=0.023; Table 3). These findings may be related to the different substrates that reached the rumen in each treatment. The presence of soluble carbohydrates in the rumen, which usually have greater digestibility, promotes competition among the ruminal microbiota, resulting in a proliferation of starch-degrading microorganisms, which grow more rapidly than fibrolytic ones (El-Shazly et al., 1961; Moura et al., 2020). This competition may lead to the preferential use of starch as an energy source, thereby increasing the concentration of fibrous substrates over time. Furthermore, this competitive dynamic could inhibit fibrolytic activity (Arroquy et al., 2005). Therefore, the treatment with 50% substitution likely resulted in higher substrate variability in the rumen than the treatments with 0% and 100% substitution of corn and soybean meal with wheat bran and urea.

Table 3
Total digestibility and nitrogen levels of suckling beef calves fed with protein-energy supplements in tropical pasture

Item[1]	Treatments[2]				SEM[3]	*P*-value[4]		
	Control	0%	50%	100%		C vs S	L	Q
				(g day^{-1})				
Dry matter	0.592	0.682	0.608	0.597	0.0220	0.213	0.052	0.305
Organic matter	0.636	0.720	0.650	0.645	0.0196	0.190	0.053	0.244
Crude protein	0.700	0.747	0.712	0.737	0.0215	0.262	0.766	0.323
Ether extract	0.858	0.874	0.850	0.874	0.0138	0.663	0.969	0.243
apNDF	0.478	0.503	0.396	0.446	0.0179	0.228	0.088	0.023
NFC	0.664	0.795	0.765	0.716	0.0262	0.035	0.099	0.789
DOM (g kg^{-1} DM)	586	677	609	603	18.3	0.111	0.046	0.237
NMIC (g day^{-1})	27.1148	35.1443	34.5337	32.2443	6.0592	0.382	0.752	0.915
MICNR (g g^{-1} N)	0.506	0.400	0.427	0.404	0.068	0.291	0.968	0.785
EMS (g kg^{-1}DOM)	127.02	97.91	115.84	111.10	18.613	0.432	0.642	0.645

[1]apNDF: neutral detergent fiber corrected for ash and protein residue; NFC: non fibrous carbohydrate; DOM: digested organic matter; DM: Dry matter; NMIC: nitrogen production in the rumen: MICNR: microbial nitrogen: ingested nitrogen ratio; EMS: efficiency of microbial protein synthesis.
[2]Control: Mineral mix only; 0%. 50%. 100%: Replacement levels of corn and soybean meal for wheat bran and urea.
[3]Means standard error of the mean.
[4]Contrast between animals fed with mineral mix only or protein-energy supplement (C vs S) and linear (L) and quadratic (Q) effects for the levels of substitution.

Despite the different CP intakes among control and supplemented animals (P=0.001; Table 2), similar values of nitrogen production in the rumen, microbial nitrogen:ingested nitrogen ratios, and efficiency of microbial protein synthesis were observed (P>0.05; Table 3). This suggests that all treatments provided enough protein and energy substrates for ruminal microbiota development, opposed to those reported by Ortega et al. (2020) and Moreno et al. (2022) in calves that received supplement under grazing conditions.

Blood constituents are closely linked to diet composition and digestibility (Calixto et al., 2008). Therefore, animals receiving supplementation exhibited elevated concentrations of IGF-1 compared to the control group (P=0.041; Table 4), which responds to the nutritional plane as the diet composition (protein and energy level). The above suggests that a higher DM, CP, and TDN intake promoted greater anabolism, consistent with higher final BW in the experimental period. Similar results were obtained by Rodríguez-Sánchez et al. (2015) and Franco et al. (2017), who supplemented cattle in tropical conditions. However, no differences were observed among supplemented animals (P>0.05). Furthermore, supplemented calves exhibited higher SUN concentrations than those in the control group (P<0.001; Table 4), likely because of their increased intake of CP from

supplementation, resulting in higher nitrogen intake. However, blood glucose, triglycerides, total proteins, or albumin concentrations were not affected by supplementation or level of replacement (P>0.05). Similarly, Silva et al. (2017) did not report an increase in the blood concentrations of total proteins or albumins in calves with or without supplementation.

Table 4
Metabolic profile of suckling beef calves fed with protein-energy supplements in tropical pasture

Item[1]	Treatments[2]				SEM[3]	*P*-value[4]			
	Control	0%	50%	100%		C vs S	L	Q	Treat × Day
IGF-1 (ng mL^{-1})	216.1	301.1	268.8	244.6	16.19	0.041	0.069	0.847	-
Glucose (mg dL^{-1})	89.8	93.3	93.0	89.8	2.86	0.530	0.427	0.704	0.109
SUN (mg dL^{-1})	9.6	13.0	13.0	12.6	0.41	<0.001	0.493	0.595	0.450
Triglycerides (mg dL^{-1})	38.3	34.9	31.9	31.4	2.51	0.058	0.322	0.685	0.656
Total Proteins (g dL^{-1})	6.34	6.42	6.24	6.24	0.168	0.836	0.482	0.704	0.311
Albumin (g dL^{-1})	3.39	3.48	3.42	3.37	0.062	0.705	0.283	0.995	0.045

[1]apNDF: neutral detergent fiber corrected for ash and protein residue; NFC: non fibrous carbohydrate; DOM: digested organic matter; DM: Dry matter; NMIC: nitrogen production in the rumen: MICNR: microbial nitrogen: ingested nitrogen ratio; EMS: efficiency of microbial protein synthesis.
[2]Control: Mineral mix only; 0%. 50%. 100%: Replacement levels of corn and soybean meal for wheat bran and urea.
[3]Means standard error of the mean.
[4]Contrast between animals fed with mineral mix only or protein-energy supplement (C vs S) and linear (L) and quadratic (Q) effects for the levels of substitution.

Regarding performance, supplemented animals had an additional gain of approximately 0.2 kg per day compared to the control group, exhibiting a higher final BW than ones (P=0.047; Table 5). This agrees with Carvalho et al. (2019), who reported 0.2 kg as supplemental weight gain for young bull calves supplemented in a creep-feeding system from 3 to 8 months of age. Despite this, the higher final BW and IGF-1 concentrations were not sufficient to influence the ribeye area, SFTL or SFTR, indicators of muscle and adipose tissue deposition among treatments. Such a pattern seems to indicate that both the protein-energy supplementation and levels of replacing were not sufficient to influence the gain composition. This may be attributed to a more even distribution of body tissue deposition in the supplemented animals. Moreover, these results suggest that wheat bran and urea are effective alternatives to corn and soybean meal, without hindering animal performance. These results are similar to those observed by Silva et al. (2017) and Ortega et al. (2020), who did not observe an increase in ADG, ribeye area, or SFTL with an increase in the supplement amount.

Table 5
Performance of suckling beef calves fed with protein-energy supplement in tropical pasture

Item[1]	Treatments[2]				SEM[3]	*P*-value[4]			
	Control	0%	50%	100%		C vs S	L	Q	Treat × Day
IBW (kg)	111.5	111.3	111.4	111.3	5.98	0.984	0.996	0.990	-
FBW (kg)	222.9	246.1	247.0	240.2	6.19	0.047	0.245	0.407	-
ADG (kg)	0.797	0.962	0.969	0.890	0.0440	0.059	0.168	0.334	-
Ribeye area (cm^2)	34.0	35.8	35.7	32.9	1.25	0.561	0.110	0.391	0.744
SFTL (mm)	1.43	1.54	1.53	1.53	0.081	0.273	0.881	0.937	0.604
SFTR (mm)	1.87	2.04	1.97	2.05	0.108	0.234	0.960	0.570	0.114

[1]IBW: initial body weight; FBW: final body weight; ADG: average daily gain; SFTL: subcutaneous fat thickness at the *Longissimus* muscle; SFTR: fat thickness at the rump.
[2]Control: Mineral mix only; 0%. 50%. 100%: Replacement levels of corn and soybean meal for wheat bran and urea.
[3]Means standard error of the mean.
[4]Contrast between animals fed with mineral mix only or protein-energy supplement (C vs S), linear (L) and quadratic (Q) effects for the levels of substitution, and treatment versus day of collection.

Conclusion

Replacing corn and soybean meal with a combination of wheat bran and urea in a protein-energy supplement does not thinder performance, nutritional status, or metabolic parameters in grazing suckling beef calves. Therefore, we recommend supplementing wheat bran and urea at 7 g kg^{-1} of BW as a protein-energy source in a creep-feeding system for grazing suckling beef calves.

Acknowledgments

To the Universidade Federal de Viçosa and Fundação de Amparo à Pesquisa do estado de Minas Gerais (FAPEMIG) for financing and help conducting this study.

References

Almeida, D. M., Marcondes, M. I., Rennó, L. N., Barros, L. V., Cabral, C. H. A., Martins, L. S., Marquez, D. E. C., Saldarriaga, F. V., Villadiego, F. A. C., Cardozo, M. A., Ortega, R. M., Cárdenas, J. E. G., Brandão, V. L. N., & Paulino, M. F. (2018). Estimation of daily milk yield of Nellore cows grazing tropical pastures. *Tropical Animal Health and Production, 50*(4), 1771-1777. doi: 10.1007/s11250-018-1617-4

Arroquy, J. I., Cochran, R. C., Nagaraja, T. G., Titgemeyer, E. C., & Johnson, D. E. (2005). Effect of types of non-fiber carbohydrate on in vitro forage fiber digestion of low-quality grass hay. *Animal Feed Science and Technology, 120*(1-2), 93-106. doi: 10.1016/j.anifeedsci.2005.01.012

Calixto, M., Jr., Arruda, D. S. R., Jobim, C. C., & Santos, G. T. D. (2008). Efeito de diferentes volumosos sobre os constituintes sanguíneos de vacas da raça Holandesa. *Revista Brasileira de Saúde e Produção Animal, 9*(1), 35-44.

Cardenas, J. E. G., Paulino, M. F., Lopes, S. A., Silva, A. G., Barros, L., & Valente, E. L. (2015). Desempeño productivo, consumo y digestibilidad de terneras lactantes criadas en pastoreo suplementadas com diferentes niveles de proteína bruta. *Archivos de Zootecnia, 64*(246), 167-174. doi: 10.21071/az.v64i246. 392

Carvalho, V. V., Paulino, M. F., Detmann, E., Valadares, S. C., Fº., Lopes, S. A., Rennó, L. N., Sampaio, C. B., & Silva, A. G. (2019). A meta-analysis of the effects of creep-feeding supplementation on performance and nutritional characteristics by beef calves grazing on tropical pastures. *Livestock Science, 227*(9), 175-182. doi: 10.1016/j.livsci.2019.07.009

Chen, X. B., & Gomes, M. J. (1992). *Estimation of microbial protein supply to sheep and cattle based on urinary excretion of purine derivatives-an overview of the technical details.* Int Rowett Research Institute.

Detmann, E., Gionbelli, M. P., Paulino, M. F., Valadares, S. C., Fº., & Rennó, L. N. (2016). Considerations on research methods applied to ruminants under grazing. *Nutritime, 13*(3), 4711-4731. https://nutritime.com.br/wp-content/uploads/2020/02/Artigo-381.pdf

Detmann, E., Souza, M. A., Valente, E. E. L., Valadares, S. C., Fº., Queiroz, A. C., Berchielli, T. T., Saliba, E. O. S., Cabral, L. S., Pina, D. S., Ladeira, M. M., & Azevedo, J. A. G. (2012). *Métodos para análise de alimentos.* SUPREMA.

Detmann, E., Valente, É. E. L., Batista, E. D., & Huhtanen, P. (2014). An evaluation of the performance and efficiency of nitrogen utilization in cattle fed tropical grass pastures with supplementation. *Livestock Science, 162*(29), 141-153. doi: 10.1016/j.livsci.2014.01.029

El-Shazly, K., Dehority, B. A., & Johnson, R. R. (1961). Effect of starch on the digestion of cellulose in vitro and in vivo by rumen microorganisms. *Journal of Animal Science, 20*(2), 268-273.

Franco, M. O., Detmann, E., Valadares, S. C., Fº., Batista, E. D., Almeida Rufino, L. M. de, Barbosa, M. M., & Lopes, A. R. (2017). Intake, digestibility, and rumen and metabolic characteristics of cattle fed low-quality tropical forage and supplemented with nitrogen and different levels of starch. *Asian-Australasian Journal of Animal Sciences, 30*(6), 797-803. doi: 10.5713/ajas.16.0629

Goes, R. H. T. B., Mancio, A., Valadares, S. C., Fº., & Lana, R. (2004). Degradação ruminal da matéria seca e proteína bruta, de alimentos concentrados utilizados como suplementos para novilhos. *Ciencia e Agrotecnologia, 28*(1), 167-173. doi: 10.1590/s1413-70542004000100022

Humer, E., & Zebeli, Q. (2017). Grains in ruminant feeding and potentials to enhance their nutritive and health value by chemical processing. *Animal Feed Science and Technology, 226*(5), 133-151. doi: 10.1016/j.anifeedsci.2017.02.005

Lazzarini, I., Detmann, E., Sampaio, C. B., Paulino, M. F., Valadares, S. C., Fº., Souza, M. A. D., & Oliveira, F. A. (2009). Intake and digestibility in cattle fed low-quality tropical forage and supplemented with nitrogenous compounds. *Resvista Brasileira de Zootecnia, 38*(10), 2021-2030. doi: 10.1590/s1516-35982009001000024

Lima, J. A. C., Paulino, M. F., Detmann, E., Rennó, L. N., Fernandes, H. J., Silva, A. G. da, Lopes, S. A., Marquez, D. E. C., Martins, L. S., & Moura, F. H. (2016). Supplementation of grazing suckling beef calves receiving different energy sources. *Semina: Ciências Agrárias, 37*(3), 1527-1538. doi: 10.5433/1679-0359.2016v37n3p1527

Lopes, S. A., Paulino, M. F., Detmann, E., Valente, É. E. L., Barros, L. V. de, Rennó, L. N., & Martins, L. S. (2016). Does supplementation of beef calves by creep feeding systems influence milk production and body condition of the dams? *Tropical Animal Health and Production, 48*(6), 1241-1246. doi: 10.1007/s 11250-016-1083-9

Marquez, D. C., Paulino, M. F., Marcondes, M. I., Rennó, L. N., Barros, L. V. de, Martins, L. S., Ortega, R. M., Silva, A. G. da, Almeida, D. M. de, Sotelo, D. P., & Moura, F. H. (2014). Parâmetros nutricionais e produtivos de bezerras suplementadas a pasto com diferentes fontes de alimentos proteicos. *Semina: Ciências Agrárias, 35*(5), 2709-2722. doi: 10.5433/1679-0359.2014v35n5p2709

Moreno, D. S., Ortega, R. M., Marquez, D. C., Moreira, T. R., Santos, E. J. D., Almeida, D. M., Paulino, M. F., Rennó, L. N., & Detmann, E. (2022). Provision of a protein-rich supplement for grazing suckling female beef calves to improve productive performance and metabolic response. *Animal Bioscience, 35*(8), 1174-1183. doi: 10.5713/ab.21.0226

Moura, F. H., Costa, T. C., Trece, A. S., Melo, L. P. de, Manso, M. R., Paulino, M. F., Rennó, L. N., Fonseca, M. A., Detmann, E., Gionbelli, M. P., & Duarte, M. S. (2020). Effects of energy-protein supplementation frequency on performance of primiparous grazing beef cows during pre and postpartum. *Asian-Australasian Journal of Animal Sciences, 33*(9), 1430-1443. doi: 10.5713/ajas.19.0784

National Research Council (1996). *Nutrient requirements of beef cattle.* National Academy Press.

Ortega, R. M., Paulino, M. F., Detmann, E., Rennó, L. N., Moreno, D. S., Marquez, D. C., Almeida, D. M. de, Martins, L. S., & Ramalho, T. (2020). Supplementation levels for suckling female calves under grazing: productive and nutritional performance and metabolic profile. *Semina: Ciências Agrárias, 41*(3), 945-958. doi: 10.5433/1679-0359.2020v41n3p945

Pan, L., Huang, K. H., Middlebrook, T., Zhang, D., Bryden, W. L., & Li, X. (2021). Rumen degradability of barley, oats, sorghum, triticale, and wheat in situ and the effect of pelleting. *Agriculture, 11*(7), 647-655. doi: 10.3390/agriculture11070647

Porto, M. O., Paulino, M. F., Valadares, S. C., Fº., Detmann, E., Sales, M. F. L., & Couto, V. R. M. (2009). Fontes de energia em suplementos múltiplos para bezerros Nelore em creepfeeding: desempenho produtivo, consumo e digestibilidade dos nutrientes. *Revista Brasileira de*

Zootecnia, 38(7), 1329-1339. doi: 10.1590/s1516-35982009000700024

Rodríguez-Sánchez, J. A., Sanz, A., Tamanini, C., & Casasús, I. (2015). Metabolic, endocrine, and reproductive responses of beef heifers submitted to different growth strategies during the lactation and rearing periods. *Journal of Animal Science, 15*(93), 3871-3885. doi: 10.2527/jas.2015-8994

Signoretti, R. D., Garcia, T. S., Vanessa, D., Souza, F. H. M., Oliveira, E. M., & Pessim, B. (2011). Uréia em dietas para bovinos de origem leiteira em crescimento: consumo de nutrientes e desenvolvimento corporal. *Boletim de Industria Animal, 68*(2), 157-164.

Silva, A. G. da, Paulino, M. F., Amorim, L. S., Rennó, L. N., Detmann, E., Moura, F. H. de, Manso, M. R., Paiva, P. H. S., Ortega, R. E. M., & Melo, L. P. (2017). Performance, endocrine, metabolic, and reproductive responses of Nellore heifers submitted to different supplementation levels pre-and post-weaning. *Tropical Animal Health and Production, 49*(1), 707-715. doi: 10.1007/s11250-017-1248-1

Universidade Federal de Viçosa (2017). *Estação climatológica principal de Viçosa.* Bol. Meteorológico 2017, UFV. https://posmet.ufv.br/wp-content/uploads/2019/09/Estacao-Automatica-2017.pdf

Valadares, S. C., Fº., Costa e Silva, L. F., Gionbelli, M. P., Rotta, P. P., Marcondes, M. I., Chizzotti, M. L., & Prados, L. F. (2016). BR-CORTE 3.0. *Nutritional requirements, diet formulation and performance prediction of zebu and crossbred cattle* (3a ed.). Suprema Gráfica.

Wallis de Vries, M. F. (1995). Estimating forage intake and quality in grazing cattle: a reconsideration of the hand-plucking method. *Journal of Range Management, 48*(4), 370-375. doi: 10.2307/4002491