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Effect of using dried distillery grains in the supplementation of pregnant beef cows on grazing systems and the association of brix degree with blood metabolites in beef calves

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Magister Scientiae

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Dissertation submitted to the Animal Science Graduate Program of the Universidade Federal de Viçosa in partial fulfillment of the requirements for the degree of *Magister Scientiae*.

Adviser: Sidnei Antonio Lopes

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Assent:

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A meus pais que tanto amo, Jacqueline e João.

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“You can conquer hate by ignoring it. You can destroy it by loving the person next to you... celebrating life”
(Chester Charles Bennington)

ABSTRACT

ALBUQUERQUE, Jean Marcelo, M.Sc., Universidade Federal de Viçosa, February, 2025. **Effect of using dried distillery grains in the supplementation of pregnant beef cows on grazing systems and the association of brix degree with blood metabolites in beef calves**. Adviser: Sidnei Antonio Lopes.

In Chapter 1, the objective was to evaluate the effects of prepartum DDG supplementation on the performance, nutritional, and metabolic characteristics of beef cows grazing on low-quality tropical pasture during the peripartum period. Forty-four multiparous Nellore beef cows with an average body weight (BW), body condition score (BCS), and gestation length of 534±43.39 kg, 5.4±0.6 points, and 200 days, respectively, were supplemented with a mineral mixture or with low-intake concentrate supplements (1 kg/animal/day) containing corn and soybean meal-based ingredients or DDG-based ingredients. Supplementation promoted an increase in CP intake and digestibility ($P<0.05$); however, there was no effect on the intake of the other variables studied ($P>0.05$). A positive variation in prepartum BCS was observed ($P<0.05$); nevertheless, no effect was observed on postpartum BCS variation ($P>0.05$). Concentrations of Beta-hydroxybutyrate and Insulin-like growth factor 1 were lower in cows supplemented during the prepartum period ($P<0.05$). Under tropical conditions, DDG supplementation yielded results similar to those supplemented with CM+SM, ensuring a better prepartum energy balance. However, prepartum concentrate supplementation did not influence postpartum effects compared to cows that consumed only the mineral mixture. For Chapter 2, the objective was to quantify the relationship between blood serum Brix, serum globulin concentrations, calf performance at 60 days of age, and maternal milk yield in Nellore cattle. The data used in this study were derived from three studies evaluating supplementation in the last third of gestation and its effects on the peripartum and offspring, conducted between 2022 and 2024, totaling 118 treatment means. There was a strong positive correlation ($r = 0.842$; $P < 0.001$) between globulin concentration and serum Brix. Regression analysis demonstrated a positive linear relationship between globulin concentrations and serum Brix. For every 1 g/dL increase in globulin concentrations, there is a 0.08% increase in Brix. For every 1% increase in serum Brix, there is an increase of 0.05 kg/day and 3.38 kg in weight at 60 days. It is estimated that for every 1 kg increase in average daily milk yield, there is an increase of 4.8 kg in calf body weight at 60 days, 0.064 kg/day, and an impact of 0.24% on calf serum Brix. Blood serum Brix in Nellore calves has a strong relationship with globulin concentration, serving as a strong indicator of passive immunity transfer. Furthermore, cow milk yield explains a significant portion of calf performance up to 60 days of age.

Keywords: zebu; metabolites; peripartum; nutrition; performance ; globulins

RESUMO

ALBUQUERQUE, Jean Marcelo, M.Sc., Universidade Federal de Viçosa, fevereiro de 2025. **Efeito do uso de grãos secos de destilaria na suplementação de vacas de corte prenhes em sistemas de pastejo e a associação do grau brix com metabólitos sanguíneos em bezerros de corte**. Orientador: Sidnei Antonio Lopes.

No capítulo 1 objetivou-se avaliar os efeitos da suplementação no pré-parto com DDG sobre a performance, características nutricionais e metabólicas de vacas de corte em pasto tropical de baixa qualidade durante o periparto. Quarenta e quatro vacas de corte multíparas da raça Nelore com peso corporal médio, escore de condição corporal e dias de gestação de 534±43,39 kg, 5,4±0,6 pontos e 200 dias, respectivamente, foram suplementadas com mistura mineral ou com suplementos concentrados de baixo consumo (1kg/animal/dia), contendo ingredientes à base de milho e farelo de soja ou à base de DDG. A suplementação promoveu aumento no consumo e digestibilidade da PB ($P<0,05$), no entanto, não houve efeito sobre o consumo das demais variáveis estudadas ($P>0,05$). Observou-se uma variação positiva do ECC no pré-parto ($P<0,05$), entretanto, não foi observado efeito na variação do ECC no pós-parto ($P>0,05$). As concentrações de -hidroxibutirato e Fator de crescimento semelhante à insulina tipo 1 foram menores em vacas suplementadas no pré-parto ($P<0,05$). Sob condição tropical, a suplementação de DDG teve resultados semelhantes aos suplementados com CM+SM, garantindo melhor balanço energético no pré-parto. Entretanto, a suplementação concentrada no pré-parto não influenciou efeitos no pós-parto em relação as vacas que consumiram apenas mistura mineral. Para o capítulo 2 objetivou-se quantificar a relação entre brix do soro sanguíneo, concentrações séricas de globulinas e desempenho dos bezerros com 60 dias de vida e a produção de leite materna de bovinos nelore. Os dados utilizados neste estudo advim de três estudos avaliando a suplementação no terço final de gestação e seus efeitos no periparto e prole, entre os anos de 2022 e 2024 e totalizando 118 média de tratamentos. Houve uma correlação forte e positiva ($r = 0,842$; $P<0,001$) entre a concentração de Globulinas e o Brix do Soro. A análise de regressão demonstrou uma relação linear e positiva entre as concentrações de globulinas e o brix do soro. Para cada aumento de 1 g/dL nas concentrações de globulinas, há um incremento de 0,08% de Brix. Para cada aumento de 1% no Brix do soro há um incremento de 0,05 kg/dia e 3,38 kg no peso de 60 dias. Estima-se que para cada aumento de 1 kg na produção média de leite por dia, há um incremento de 4,8 kg de peso vivo dos bezerros aos 60 dias, 0,064 kg/dia e um impacto de 0,24%

de Brix do soro dos bezerros. O brix do soro sanguíneo de bezerros Nelore possui forte relação com a concentração de globulinas, evidenciando forte indicativo da transferência de imunidade passiva. E a produção de leite das vacas explica boa parte do desempenho dos bezerros até os 60 dias de vida.

Palavras-chave: zebu; metabólitos; periparto; nutrição; desempenho; globulinas

SUMMARY

CHAPTER 1 - EFFECT OF USING DRIED DISTILLERY GRAINS IN THE SUPPLEMENTATION OF PREGNANT BEEF COWS ON GRAZING SYSTEMS

Abstract

The objective was to evaluate the effects of prepartum supplementation with DDG on the performance, nutritional, and metabolic characteristics of beef cows grazing on low-quality tropical pasture during the peripartum period. Forty-four multiparous Nellore beef cows with an average body weight (BW), body condition score (BCS), and gestation length of 534±43.39 kg, 5.4±0.6 points, and 200 days, respectively, were supplemented with a mineral mixture or with low-intake concentrate supplements (1 kg/animal/day) containing corn and soybean meal-based ingredients or DDG-based ingredients. Supplementation promoted an increase in CP intake and digestibility ($P<0.05$); however, there was no effect on the intake of the other variables studied ($P>0.05$). A positive variation in prepartum BCS was observed ($P<0.05$); nevertheless, no effect was observed on postpartum BCS variation ($P>0.05$). Concentrations of β-hydroxybutyrate and Insulin-like growth factor 1 were lower in cows supplemented during the prepartum period ($P<0.05$). Under tropical conditions, DDG supplementation yielded results like those supplemented with corn + soybean meal, ensuring a better prepartum energy balance. However, prepartum concentrate supplementation did not influence postpartum effects compared to cows that consumed only the mineral mixture.

Keywords: Zebu, peripartum, metabolism, nutrition

Introduction

During gestation, beef cows have increased requirements due to greater nutrient uptake by the fetus. Consequently, feed restriction directly impacts progeny development (Gionbelli et al., 2023) and the return to cyclicity, potentially reducing the pregnancy rate in the subsequent breeding season.

In tropical conditions, cows in the last third of gestation require special attention because this phase coincides with the dry season, leading to a reduction in forage supply and quality (Paulino et al., 2014). This forage is characterized by lower protein content and high levels of indigestible fiber (Detmann et al., 2014a); additionally, this is the gestation period when maximum fetal growth occurs (Gionbelli et al., 2023). Therefore, the use of protein supplements can correct nutritional deficiencies, improve the digestion of potentially digestible fiber, and increase dry matter intake (DMI) of low-quality forage (Lazzarini et al., 2009), providing low-cost energy compounds derived from the forage (Detmann et al., 2024). Studies evaluating protein supplementation for pregnant cows prepartum observed improvements in body weight (Almeida et al., 2020), reproductive performance (Rodrigues et al., 2020), and the dams' energy balance (Ferreira et al., 2020). However, the use of concentrate supplementation increases production costs, which hinders its adoption for the beef cow category.

The use of dried distillers' grains (DDG), a co-product of corn ethanol production, has been widely employed in cattle feeding in Brazil. This product is characterized by high rumen-undegradable protein (RUP), digestible fiber, and low-fat content (Valadares Filho et al., 2018). Regarding the supplementation of beef cows consuming low-quality forage, the utilization of dried distillers' grains with soluble (DDGS) allowed for the maintenance of body condition score (BCS) and weight gain in late gestation (Kennedy et al., 2016) and led to improved performance and BCS from calving to the breeding season (Wilson; Faulkner; Shike, 2015). However, the inclusion of DDG in supplements for grazing beef cows is still poorly understood.

Thus, our hypothesis is that providing DDG supplements to beef cows during the prepartum period improves productive performance, nutritional characteristics, and metabolic status in the peripartum. Therefore, the objective was to evaluate the effects of prepartum DDG supplementation on the performance, nutritional, and metabolic characteristics of beef cows grazing on low-quality tropical pasture during the peripartum period.

Material and methods

Animals and treatments

All procedures involving the use of animals were previously approved by the Ethics Committee on the Use of Production Animals of the Federal University of Viçosa (Protocol No. 057/2023).

The experiment was conducted at the facilities of the UEPE – Unidade de Ensino, Pesquisa e Extensão of the Department of Animal Science at the Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil, between June and November 2023, corresponding to 154 experimental days. Prepartum samplings were carried out based on the expected calving date (corresponding to day 0), with results presented as negative values (-85, -30, -15, and -1). Postpartum samplings were conducted according to the actual individual calving date, presented as positive values (1, 15, 30, 45, and 60).

Forty-four multiparous Nellore beef cows at 200 days of gestation (inseminated on the same day) were used during the peripartum period, with an initial average weight of 534±43.39 kg and BCS of 5.4±0.6.

The animals were allocated to 12 paddocks uniformly covered with *Urochloa decumbens* grass, measuring approximately 3 ha, provided with water troughs and covered feed bunks accessible from both sides. The chemical composition of the forage is shown in Table 1.

The evaluated treatments were: 1) Control: cows receiving mineral mixture *ad libitum*; 2) Corn meal + soybean meal (CM+SM): cows receiving a concentrate supplement based on corn, soybean meal, mineral mixture, and urea + ammonium sulfate; and 3) DDG: cows supplemented with a concentrate based on DDG, mineral mixture, and urea + ammonium sulfate (Table 2). The concentrate supplements were formulated to supply 40% CP (Valadares Filho et al., 2023) and were offered daily at 11:00 h at a rate of 1 kg/cow during the prepartum period. After calving, all animals received only mineral mixture *ad libitum*.

Performance

Cows were weighed every 30 days at 07:00 h without fasting for monitoring purposes. However, to evaluate weight variation, weights recorded on days -85, 1, and 60 were used. The weight relative to the prepartum period (-1 day) was also utilized, which was individually adjusted according to the average daily gain (ADG) between prepartum weighings (BW at calving). To assess body condition score (BCS) variation, evaluations were performed on days -85, on the expected calving date, and at day 60, using the 1-to-9 scale recommended by the

NRC (1996), conducted by three previously trained evaluators. The following equations were used:

$$PreBW\ change = \frac{BW\ at\ calving - Initial\ BW}{Initial\ BW} x\ 100 \quad (1)$$

$$PostBW\ change = \frac{Final\ BW - BW\ at\ calving}{BW\ at\ calving} x\ 100 \quad (2)$$

$$PreBCS\ change = \frac{BCS\ at\ calving - Initial\ BCS}{Initial\ BCS} x\ 100 \quad (3)$$

$$PostBCS\ change = \frac{Final\ BCS - BCS\ at\ calving}{BCS\ at\ calving} x\ 100 \quad (4)$$

where: PreBW change and PreBCS change: change in body weight or body condition score in late gestation; BW at calving: body weight on the expected calving date; Initial BW and Initial BCS: initial body weight or body condition score of cows in late gestation; PostBW change and PostBCS change: change in body weight or body condition score from calving to beginning of the breeding season; BW at calving and BCS at calving: body weight or body condition score measured on the expected calving date. Final BW and Final BCS: body weight or body condition score at beginning of the breeding session.

Carcass ultrasonography

Concomitantly with BCS evaluations, ultrasound images were taken to measure body composition, assessing the ribeye area (RA), thickness of subcutaneous rib fat (TSR), and thickness of subcutaneous fat on the croup (TSC). The TSC images were taken between the ileum and ischium in a rectilinear position between the two tuberosities until the identification of the upper border of the *Biceps femuris*, while the TSR and RA images were obtained in the intercostal region, between the 12th and 13th ribs, in the middle distal third of the loin eye area. The equipment used was the Aloka ultrasound (SSD 500V®, Aloka, Ltd., Tokyo, Japan) with an 18 cm linear probe. The images were analyzed with BioSoft Toolbox® II for beef (Biotronics Inc., Ames, IA, USA).

Pasture sampling

Pasture sampling was performed every 28 days to determine forage mass per hectare (ha) and potentially digestible dry matter (pdDM)/ha. In each paddock, four samples were randomly collected by cutting at ground level using a 0.5 x 0.5 m metal quadrat to obtain a representative sample of the paddock. Sampling for qualitative evaluation of the pasture consumed by the animals was carried out every 14 days via manual grazing simulation. Samples were weighed and immediately placed in a forced-air oven at 55°C for 72 hours. Subsequently, they were ground in a knife mill and stored for analysis.

The pdDM was estimated according to the following equation by Paulino et al. (2008):

$$pdDM = 0.98\, x\, (100 - NDF) + (NDF - iNDF)\ (5)$$

where: pdDM = forage content of potentially digestible DM; 0.98 = true digestible coefficient of cell content; and NDF and iNDF = forage content of NDF and iNDF, respectively.

Intake and digestibility

To assess intake and digestibility, a trial was performed during the prepartum period, starting 43 days before the expected calving date and lasting 9 days. This consisted of a 5-day adaptation period to chromic oxide (Cr_2O_3) followed by 4 days of fecal collection at different times (10:00, 14:00, 16:00, and 06:00 h) to ensure representative sampling for each animal (Sampaio et al., 2011). The Cr_2O_3 (15 g/animal/day), packaged in paper cartridges, was administered via the esophagus using a metal applicator daily at 10:00 h. Fecal samples were collected immediately after defecation or directly from the rectum and dried in a forced-air oven (55°C for 72 hours). A composite sample was created for each animal based on the four days of collection.

Fecal DM excretion was estimated based on the ratio between the amount of markers administered and its concentration in the feces. Forage dry matter intake was estimated to use iNDF as an internal marker, according to Detmann et al. (2001). It was assumed that supplement intake was equivalent to the amount offered per animal/day.

On the fifth day of the trial, a forage sample was obtained via manual grazing simulation in each paddock separately; this sample was used for chemical analyses and to determine forage intake and digestibility.

Total dry matter intake was calculated as the sum of forage dry matter intake and supplement dry matter.

Milk yield and composition

To estimate milk yield, two milk collections were performed on days 24 and 40 postpartum to obtain an average of production and composition. Calves were separated from their dams at 14:00 h on the day prior to collection. At 16:00 h, calves were reunited with their dams to empty the udder, separated again at 17:00 h, and kept for a 12-hour period in a corral with access to water. The cows were released into a nearby pasture, and the following day at 05:00 h, mechanical milking was performed following the administration of 0.5 mL of oxytocin (10 IU/mL; Ocitovet®, Brazil) into the mammary vein. Daily milk yield for each cow was estimated based on the production during the period (considering the time of calf separation and the time of milking), adjusted to 24 hours (Lopes et al., 2022). After weighing the milk to estimate yield, samples were collected for subsequent analysis. Milk yield was corrected to 4% fat according to the NRC (2001).

Blood sampling

To evaluate metabolic status, blood samples were collected from cows on days -30, -15, 1, 15, 30, 45, and 60 relatives to calving, and from calves on days 1 and 15. Samples were obtained via jugular vein puncture using vacuum tubes with separator gel (BD Vacutainer® SST II Advance). Subsequently, the tubes were centrifuged for 15 minutes, and the serum was immediately frozen at -20ºC for further analysis. All samplings were performed at 07:00 h, except for day 1, when collection occurred between 24 and 32 hours postpartum.

Chemical analysis

Forage, feces, and supplement samples, processed to pass through a 1 mm sieve, were analyzed according to the standard analytical procedures of the National Institute of Science and Technology in Animal Science (INCT-CA; Detmann et al., 2021) for dry matter (DM; method INCT-CA G-003/1), mineral matter (MM; method INCT-CA M-001/2), CP (method INCT-CA N-001/2), and neutral detergent fiber corrected for ash and protein (NDFap) (method INCT-CA F-002/1). Indigestible neutral detergent fiber (iNDF; method INCT-CA F-009/1) was obtained following *in situ* incubation in non-woven fabric bags (100 g/m^2) for 288 hours using samples processed to pass through a 2 mm sieve. Additionally, fecal samples were analyzed for chromium content (method INCT-CA M-005/2). Milk samples were analyzed for protein, fat, lactose, and total solids content using infrared spectroscopy (Ultrasonic Milkanalyser; Lactoscan).

Cow serum concentrations were evaluated for total proteins (K031-Biuret Method, Bioclin®), albumin (K040-Bromocresol Green Method, Bioclin®), glucose (K082–Enzymatic Colorimetric Method, Bioclin®), urea (K056-UV Kinetic Method, Bioclin®), and total cholesterol (K083-Enzymatic Colorimetric Method, Bioclin®). Non-esterified fatty acids (NEFA) and β-hydroxybutyrate (BHB) were analyzed using Randox® kits (FA115-Colorimetric Method and RB1007-Enzymatic Method, respectively). All analyses were performed at the Animal Physiology Laboratory of the Department of Animal Science at the Universidade Federal de Viçosa using an automated biochemical analyzer (Mindray, BS200E).

Serum concentrations of insulin-like growth factor type 1 (IGF-1) in cows and calves were determined using DiaSorin kits on an automated chemiluminescence analyzer (LIASON XL® - DiaSorin).

Serum urea nitrogen (SUN) concentrations were obtained by multiplying urea content by 0.4667. Globulin levels were calculated as the difference between total protein and albumin concentrations.

Statistical analysis

Data was analyzed using the MIXED procedure of the Statistical Analysis System 9.4 (SAS Institute Inc.). This procedure was applied to the response variables of intake, digestibility, weight and BCS variation, and milk yield and composition, considering comparisons between treatments through a set of orthogonal contrasts: control vs. supplemented (CxS) and CM+SM vs. DDG (SxS). Repeated measures over time, such as carcass ultrasound and metabolites, were analyzed considering days (D) and the interaction between treatments and days (TxD) as fixed effects.

The effect of treatment nested within group was considered a random effect; degrees of freedom were estimated using the Kenward-Roger method, and covariance matrices were tested, with the best structure selected based on the corrected Akaike information criterion. A critical probability level of 0.05 was adopted for type I error.

Results

Forage allowance was 1.11 t/ha of pdDM and 57.4 g pdDM/kg of BW (Figure 1).

Supplementation promoted an increase in CP intake and digestibility (Table 3; $P<0.05$), both for cows receiving the CM+SM supplement and for those receiving DDG. However, there was no effect on the intake of the other variables studied ($P>0.05$; Table 3).

A positive variation in prepartum BCS was observed ($P<0.05$; Table 4). However, no effect was observed on postpartum BCS variation ($P>0.05$) or on BW variation in the pre- and postpartum periods ($P>0.05$). There was no effect of supplementation or supplement type on RA, TSR, and TSC in either of the evaluated periods ($P>0.05$). However, there was an effect of evaluation day for RA, TSR, and TSC ($P<0.05$; Figure 2), which were generally lower in the postpartum period. Additionally, there was no effect on milk yield and composition ($P>0.05$; Table 5).

Supplemented cows presented a lower overall average regarding serum glucose concentration (P<0.05). Although no interaction between treatment and measurement day was observed for this variable, levels were generally higher on day 1 and lower at 60 days (Figure 3). There was an interaction between treatment and measurement day for SUN (P<0.05; Figure 4), in which SUN concentration was higher in cows receiving CM+SM on day -30, lower for the control on day -15, and on day 15, it was higher for the DDG treatment compared to the control.

Regarding total serum protein concentrations, contrasts revealed a difference among supplemented cows, where those supplemented with DDG obtained higher concentrations compared to those supplemented with CM+SM. In addition, there was an effect of measurement day on this variable (P<0.05). In general, 30 days before calving, cows showed lower serum concentrations of total proteins (Figure 5). For serum albumin and globulin concentrations, an effect was observed only for sampling days (P<0.05; Figure 5). For albumin, an increase occurred 15 days prepartum followed by a reduction in serum concentrations throughout the remaining sampling days. For globulins, increasing increments in serum concentration were observed, peaking on day 15 postpartum.

Cows receiving the DDG supplement presented higher total cholesterol concentrations compared to those receiving the CM+SM supplement (Table 6; P<0.05). Additionally, there was an effect of sampling day (P<0.05), with an increase in serum concentration 15 days prepartum compared to 30 days prepartum. However, a drop in concentration occurred 1 day postpartum, after which total cholesterol concentration increased and stabilized from 15 days postpartum onwards (Figure 6).

Serum NEFA concentrations varied only with measurement day (P<0.05; Figure 7), with the highest overall mean values observed on days 1 and 15 postpartum, followed by a decline after this period. For BHB, there was an interaction between treatment and sampling day (P<0.05; Figure 8), in which lower BHB concentrations were observed during the prepartum period (days -30 and -15) and 1 day after calving for supplemented cows. Thereafter, BHB concentration did not vary among treatments.

Regarding IGF-1, an interaction between treatment and measurement day was observed (P<0.05; Figure 9). On concentration calving, supplemented cows presented higher serum IGF-1 concentrations compared to the control. However, 15 days before calving, no difference was observed between treatments. Subsequently, 1 day after calving, it was found that cows consuming the DDG supplement presented higher IGF-1 concentrations compared to cows supplemented with CM + SM, while cows in the mineral mixture presented intermediate values. Cow supplementation during the prepartum period did not influence the offspring's birth weight or weight at 60 days of age, nor was the nutritional status affected, as observed by the similar levels of IGF-1 ($P>0.05$; Table 7).

Discussion

Under tropical conditions, protein is the primary limiting nutrient for grazing cattle production. Throughout the study, the forage presented an average CP content of 62.9 g/kg DM, being, therefore, lower than the minimum required for the optimization of NDF degradation (<70-80 g CP kg^{-1} DM; Figueiras et al., 2010; Lazzarini et al., 2009). According to studies conducted in tropical conditions (Detmann; Valadares Filho, 2010; Paulino; Detmann; Valadares Filho, 2008; Poppi et al., 2018), supplying protein to animals consuming low-quality forage promotes the growth of fibrolytic bacteria, increasing ruminal NDF degradation, voluntary forage intake, and energy extraction from fibrous carbohydrates, providing "low-cost" energy to the system. This is highly beneficial, particularly for the beef cow category.

However, positive effects on voluntary forage intake and fiber digestibility were not observed in this study. The absence of an effect on digestible NDF intake indicates that supplementation did not affect forage digestibility. According to Detmann et al. (2014a), positive responses in fiber degradation have been observed with increased dietary CP levels reaching concentrations close to 100 g kg^{-1} DM; furthermore, voluntary forage intake has been stimulated when concentrations approach 145 g kg^{-1} DM (Detmann et al., 2014b). Although an increase in CP intake was observed among supplemented animals, dietary CP content increased only slightly, remaining below the levels suggested by the authors.

During the prepartum period, cows supplemented with concentrate exhibited a positive BW variation due to the additional nutrient supply provided by the supplement. However, no effects were observed in the postpartum period. Other studies (Ferreira et al., 2020; Moreno et al., 2023; Calderaro et al., 2024) also observed similar responses, where supplementation increased ADG during the prepartum phase, but following the cessation of supplementation and the onset of the lactation period, it did not promote a beneficial effect on productive performance.

Cows in the control treatment lost BCS during the prepartum period, indicating that concentrate supplementation was efficient in maintaining body condition during this phase. The effects of supplementation on preserving body reserves have been reported in the literature (Moriel et al., 2024) and may be associated with nutritional constraints arising from the sharp decline in forage CP content, in addition to the physiological and morphological challenges imposed by late gestation on intake. However, in the postpartum period, BCS losses were similar among treatments. According to Gionbelli et al. (2023), maintenance requirements increase with body weight and condition, thus, cows receiving concentrate supplementation calved with better body condition, which led to greater fat mobilization, observed by the reduction in the thickness of subcutaneous fat on the croup (TSC) as well as BCS losses similar to control cows postpartum.

Nutrition is a key factor regulating IGF-1, with protein and energy being essential for the serum regulation of this hormone (Thissen; Ketelslegers; Underwood, 1994). Thus, feed restriction is a primary cause of reduced IGF-1 concentrations (McGuire et al., 1992). Consequently, cows in the control treatment exhibited lower IGF-1 concentrations on day -30. However, due to a possible reduction in DMI in the days leading up to calving, as previously reported by David et al. (2024), this difference was not observed 15 days prepartum.

Prepartum SUN concentration was higher for cows supplemented with concentrate, particularly on day -15 for those receiving CM+SM. This effect may be related to the higher RDP content in these animals' supplements, given that the concentration of this metabolite is positively associated with rumen degradable protein and ruminal ammonia concentrations (Hammond, 1997). The higher SUN concentration observed 15 days postpartum in supplemented cows suggests greater muscle protein mobilization (Ndlovu et al., 2007).

Serum albumin concentrations decreased postpartum, possibly due to the increased demand for amino acids for milk protein synthesis (Contreras, 2000). In contrast, lower serum globulin values were observed from day -30 until 1 day after calving, likely due to colostrogenesis.

As gestation progresses, there is a gradual increase in fetal glucose demand (Freetly; Ferrell, 1998), and after calving, there is a glucose demand for lactation due to uptake by the mammary gland (Larson, 1974; Sletmoen-Olson et al., 2000), reducing serum glucose concentrations in peripartum cows. Cows in the control treatment exhibited higher glucose concentrations. However, these values could only be verified via the difference in least squares means 1 day after calving (76.6 mg/dL for the control; 63.26 mg/dL for CM+SM; and 64.85 mg/dL). Nevertheless, it is important to note that this distinction occurred at calving, when adrenaline stimulates glycogen catabolism (Kolnes et al., 2015) to mitigate stress, a process that likely occurred to a greater extent in control cows, thereby affecting the overall mean.

After calving, there is a need for lipoproteins to transport triglycerides to the mammary gland (Ferreira et al., 2021), resulting in lower circulating total cholesterol concentrations in the first few days postpartum. However, serum concentrations progressively increase over the subsequent days.

Control cows lost more BCS during the prepartum period compared to supplemented cows, indicating greater adipose tissue mobilization. This is corroborated by the higher circulating BHB concentrations in these animals, serving as a mechanism to utilize energy reserves to meet demands beyond fetal growth (Mulliniks et al., 2013; Wood et al., 2013). These results are consistent with other studies (Ferreira et al., 2020; Saraiva et al., 2024). However, it is important to highlight that the values found in this study do not suggest a severe energy deficit in animals.

Conclusion

Under tropical conditions, DDG supplementation yielded results like those supplemented with CM+SM, ensuring a better prepartum energy balance. Thus, DDG can be used as a total replacement for corn and soybean meal. However, prepartum concentrate supplementation did not influence postpartum effects compared to cows that consumed only mineral mixture.

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Tables and figures

Table 1. Chemical composition of *Urochloa decumbens* during the experimental period.

Item	Month					
	June	July	August[3]	September	October	November
Dry matter[1]	441.2	470.2	510.4	522.8	428.8	352.7
Organic matter[2]	899.2	903.6	904.8	905.9	916.3	910.2
Crude protein[2]	61.1	57.7	58.5	59.7	64.9	75.6
Neutral detergent fiber corrected for ash and protein [2]	650.1	671.3	664.8	679.3	665.3	611.3
Indigestible neutral detergent fiber[2]	284.7	304.6	308.1	314.3	310.4	255.1

[1]g/kg of natural matter; [2]g/kg of dry matter; [3]Intake and digestibility analysis.

Table 2. Composition of supplements provided to grazing pregnant Nellore cows during the prepartum period.

Item	Treatments		
	Control	CM+SM	DDG
Ingredients (g/kg of DM)			
Corn meal	.	480	0
Soybean meal	.	350	0
Dried distillers' grains	.	0	830
Urea + ammonium sulfate	.	80	80
Mineral mixture[1]	1000	90	90
Chemical composition			
Dry matter[2]	.	880.3	893.0
Organic matter[3]		884.4	892.4
Crude protein[3]	.	393.1	385.7
Neutral detergent fiber corrected for ash and protein[3]	.	111.2	345.6
Indigestible neutral detergent fiber[3]	.	8.6	37.2

[1]Percentage composition: dicalcium phosphate 50.00; sodium chloride, 47.2; zinc sulfate, 1.50; copper sulfate, 0.7; cobalt sulfate, 0.05; potassium iodate, 0.05; and manganese sulfate 0.5; [2]g/kg of natural matter; [3]g/kg of dry matter.

Table 3. Intake and digestibility of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplement (CM+SM), or dried distillers' grains-based supplement (DDG) during the prepartum period.

Item	Treatments			SEM[1]	P-value[2]	
	Control	CM+SM	DDG		CxS	SxS
Intake		kg/d				
Forage dry matter	6.76	6.77	6.02	0.596	0.629	0.400
Total dry matter	6.76	7.65	6.92	0.596	0.495	0.409
Total organic matter	6.16	6.82	6.23	0.523	0.586	0.449
Neutral detergent fiber corrected for ash and protein	4.41	4.48	4.32	0.379	0.980	0.769
Indigestible neutral detergent fiber	1.85	2.05	1.94	0.106	0.317	0.494
Crude protein	0.44	0.77	0.64	0.059	0.006	0.157
Digestible dry matter	2.21	2.52	2.10	0.392	0.835	0.471
Digestible neutral fiber	0.94	0.99	0.92	0.099	0.901	0.599
Digestible organic matter	2.33	2.54	2.18	0.370	0.952	0.514
Intake		g/kg PV				
Forage dry matter	12.45	12.29	10.86	1.002	0.495	0.342
Total dry matter	12.46	13.90	12.48	0.996	0.566	0.346
Total organic matter	11.35	12.39	11.24	0.876	0.677	0.385
Neutral detergent fiber corrected for ash and protein	8.11	7.88	7.33	0.656	0.541	0.568
Indigestible neutral detergent fiber	3.42	3.72	3.50	0.192	0.421	0.427
Digestible dry matter	4.06	4.55	3.79	0.694	0.895	0.460
Digestible neutral fiber	1.72	1.80	1.65	0.170	0.995	0.555
Digestibility		%				
Dry matter	31.99	32.53	29.98	3.35	0.861	0.604
Organic matter	37.19	36.64	34.74	3.22	0.711	0.687
Crude protein	23.50	43.87	35.29	6.31	0.067	0.362
Neutral detergent fiber corrected for ash and protein	50.60	47.74	47.02	3.29	0.445	0.882

[1]SEM: Standard error of the mean; [2] P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS).

Table 4. BW and BCS change (%) of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplement (CM+SM), or dried distillers' grains-based supplement (DDG) during the prepartum period and their effects in the postpartum period.

Item	Treatments			SEM[1]	P-value[2]	
	Control	CM+SM	DDG		CxS	SxS
Body weight change		%				
Prepartum	-0.91	3.12	2.32	1.33	0.056	0.686
Postpartum	-7.57	-7.15	-7.21	1.20	0.799	0.971
Body condition score change		%				
Prepartum	-7.31	0.96	1.72	2.56	0.025	0.845
Postpartum	-5.49	-9.83	-6.08	2.94	0.515	0.402

[1]SEM: Standard error of the mean; [2] P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS).

Table 5. Milk yield and composition of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or supplemented with dried distillers' grains (DDG) during the prepartum period and their effects in the postpartum period.

Item	Treatments			SEM[1]	P-value[2]	
	Control	CM+SM	DDG		CxS	SxS
		kg/dia				
Milk yield 4%	6.06	7.04	6.04	0.680	0.590	0.335
Composition		%				
Protein	3.18	3.23	3.35	0.040	0.063	0.076
Lactose	4.79	4.87	5.05	0.060	0.059	0.078
Fat	4.56	4.62	4.63	0.330	0.879	0.976
Total solids	13.14	13.50	13.84	0.332	0.224	0.490

[1]SEM: Standard error of the mean; [2] P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS).

Table 6. Serum concentration of glucose, total proteins, and total cholesterol of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplement

(CM+SM), or dried distillers' grains-based supplement (DDG) during the prepartum period and their effects in the postpartum period.

Item	Treatments			SEM[1]	P-value[2]			
	Control	CM+SM	DDG		CxS	SxS	D	TxD
Glucose, mg/dL	58.31	53.44	53.97	1.52	0.020	0.806	<0,0001	0.470
Total proteins, g/dL	6.39	6.68	6.37	0.09	0.285	0.032	<0,0001	0.142
Total Cholesterol, mg/dL	119.49	109.32	129.5	5.37	0.990	0.012	<0,0001	0.179

[1] SEM: Standard error of the mean; [2] P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS).

Table 7. Performance and IGF-1 of the progeny of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplement (CM+SM), or dried distillers' grains-based supplement (DDG) during the prepartum period and their effects in the postpartum period.

Item	Treatments			SEM[1]	P-value[2]			
	Control	CM+SM	DDG		CxS	SxS	D	TxD
Body weight		kg			0.336	0.499	<0,0001	0.837
Birth	37.5	34.8	31.3	2.84				
60 day	84.4	83.3	81.2	3.15				
IGF-1		ng/dL			0.485	0.493	<0,0001	0.0905
Birth	210.1	206.4	184.7	16.76				
15 day	251.5	296.5	286.8	17.45				

[1] SEM: Standard error of the mean; [2] P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS).

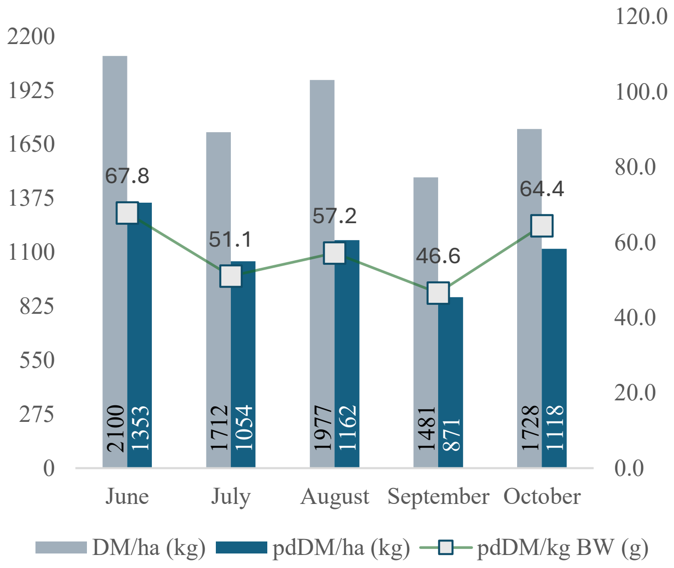
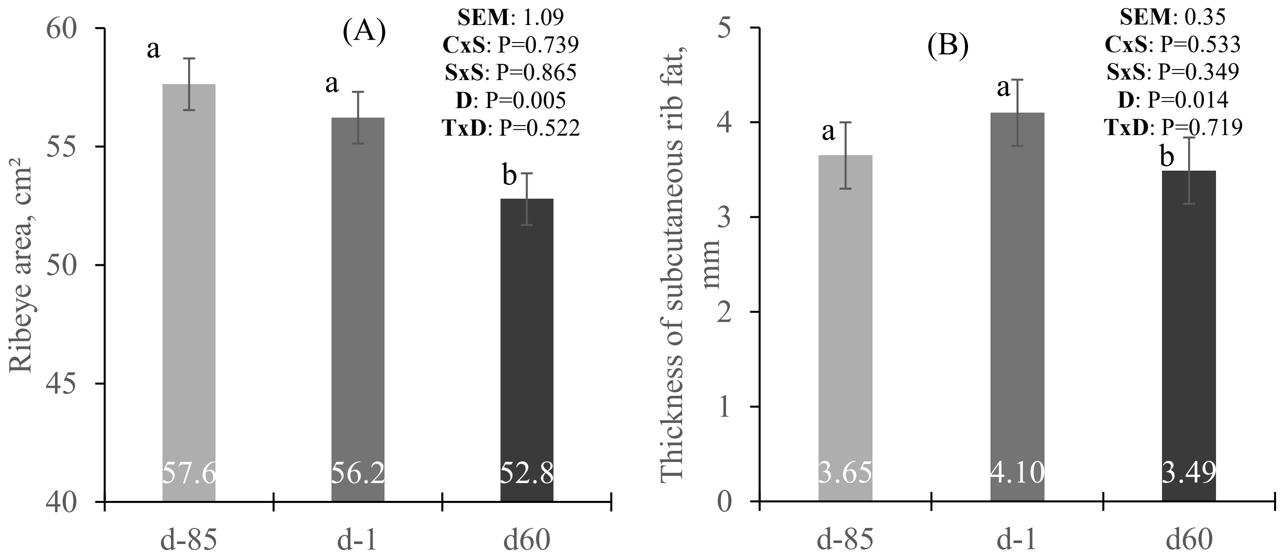


Figure 1. Availability of DM/ha, pdDM/ha, and pdDM/kg BW of *Urochloa decumbens* throughout the experimental period.



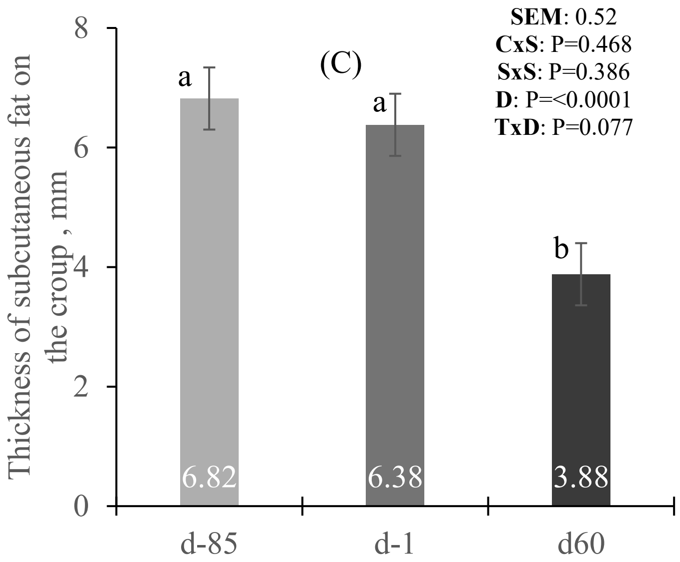


Figure 2. Carcass ultrasound and the characteristics ribeye area (A), thickness of subcutaneous rib fat (B), and thickness of subcutaneous fat on the croup (C) of grazing Nellore cows during the peripartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences between sampling days (P<0.05).

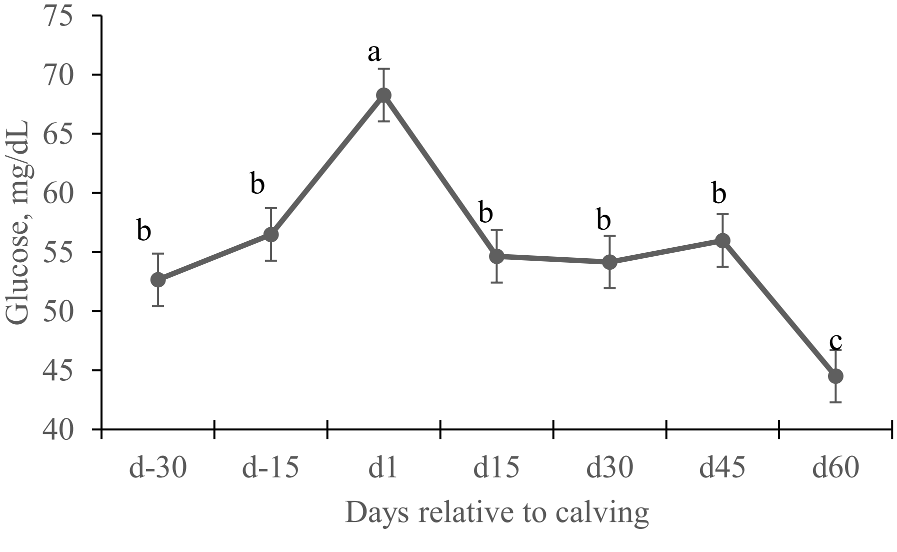


Figure 3. Serum glucose concentration of grazing Nellore cows during the peripartum period. Different letters indicate statistical differences between sampling days (P<0.05).

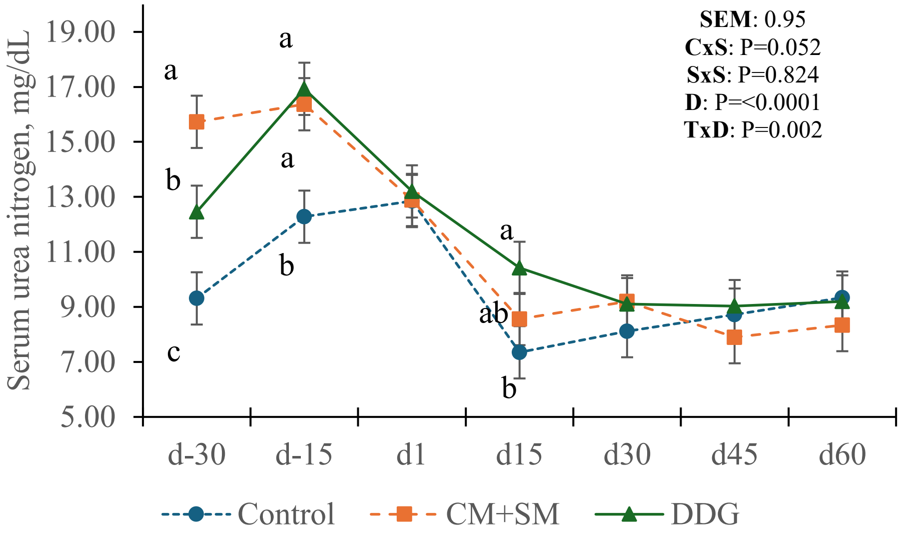


Figure 4. Serum SUN concentration of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or supplemented with dried distillers' grains (DDG) during the prepartum period and its effect in the postpartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences for the interaction between treatments and days (P<0.05).

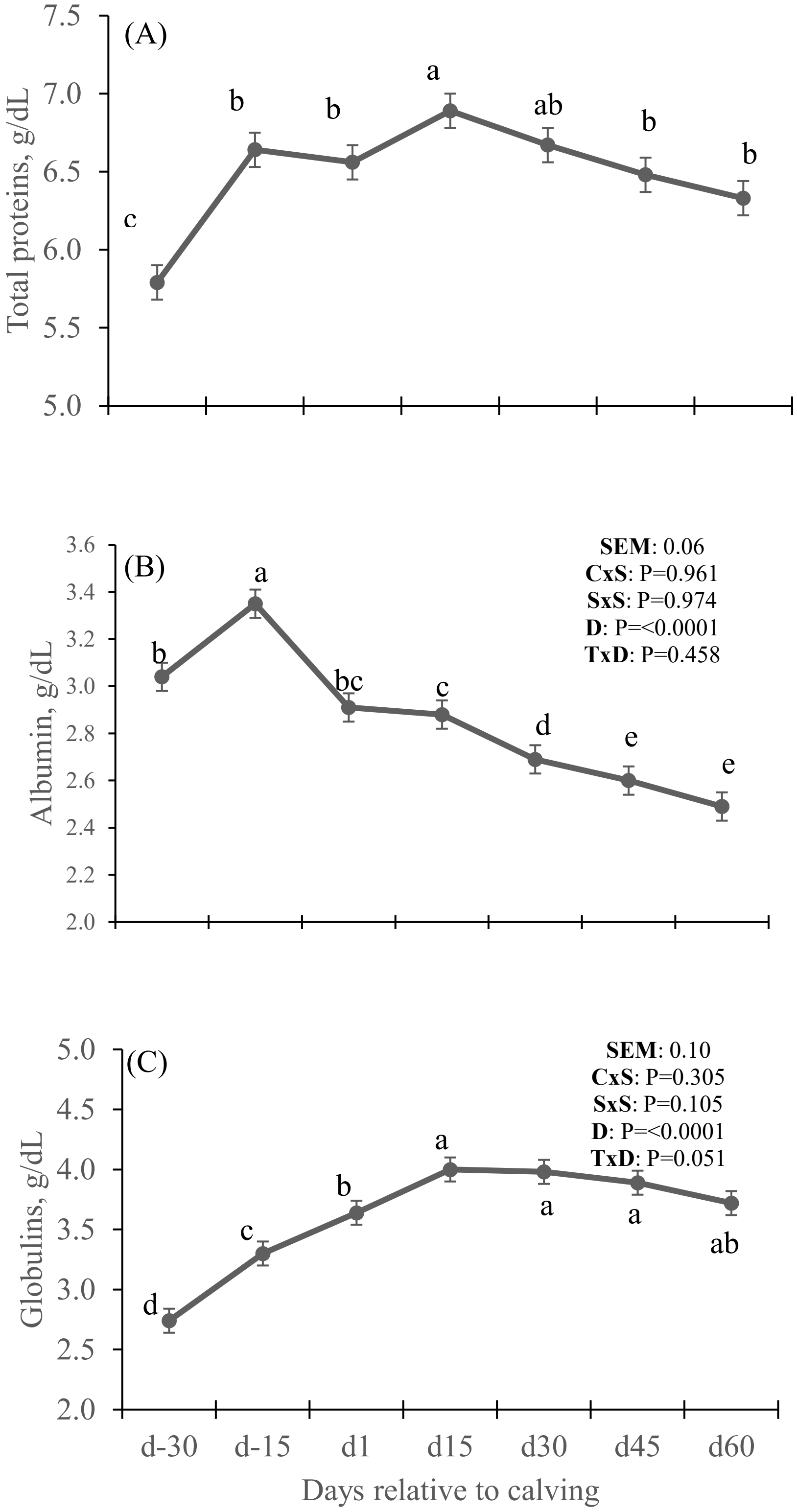


Figure 5. Serum concentration of total proteins (A), albumin (B), and globulins (C) of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or dried distillers' grains-based supplementation (DDG) during the

prepartum period and its effect in the postpartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences between sampling days (P<0.05).

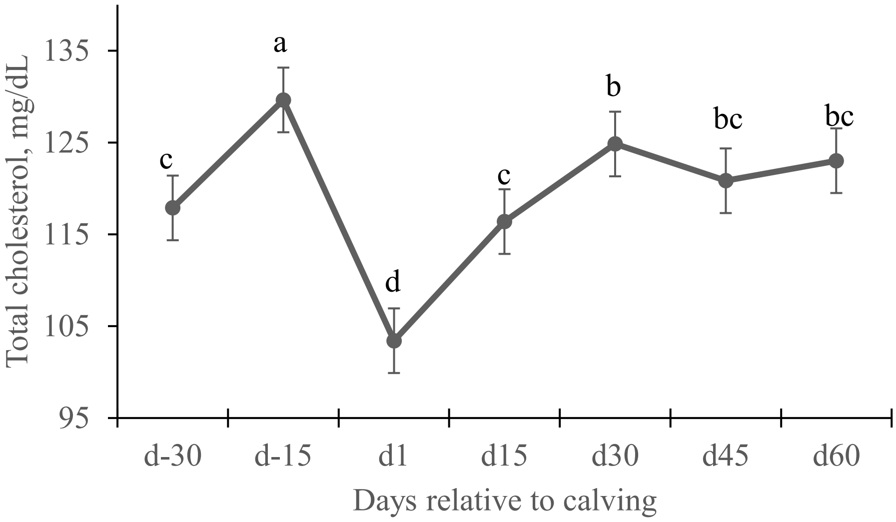


Figure 6. Serum total cholesterol concentration of grazing Nellore cows during the peripartum period. Different letters indicate statistical differences between sampling days (P<0.05).

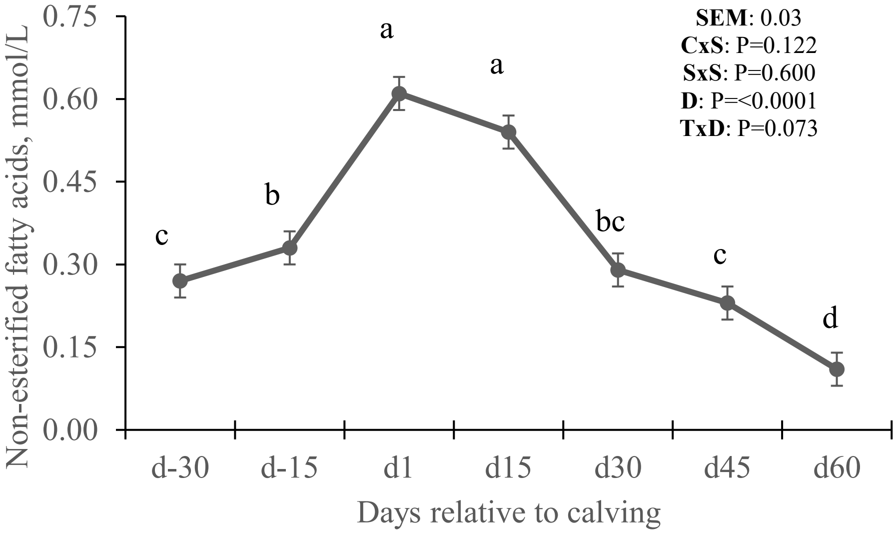


Figure 7. Serum NEFA concentration of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or supplemented with dried distillers' grains (DDG) during the prepartum period and its effect in the postpartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences between sampling days (P<0.05).

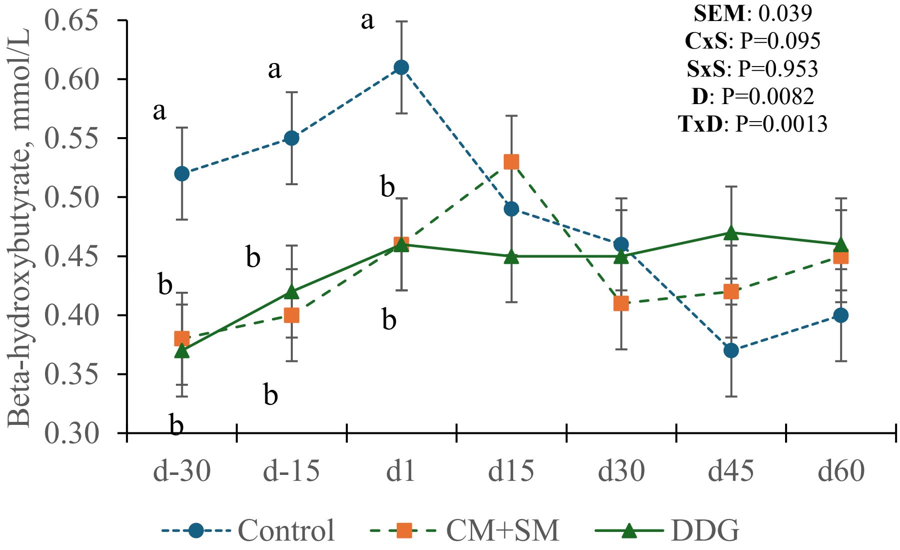


Figure 8. Serum BHB concentration of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or supplemented with dried distillers' grains (DDG) during the prepartum period and its effect in the postpartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences for the interaction between treatments and days (P<0.05).

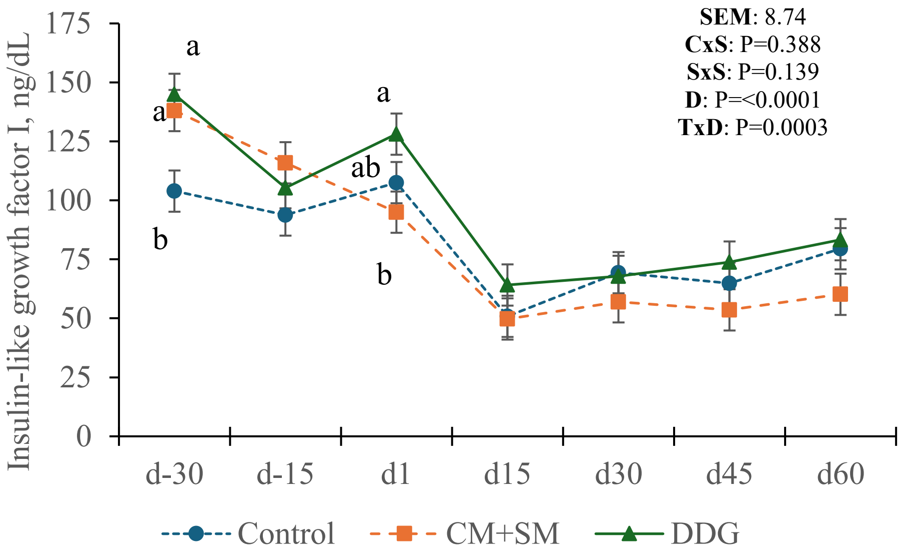


Figure 9. Serum IGF-1 concentration of Nellore cows consuming only mineral mixture (Control), corn and soybean meal-based supplementation (CM+SM), or supplemented with dried distillers' grains (DDG) during the prepartum period and its effect in the postpartum period. Standard error of the mean (SEM), P-value: contrast between supplemented and non-supplemented cows (CxS); contrast between supplemented cow treatments (SxS); effect of sampling days (D); interaction effect of treatment and days (TxD). Different letters indicate statistical differences for the interaction between treatments and days ($P<0.05$).

CHAPTER 2 - ASSOCIATION OF BRIX DEGREE WITH BLOOD METABOLITES IN BEEF CALVES

Abstract

The objective was to quantify the relationship between blood serum Brix, serum globulin concentrations, calf performance at 60 days of age, and maternal milk yield in Nellore cattle. The data used in this study were derived from three studies evaluating supplementation in the last third of gestation and its effects on the peripartum and offspring, conducted between 2022 and 2024, totaling 118 treatment means. There was a strong positive correlation (r = 0.842; P<0.001) between globulin concentration and serum Brix. Regression analysis demonstrated a positive linear relationship between globulin concentrations and serum Brix. For every 1 g/dL increase in globulin concentrations, there is a 0.08% increase in Brix. For every 1% increase in serum Brix, there is an increase of 0.05 kg/day and 3.38 kg in weight at 60 days. It is estimated that for every 1 kg increase in average daily milk yield, there is an increase of 4.8 kg in calf body weight at 60 days, 0.064 kg/day, and an impact of 0.24% on calf serum Brix. Blood serum Brix in Nellore calves is strongly related to globulin concentration, providing a strong indication of passive immunity transfer. Furthermore, cow milk yield explains a significant portion of calf performance up to 60 days of age.

Keywords: Zebu, calves, average daily gain, performance

Introduction

Colostrum intake within the first 6 hours of life is extremely important for calf health, as the absorption capacity is reduced by up to 50% after this period (Cortese, 2009). This is the primary method for the passive transfer of immunity, given that there is no placental transfer of antibodies in cattle (Tizard, 2012) to protect against pathogens.

The transfer of passive immunity (TPI) can be assessed by verifying the total protein or immunoglobulin (IgG) in the calf's serum or plasma (Lombard et al., 2020). Studies indicate that at least 10 g/L of serum IgG is necessary to ensure health and prevent disease in calves (Besser et al., 1991; Furman-Fratczak et al., 2011), along with 5.0 to 6.3 g/dL of total serum protein (Hogan et al., 2015; Godden et al., 2019; Lombard et al., 2020) for dairy cattle.

There are direct methods to analyze these parameters, however, they are generally costly and/or require sending samples to laboratories, which delays the verification of results. Thus, indirect methods, such as the use of a Brix refractometer (% Brix), are employed to correlate colostrum intake quality due to their ease of on-farm use. Studies have found that serum Brix values lower than 8.4% in dairy calves (Deelen et al., 2014) and 7.9% (Gamsjäger et al., 2021) or 8.4% in beef calves (Marcos, 2024) can result in failure of transfer of passive immunity (FTPI).

However, beyond ensuring survival, it is necessary to understand how the initial immune status influences the performance of these animals throughout their lives. Therefore, the objective of this study was to quantify the relationship between blood serum Brix, serum globulin concentrations, calf performance at 60 days of age, and maternal milk yield in Nellore cattle.

Material and methods

Animals and Procedures

All procedures involving the use of animals were previously approved by the Ethics Committee on the Use of Production Animals of the Federal University of Viçosa (Protocol Nos. 011/2022, 057/2023, and 034/2024).

Data were collected during three parallel studies (Gonçalves, 2025; Albuquerque, 2025; Coelho, *[S.d.]*) at the UEPE – Unidade de Ensino, Pesquisa e Extensão of the Department of Animal Science at the Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil. The collection period covered the calving seasons (September to November) between 2022 and

2024 (Table 1). All studies included control treatments (mineral mixture *ad libitum* only) and supplemented treatments (protein supplement at 2 g/kg BW) for dams during the prepartum period.

Calf blood samples were collected between 24 and 36 hours after birth to avoid interfering with the mother-offspring bond. Collection was performed via jugular vein puncture using vacuum tubes with a separator gel (BD Vacutainer® SST II Advance). Samples were centrifuged for 15 minutes, and the serum was immediately frozen at -20ºC for subsequent analyses. An ATC Optical Refractometer with a 0 to 32% Brix scale was used to evaluate calf serum samples immediately after collection. The refractometer was calibrated and washed with distilled water between samples.

To evaluate performance, calves were weighed between 24 and 36 hours after birth and at 60 days of age. Average daily gain (ADG) was calculated based on the relationship between the 60-day weight and the weight after birth.

To estimate milk yield, mechanical milking was performed in each study. Calves were separated from their dams at 14:00 h on the day prior to milk collection. At 16:00 h, calves were reunited with their dams to empty the udder, separated again at 17:00 h, and kept for a 12-hour period in a corral with access to water. The cows were released into a nearby pasture, and the following day at 05:00 h, mechanical milking was performed following the administration of 0.5 mL of oxytocin (10 IU; Ocitovet®, Brazil) into the mammary vein. Daily milk yield for each cow was estimated based on the production during the period (considering the time of calf separation and the time of milking), adjusted to 24 hours (Lopes et al., 2022).

Chemical Analyses

Serum globulin concentrations were calculated as the difference between total protein and albumin concentrations, determined using Bioclin® kits (Belo Horizonte, Brazil) via the chemiluminescence method on an automated biochemical analyzer (Mindray, BS200E, Shenzhen, China).

Statistical Analyses

Statistical analyses were performed using SAS software (version 9.4). Initially, descriptive statistics for performance variables (BW1, BW60, ADG), blood parameters, and milk yield were obtained using the UNIVARIATE procedure. Pearson correlations between dependent variables were calculated to verify the degree of linear association between traits.

Data were subjected to the MIXED procedure of SAS. The model included the fixed effect of treatment and the random effect of study. The effect of sex and the treatment x sex interaction were initially tested but removed from the final model due to a lack of statistical significance for the analyzed variables. Covariance matrices were tested, and the best structure was selected based on the corrected Akaike information criterion.

Additionally, linear mixed regression analyses were performed to evaluate the association between variables. Data were adjusted for the random effect of study. A critical probability level of 0.05 was adopted for type I error.

Results

Pearson correlations were evaluated among the variables milk yield, milk Brix, birth weight, body weight at 60 days of age, average daily gain, and calf blood serum Brix (Table 2).

There was a strong positive correlation (r = 0.842; P<0.001) between globulin concentration and serum Brix. Milk yield showed a strong positive correlation with weight at 60 days (r = 0.764; P<0.001) and average daily gain (r = 0.732; P<0.001). A very strong positive correlation was observed between weight at 60 days and average daily gain (r = 0.920; P<0.001). A weak positive correlation was noted between serum Brix and body weight at 60 days (r = 0.389; P<0.001) and with average daily gain (r = 0.377; P = 0.001).

No significant effects were found on milk yield, milk Brix, weight at 60 days, average daily gain, serum Brix, or globulins (Table 3) regarding prepartum supplementation (P>0.05), calf sex (P>0.05), or the interaction between prepartum supplementation and calf sex (P>0.05).

Birth weight was significantly affected by prepartum supplementation (P = 0.002), with non-supplemented cows calving heavier calves. A sex effect was also observed (P = 0.002), where males were heavier than females at birth.

Regression analysis demonstrated a positive linear relationship between globulin concentrations and serum Brix (Figure 1). For every 1 g/dL increase in globulin concentrations, there is a 0.08% increase in Brix. For every 1% increase in serum Brix, there is an increase of 0.05 kg/day (Figure 2) and 3.38 kg in weight at 60 days (Figure 3). It is estimated that for every 1 kg increase in average daily milk yield, there is an increase of 4.8 kg in calf body weight at 60 days (Figure 4), 0.064 kg/day (Figure 5), and an impact of 0.24% on calf serum Brix (Figure 6).

Discussion

Calf data in this study were obtained from three different trials on supplementation in the last third of gestation of grazing Nellore cows. It was observed that this supplementation negatively influenced the offspring's birth weight. However, this is an unexpected result that may have been subject to other external influences, such as genetics and the cow's phenotype.

The use of the Brix refractometer proved effective for evaluating the transfer of passive immunity in newborn calves. This result was evidenced by the relationship between serum Brix and globulins, with a coefficient of determination indicating that 73.4% of the observed variation in serum Brix can be explained by serum globulin concentration. Several studies have validated the use of the Brix refractometer for this function (Deelen et al., 2014; Godden et al., 2019; Lombard et al., 2020; Gamsjäger et al., 2021; Akköse et al., 2022; Marcos, 2024) and defined the serum Brix threshold between 7.9 and 8.4% to identify potential FTPI in beef and dairy calves.

Although having lower predictive power, serum Brix showed a positive and significant relationship regarding these animals' performance. Sutter et al. (2023), associating FTPI and the performance of dairy calves, verified that the better the passive immunity transfer, the higher the ADG. This aspect may be related to a lower incidence of diseases (Waldner; Rosengren, 2009; Windeyer et al., 2014; Lombard et al., 2020) in animals that had good passive immunity transfer, which directly influences performance.

Cow milk yield is the primary nutrient source for calves during the first weeks, being sufficient to meet all energy and protein requirements of Nellore calves up to the 12th week of life (Lopes et al., 2023), which explains the results obtained in the relationships involving maternal milk production and their calves' performance. Even after this period, the animal does not replace milk intake with pasture and/or supplement (Lopes et al., 2017), leading to better performance until weaning.

Conclusion

Blood serum Brix in Nellore calves has a strong relationship with globulin concentration, serving as a strong indicator of passive immunity transfer. Furthermore, cow milk yield explains a significant portion of calf performance up to 60 days of age.

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Tables and figures

Table 1. Summary of experimental data.

Item	Average	Minimum	Median	Maximum	s	n
			Cows			
Milk Yield, kg/d	6.07	1.6	6.1	9.7	1.708	101
Milk Brix, %	12.0	8.4	11.8	21.2	2.061	117
			Calves			
Body Weight at Birth, kg	34.9	20	35	46	5.069	118
Body Weight at 60 d, kg	81.4	50	80	110	11.75	92
Average Daily Gain, kg	0.786	0.339	0.780	1.144	0.1572	92
Serum Brix, %	10.2	7.2	10.2	13.1	1.242	117
Globulins, g/dL	3.86	1.0	3.9	8.0	1.241	118

Table 2. Correlation matrix of maternal and Nellore calf variables.

Variable	Variable					
	Milk Yield, kg/d	Milk Brix, %	Body Weight at Birth, kg	Body Weight at 60 d, kg	Average Daily Gain, kg	Serum Brix, %

Milk Brix, %	0.315 *0.007*					
Body Weight at Birth, kg	0.415 *<0.001*	-0.006 *0.957*				
Body Weight at 60 d, kg	0.764 *<0.001*	0.283 *0.017*	0.620 *<0.001*			
Average Daily Gain, kg	0.732 *<0.001*	0.351 *0.002*	0.263 *0.027*	0.920 *<0.001*		
Serum Brix, %	0.338 *0.004*	-0.138 *0.254*	0.202 *0.093*	0.389 *<0.001*	0.377 *0.001*	
Globulins, g/dL	0.348 *0.003*	-0.150 *0.214*	0.372 *0.001*	0.459 *<0.001*	0.378 *0.001*	0.842 *<0.001*

Table 3. Effect of prepartum cow supplementation on maternal and Nellore calf variables.

Item	Prepartum Supplementation						
	No		Yes		P-Value[1]		
	Male	Female	Male	Female	Sup.	Sex	Sup x Sex
				Cows			
Milk Yield, kg/d	5.83±0.684	6.38±0.684	6.48±0.594	6.22±0.606	0.47	0.66	0.22

Milk Brix, %	11.4±0.42	11.7±0.53	12.0±0.31	12.6±0.34	0.056	0.252	0.714
Calves							
Body Weight at Birth, kg	38.4±1.40	34.8±1.57	34.9±1.26	32.9±1.29	0.002	0.002	0.37
Body Weight at 60 d, kg	84.7±3.82	81.8±4.08	80.7±3.40	78.2±3.51	0.12	0.28	0.95
Average Daily Gain, kg	0.790±0.0444	0.793±0.0488	0.779±0.0369	0.776±0.0390	0.689	0.984	0.932
Serum Brix, %	9.94±0.254	10.1±0.33	10.4±0.19	10.3±0.21	0.18	0.99	0.55
Globulins, g/dL	3.55±0.275	3.97±0.338	3.98±0.217	3.89±0.232	0.48	0.50	0.31

[1] P-value: effect of maternal supplementation (Sup.); effect of calf sex (Sex); interaction between maternal supplementation effect and calf sex (Sup x Sex).

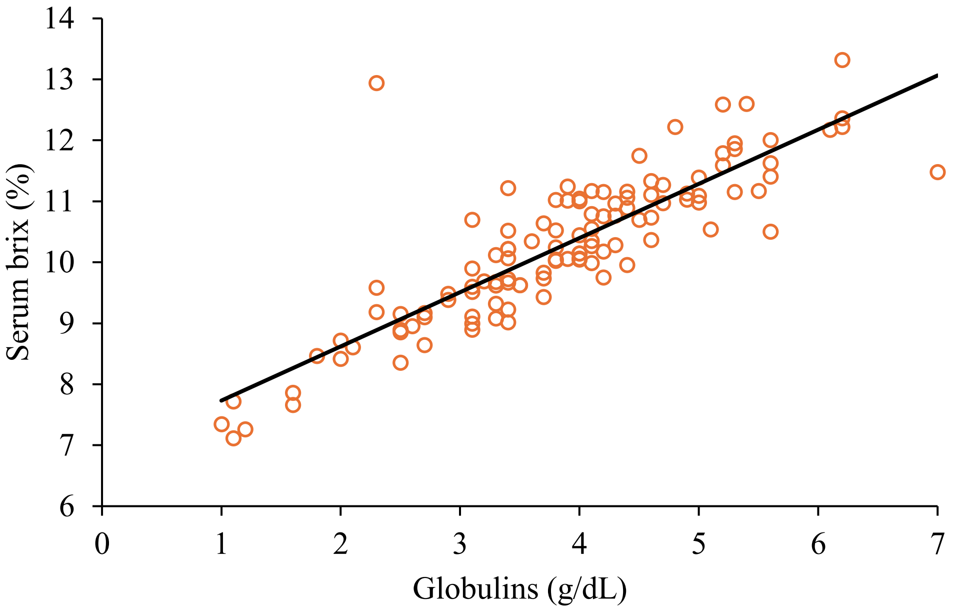

Figure 1. Relationship between globulins and the serum brix of 1-day-old calves ($\hat{Y}$ = 6.84±0.27 + 0.887±0.081×X; s_{XY} = 0.644; r^2 = 0.734; n = 117). The data points were adjusted for random study effects.

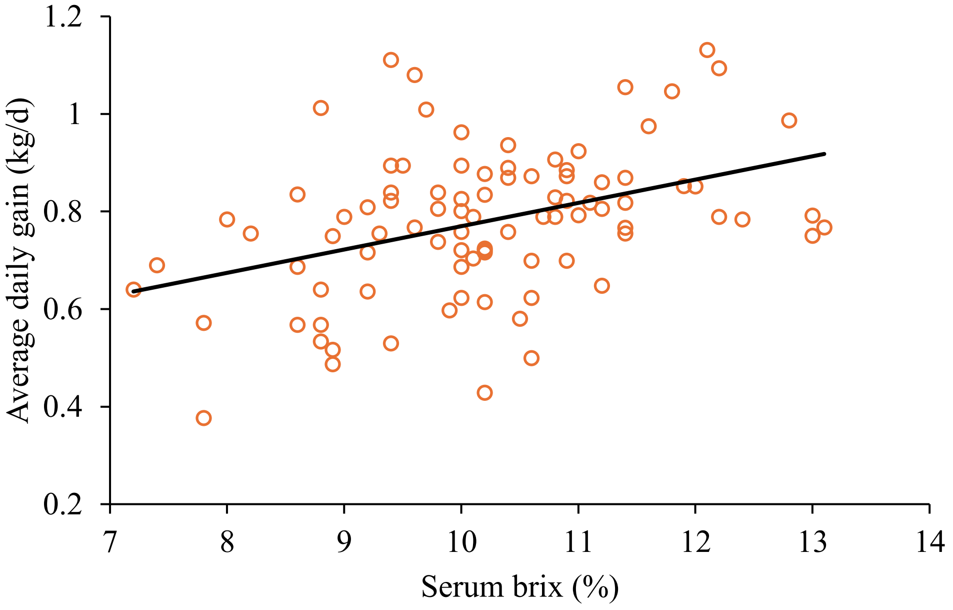

Figure 2. Relationship between serum brix of 1-day-old calves and the average daily gain of the calves ($\hat{Y}$ = 0.29±0.124 + 0.05±0.012×X; s_{XY} = 0.141; r^2 = 0.158; n = 91). The data points were adjusted for random study effects.

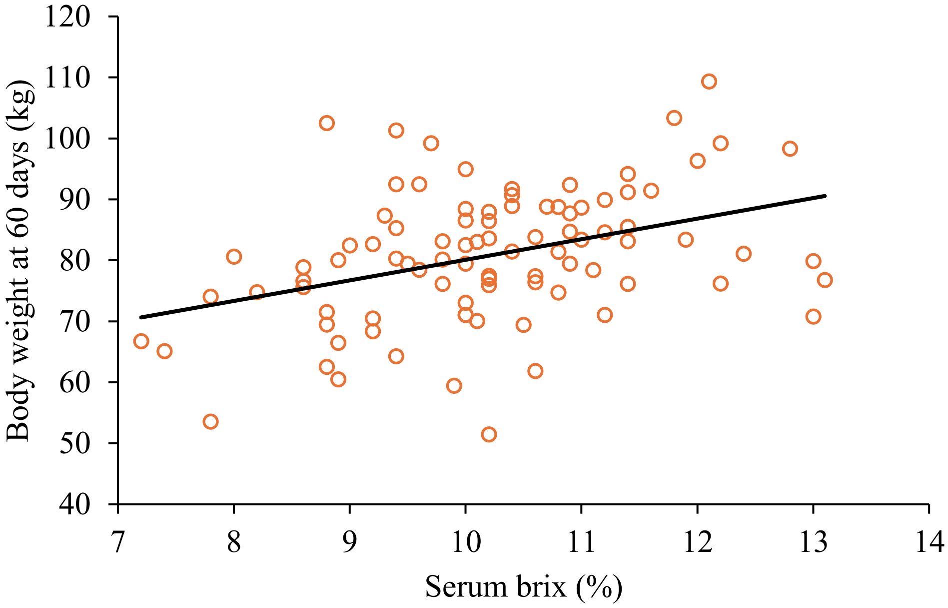

Figure 3. Relationship between serum brix of 1-day-old calves and the body weight at 60 days of the calves ($\hat{Y} = 46.3 \pm 8.98 + 3.38 \pm 0.910 \times X$; $s_{XY} = 10.35$; $r^2 = 0.148$; $n = 76$). The data points were adjusted for random study effects.

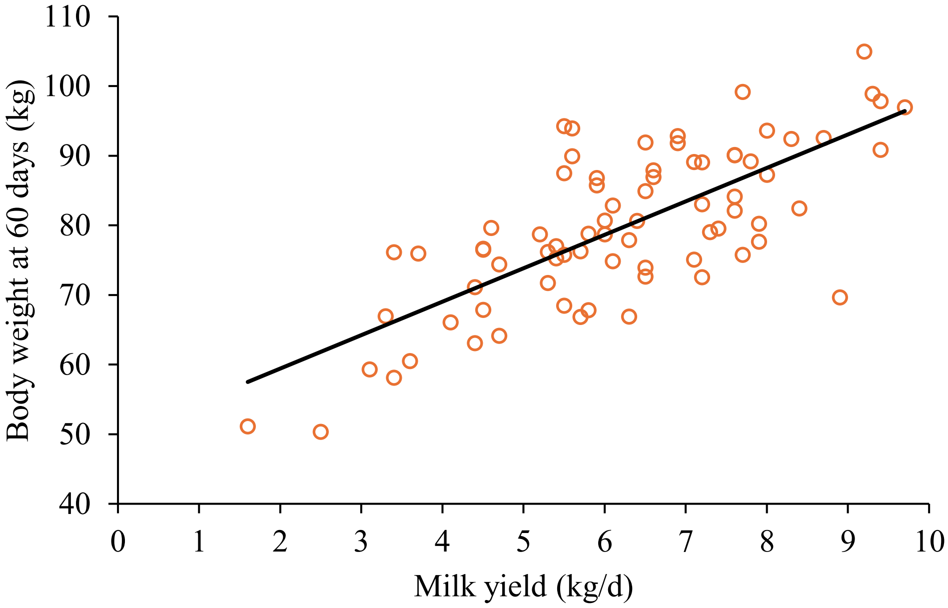

Figure 4. Relationship between milk yield of the dams and the body weight at 60 days of the calves ($\hat{Y} = 49.8 \pm 3.59 + 4.80 \pm 0.642 \times X$; $s_{XY} = 7.82$; $r^2 = 0.540$; $n = 76$). The data points were adjusted for random study effects.

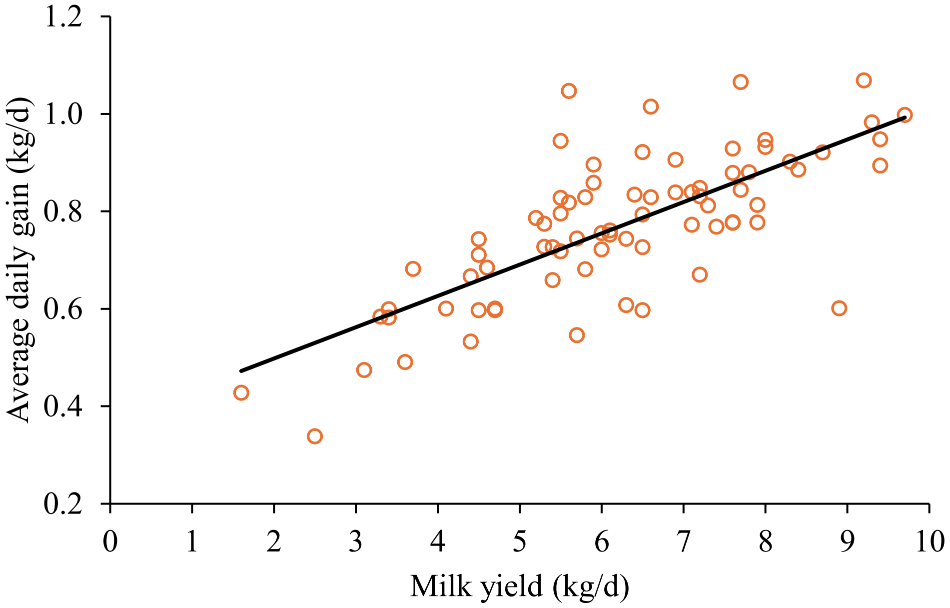

Figure 5. Relationship between milk yield of the dams and the average daily gain of the calves ($\hat{Y} = 0.37 \pm 0.048 + 0.064 \pm 0.0090 \times X$; $s_{XY} = 0.104$; $r^2 = 0.545$; $n = 76$). The data points were adjusted for random study effects.

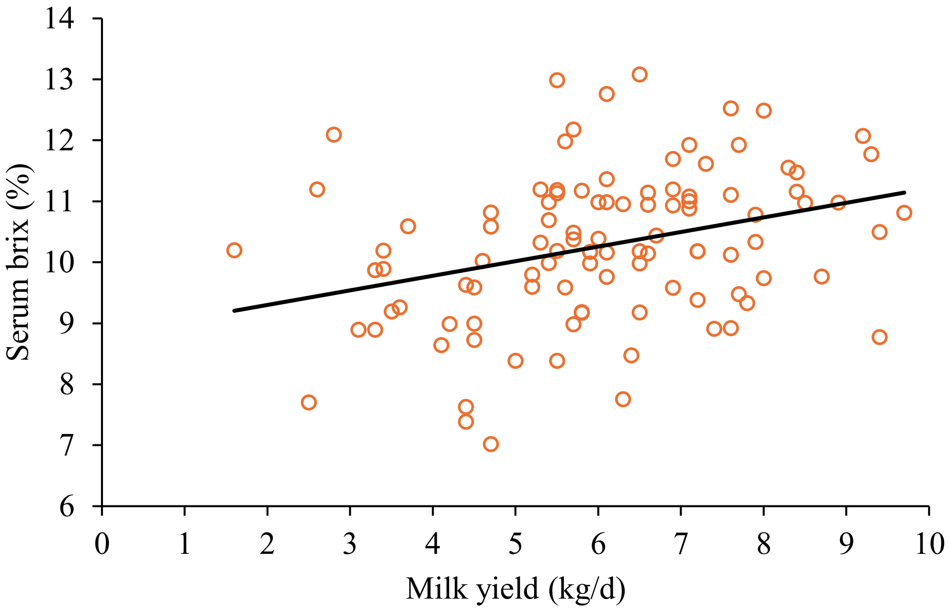

Figure 6. Relationship between milk yield of the dams and the serum brix of 1-day-old calves ($\hat{Y} = 8.82 \pm 0.460 + 0.24 \pm 0.078 \times X$; $s_{XY} = 1.192$; $r^2 = 0.108$; $n = 101$). The data points were adjusted for random study effects.