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VOLUNTARY INTAKE OF LACTATING BEEF COWS DURING PREGNANCY UNDER TROPICAL PASTURE AND EFFECTS OF DDG OR SUGAR CANE MOLASSES ON PERFORMANCE OF CALVES AND HEIFERS RESPECTIVELY.

Éllem Maria de Almeida Matos
Doctor Scientiae

VIÇOSA - MINAS GERAIS
2024

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Adviser: Sebastiao de C V Filho

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Assent:

Éllem Maria de Almeida Matos
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Sebastiao de Campos Valadares Filho

To God.

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“Se algum de vocês tem falta de sabedoria, peça-a a Deus, que a todos dá livremente, de boa vontade; e lhe será concedida”.

(Tiago 1:5)

ABSTRACT

MATOS, Éllem Maria de Almeida, D.Sc., Universidade Federal de Viçosa, August, 2024. **Voluntary intake of lactating beef cows during pregnancy under tropical pasture and effects of ddg or sugar cane molasses on performance of calves and heifers respectively.**. Adviser: Sebastiao de Campos Valadares Filho.

Our objectives in the current study were: 1) to understand the voluntary intake of beef cows during gestation under grazing in tropical pastures; 2) to evaluate the effects of DDG supplementation on performance and digestion characteristics of suckling beef calves on tropical pastures and 3) to evaluate whether the inclusion of molasses affect voluntary intake, performance, digestive characteristics, and rumen epithelium cell development in young beef finishing heifers. For the first objective, thirty-seven multiparous Nellore cows, along their respective calves, with an average initial body weight (BW) of 517 ±15.7 kg and initial body condition score (BCS) of 5.0, were used. All cows were randomly assigned into eight paddocks with approximately 7.0 ha each, covered evenly with Urochloa decumbens signal grass. Intake and digestibility trials were carried out at approximately 4 days before the insemination (this time point was considered day 0 of gestation) and, 30, 60, 90, 120, 150, 180, 210, 240, and 270 days of gestation. Fecal output was estimated using chromic oxide (Cr_2O_3) as an external marker. We detected a quadratic pattern ($P < 0.05$) between week of pregnancy (WP) and dry matter intake (DMI, g/kg BW).In conclusion, the voluntary intake can be adequately described as a function of the week of pregnancy according to the following model: $DMI = 20.978 \pm 0.9059 + 0.316 \pm 0.0947 \times WP – 0.0134 \pm 0.0023 \times WP^2$ For the second objective, thirty-seven Red Angus × Nellore male calves (162±4.1 kg BW; 120 days old) were used in this study. Cow-calf pairs were randomly assigned into eight paddocks with approximately 7.0 ha each. Four treatments were evaluated, as follows: control (mineral mixture only; n=8), supplement without DDG (n=10), or supplements including 375 (n = 9) and 750 (n = 10) g/kg DDG on a dry matter (DM) basis. The supplements were composed by ground corn, soybean meal, urea, and DDG, and formulated to contain 300 g CP/kg DM. Performance characteristics were evaluated by recording calves' BW at the beginning and end of the experiment without prior fasting. The ADG and FBW decreased (P = 0.04) linearly as the level of inclusion of DDG in the supplement increased. In conclusion, including 375 or 750 g DDG/kg DM in supplements decreases performance suckling beef calves in tropical pastures. For the third objective, twelve newly weaned Red Angus × Nellore heifers, averaging 238±5.14 kg, were used. The basal diet consisted of corn silage, sorghum silage, ground corn, soybean meal, urea, and mineral mixture, with a forage-to-concentrate ratio of 50:50. The following treatments were evaluated: control (no addition of powdered molasses) or addition of 50 g/kg of diet (DM basis) of powdered molasses. On day 55 of the experiment, a digestibility carried out over a period of 5 days. Briefly, feces samples were collected immediately after defecation on days 1, 2, 3, 4, and 5 of the trial, respectively and on days 3 and 5 of the digestibility trial, urine samples were collected. All heifers were weighed at the beginning and end of the experiment after 16-hour solids fasting. Subsequently, the heifers were slaughtered. There was no effect (P = 0.15) of molasses inclusion in either intake variables or fatty acids concentration. Providing sugarcane molasses does not affect the intake, digestion characteristics, and performance of young beef finishing heifers fed with medium-quality diets.

Keywords: beef cattle; by-products; digestion; forage intake.

RESUMO

MATOS, Éllem Maria de Almeida, D.Sc., Universidade Federal de Viçosa, agosto de 2024. **Consumo voluntário de vacas de corte em lactação durante a gestação em pastagens tropicais e efeitos do ddg ou melaço de cana-de-açúcar no desempenho de bezerros e novilhas, respetivamente.**. Orientador: Sebastiao de Campos Valadares Filho.

Os objetivos deste estudo foram: 1) compreender o consumo voluntário de vacas de corte durante a gestação sob pastejo em pastagens tropicais; 2) avaliar os efeitos da suplementação com DDG sobre o desempenho e as características de digestão de bezerros lactentes em pastagens tropicais e 3) avaliar o efeito da inclusão de melaço no consumo voluntário, desempenho, características digestivas e desenvolvimento das células do epitélio ruminal em novilhas de corte em terminação. Para o primeiro objetivo, trinta e sete vacas Nelore multíparas, junto com seus respectivos bezerros, com peso corporal (PC) inicial médio de 517±15,7 kg e escore de condição corporal (ECC) inicial de 5,0 foram utilizadas. Todas as vacas foram distribuídas aleatoriamente em oito piquetes com aproximadamente 7,0 ha cada, cobertos uniformemente com capim-braquiária (Urochloa decumbens). Ensaios de consumo e digestibilidade foram realizados aproximadamente 4 dias antes da inseminação (este ponto temporal foi considerado o dia 0 de gestação) e aos 30, 60, 90, 120, 150, 180, 210, 240 e 270 dias de gestação. A excreção fecal foi estimada usando óxido crômico (Cr_2O_3) como marcador externo. Detectamos um padrão quadrático ($P < 0,05$) entre a semana de gestação (SG) e o consumo de matéria seca (CMS, g/kg PC). Concluímos que o consumo voluntário pode ser descrito adequadamente em função da semana de gestação de acordo com o seguinte modelo: $CMS = 20,978 \pm 0,9059 + 0,316 \pm 0,0947 \times SG – 0,0134 \pm 0,0023 \times SG^2$. Para o segundo objetivo, trinta e sete bezerros machos Red Angus × Nelore (162±4,1 kg PC; 120 dias de idade) foram utilizados. Os pares vaca-bezerro foram distribuídos aleatoriamente em oito piquetes com aproximadamente 7,0 ha cada. Quatro tratamentos foram avaliados: controle (apenas mistura mineral; n = 8), suplemento sem DDG (n = 10), e suplementos com 375 (n = 9) e 750 (n = 10) g/kg de DDG na matéria seca (MS). Os suplementos foram compostos por milho moído, farelo de soja, ureia e DDG, formulados para conter 300 g de proteína bruta (PB)/kg MS. As características de desempenho foram avaliadas registrando o PC dos bezerros no início e no final do experimento, sem jejum prévio. O ganho médio diário (GMD) e o peso corporal final (PCF) diminuíram (P = 0,04) linearmente à medida que o nível de inclusão de DDG no suplemento aumentou. Concluímos que a inclusão de 375 ou 750 g de DDG/kg MS nos suplementos reduz o desempenho de bezerros lactentes em pastagens tropicais. Para o terceiro objetivo, doze novilhas recém-desmamadas Red Angus × Nelore, com peso médio de 238±5,14 kg, foram utilizadas. A dieta basal consistiu de silagem de milho, silagem de sorgo, milho moído, farelo de soja, ureia e mistura mineral, com uma relação volumoso de 50:50. Os seguintes tratamentos foram avaliados: controle (sem adição de melaço em pó) ou adição de 50 g/kg de dieta (base na MS) de melaço em pó. No 55º dia do experimento, foi realizado um ensaio de digestibilidade por um período de 5 dias. As amostras de fezes foram coletadas imediatamente após a defecação nos dias 1, 2, 3, 4 e 5 do ensaio, e, nos dias 3 e 5, amostras de urina também foram coletadas. Todas as novilhas foram pesadas no início e no final do experimento, após jejum sólido de 16 horas, e posteriormente abatidas. Não houve efeito (P = 0,15) da inclusão de melaço sobre as variáveis de consumo ou concentração de ácidos graxos. Concluímos que a oferta de melaço de cana não afeta o consumo, as características de digestão e o desempenho de novilhas em terminação alimentadas com dietas de qualidade média

Palavras-chave: bovinos de corte; coproduto; digestão; ingestão de forragem.

SUMMARY

INTRODUCTION

The use of cereal grains in animal feed originated from the need to supplement nutrients that pastures lacked, particularly during drought periods when the nutritional value of pasture significantly diminishes. Beyond supplementing grazing cattle, cereal grains are also extensively used in feedlot diets worldwide. This reliance on grains has intensified competition between the livestock industry and the human food market, driving prices higher.

In response to the need to reduce animal production's dependence on grains also essential for human consumption, feeding systems globally have turned to research on the use of locally available agro-industrial byproducts in ruminant diets. The remarkable ability of the ruminal microbial ecosystem to break down fibrous carbohydrates and synthesize high-quality protein from low-value protein sources and non-protein nitrogen has facilitated the increased use of alternative feeds, commonly referred to as byproducts (Oliveira et al., 2012). By-products are derived from various stages of industrial processes, where the primary product is refined and the residual materials are repurposed. In many cases, byproducts have found significant applications in the animal feed industry such as characteristics that enhance diet formulation (e.g., high protein content) and offer health and performance benefits to animals. Among the most used byproducts in ruminant nutrition are dried distillers grains and sugarcane molasses.

Dried distillers grains (DDG) have been used extensively in diets of growing and finishing cattle due to their high energy and rumen undegradable protein content (RUP; Martin et al., 2007; Kopflestein et al., 2008; Buttrey et al.,2012). Overall, DDG shows significant variation in chemical composition, primarily due to differences in the type of grain and processing methods used in the ethanol production (Boever et al., 2014; Nkomba et al., 2016; Böttger et al., 2018). This variability is a key factor contributing to the wide range of performance outcomes observed in cattle fed diets containing DDG (Klopfenstein et al., 2008; Griffin et al., 2012; Torres et al., 2022).

In tropical conditions, DDG has been utilized in cattle supplements as a strategy to increase the flow of amino acids to the intestine, thereby increasing the efficiency of nitrogen (N) utilization in cattle under grazing in tropical pastures (Ferrari et al., 2021). However, most research on the addition of DDG in cattle diets has focused on growing and finishing beef cattle. To the best of our knowledge, no studies have investigated the use of DDG in supplements for suckling beef calves in pastures. Consequently, the effects of DDG on the performance and digestion characteristics of suckling beef calves remain to be understood.

Molasses is a widely used feed in the diets of production animals, especially during early life in order to stimulate feed intake. In cattle, the use of molasses has benefits related to improved voluntary intake and increased production of volatile fatty acid (VFA), especially butyrate (Brown et al., 1987; Lesmeister, et al., 2005). Butyrate is closely related to gastrointestinal tract (GIT) health, as it contributes to the development and maintenance of GIT integrity (Górka, et al., 2017). In theory, this could increase the absorption area of the ruminal epithelium and improve the utilization of diet components, resulting in enhanced voluntary intake and immune response capacity in stressful situations, including the weaning.

Additionally, incorporating molasses into the diets of finishing cattle can enhance immune system resilience, making the animals less susceptible to potential challenges upon arrival at feedlot, such as immune system decline, respiratory diseases, and ruminal environment imbalances. Furthermore, the inclusion of molasses can help stabilize feed intake throughout the entire feedlot period, thereby ensuring consistently satisfactory production levels (Reinhardt et al., 2015; Kos et al., 2023).

However, the key to meeting nutrient requirements and formulating effective diets is accurately predicting the animals' actual dry matter intake, which can vary based on the characteristics of the feed and the animals' physiological state. Predicting voluntary intake of ruminant animals is quite challenging due to the multifactorial nature of intake control (Forbes, 2007). This complexity is heightened in pregnant beef cows, as pregnancy induces several changes. For instance, physical, physiological, metabolic, and behavioral alterations can influence the voluntary intake of pregnant cows, making it difficult to incorporate all these factors into predictive equations (Gionbelli et al., 2023).

Furthermore, most research aimed at understanding and/or predicting the voluntary intake of pregnant beef cows has focused on Bos taurus cows mostly from pen-fed animals (Stanley et al., 1993; Ingvartsen, 1994; Coleman et al., 2014; Linder et al., 2014). Conversely, there is a lack of data in both national and international literature on the voluntary intake of beef cows during gestation under grazing in tropical pastures. Most studies are carried out at specific time points during gestation, failing to account for variations throughout the cow's production cycle (Marquez et al., 2017; Ferreira et al., 2020; David et al., 2024). This data gap is primarily attributed to the difficulty in measuring the intake of ruminants under grazing conditions (Coleman et al., 2014). Moreover, due to the intensive management required during intake trials (e.g., restraint, marker infusion, sample collection), special care must be taken with pregnant cows to prevent any productive losses, such as difficulty conceiving and abortions. As a result, the variation in voluntary intake throughout gestation in beef cows and the factors affecting it still need to be understood.

Hence, the objectives in the current study were: 1) to understand the voluntary intake of beef cows during gestation under grazing in tropical pastures.

2) to evaluate the effects of DDG supplementation on performance and digestion characteristics of suckling beef calves on tropical pastures; 3) to evaluate whether the inclusion of molasses affect voluntary intake, performance, digestive characteristics, and rumen development in young beef finishing heifers.

REFERENCE

Brown, W. F.; Johnson, D. D. 1991. Effects of energy and protein supplementation of ammoniated tropical grass hay on the growth and carcass characteristics of cull cows. J. Anim. Sci., 69, 348-357.

Boever, J.A., Blok, M.C., Millet, S., Vanacker, J., De Campeneere, S., 2014. The energy and protein value of wheat, maize and blend DDGS for cattle and evaluation of prediction methods. Animal, 8, 1839-1850.

Böttger, C., Südekum, K.H., 2018. Protein value of distillers dried grains with solubles (DDGS) in animal nutrition as affected by the ethanol production process. Anim. Feed Sci. Technol. 244, 11-17.

Buttrey, E.K., McCollum III, F.T., Jenkins, K.H., Patterson, J.M., Clark, B.E., Luebbe, M.K., Lawrence, T.E., MacDonald, J.C. 2012. Use of dried distillers grains throughout a beef production system: Effects on stocker and finishing performance, carcass characteristics, and fatty acid composition of beef. J. Anim. Sci. 90, 2381-2393.

Coleman, S.W., Gunter, S.A., Sprinkle, J.E., Neel, J.P. 2014. Difficulties associated with predicting forage intake by grazing beef cows. J Anim Sci. 7, 2775-2784.

David, G.S.S., Matos, E.M.A., Domingos, B.R., Ebani, Y.C., Sousa, L.C.O., Leite, G.D.O., Pereira, P.H.B., Renno, L.N., Lopes, S.A., Valadares Filho, S.C., Paulino, M.F. 2024. Periparturient Changes in Voluntary Intake, Digestibility, and Performance of Grazing Zebu Beef Cows with or without Protein Supplementation. Animals. 14, 1710.

Ferrari, A.C., Leite, R.G., Fonseca, N.V., Romanzini, E.P., Cardoso, A.D.S., Barbero, R.P., Costa, D.F., Ruggieri, A.C., Reis, R.A. 2021. Performance, nutrient use, and methanogenesis of Nellore cattle on a continuous grazing system of Urochloa brizantha and fed supplement types varying on protein and energy sources. Livest. Sci., 253, 104716. https://doi.org/10.1016/j.livsci.2021.104716

Ferreira M.F.L., Renno, L.N., Detmann, E., Paulino, M.F., Valadares Filho, S.C., Moreira, S.S., Martins, H.C., Oliveira, B.I.C., Marquez, J.A., Cidrine, I.P. 2020. Performance, metabolic and hormonal responses of grazing Nellore cows to an energy-protein supplementation during the pre-partum phase. BMC Vet Res. 1, 108.

Forbes, J.M. 2007. Voluntary Food Intake and Diet Selection in Farm Animals. Second edition. CABI Publishing, Wallingford, UK.

Gionbelli, M.P., Valadares Filho, S.C., Souza, M.D. Nutritional requirements for pregnant and non-pregnant beef cows. 2023. In: Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Suprema Grafica Ltda, Visconde do Rio Branco, 299-328.

Górka, P., B. Śliwiński, J., Flaga, J., Wieczorek, M.M., Godlewski, E., Wierzchoś, R., Zabielski., Kowalski, Z.M. 2017. Effect of butyrate infusion into the rumen on butyrate flow to the duodenum, selected gene expression in the duodenum epithelium, and nutrient digestion in sheep. J. Anim. Sci. 95, 2144-2155.

Griffin, W.A., Bremer, V.R., Klopfenstein, T.J., Stalker, L.A., Lomas, L.W., Moyer, J.L. and Erickson, G.E. 2012. A meta-analysis evaluation of supplementing dried distillers grains plus solubles to cattle consuming forage-based diets. Prof. Anim. Sci. 28, 306-312.

Ingvartsen, L.K. 1994. Models of voluntary food intake in cattle. Livest. Prod. Sci. 39, 19-39.

Klopfenstein, T.J., Erickson, G.E. and Bremer, V.R., 2008. Board-invited review: Use of distillers by-products in the beef cattle feeding industry. J. Anim Sci., 86, 1223-1231. https://doi.org/10.2527/jas.2007-0550

Kos, D., Schreiner, B., Thiessen, S., McAllister, T., Jelinski, M., Ruzzini, A. 2023. Insight into antimicrobial resistance at a new beef cattle feedlot in western Canada. Msphere. 8, 00317-23.

Lesmeister, K. E.; Heinrichs, A. J. 2005. Effects of adding extra molasses to a texturized calf starter on rumen development, growth characteristics, and blood parameters in neonatal dairy calves. J. Dairy Sci. 88, 411-418.

Linder, D.R., Tigemeyer, E.C., Olson, K.C., Anderson, D.E. Effects of gestation and lactation on forage intake, digestion, and passage rates of primiparous beef heifers and multiparous beef cows. J. Anim. Sci. 2014, 92, 2141–215.

Martin, J.L., Cupp, A.S., Rasby, R.J., Hall, Z.C., Funston, R.N. 2007. Utilization of dried distillers grains for developing beef heifers. J. Anim Sci. 85, 2298-2303. https://doi.org/10.2527/jas.2007-0076.

Marquez, D.C., Paulino, M.F., Renno, L.N., Villadiego, F.C., Ortega, R.M., Moreno, D.S., Martins, L.S., De Almeida, D.M., Gionbelli, M.P., Manso, M.R., Melo, L.P., Moura, F.H., Duarte, M.S., 2017. Supplementation of grazing beef cows during gestation as a strategy to improve skeletal muscle development of the offspring. Animal, 11, 2184-2192.

Nkomba, E. Y., E. Van Rensburg, A. F. A. Chimphango., J. F. Gorgens. 2016. The influence of sorghum grain decortication on bioethanol production and quality of the distillers dried grains with solubles using cold and conventional warm starch processing. Bioresour. Technol. 203, 181–189. https://doi.org/10.1016/j.biortech.2015.12.045

Oliveira, A.S., Rodrigues, M., Oliveira, C., De Souza, M.C., De Moura, D.C., De Souza, J.G., 2012. Utilização de coprodutos agroindustriais na alimentação de bovinos. In: Anais do XI Congresso sobre Manejo e Nutrição de Bovinos CBNA, Campinas, SP.

Reinhardt, C., Thomson, D.U. 2015. Nutrition of newly received feedlot cattle. Vet. Clinics: Food Anim. Pract. 31, 283-294.

Stanley, T.A., Cochran, R.C., Vanzant, E.S., Harmon, D., Corah, L. 1993. Periparturient changer in intake, ruminal capacity, and digestive characteristics in beef cows consuming alfalfa hay. J. Anim. Sci. 71, 788-795.

Torres, R.N.S.; Coelho, L.M., Paschoaloto, J.R., Ghedini, C.P., Neto, O.R.M., Chardulo, L.A.L., Almeida, M.T.C. 2022. Does distillers' grains supplementation affect beef cattle performance, carcass characteristics, and meat quality? A meta-analysis study. Res. Vet. Sci. 149, 21–35. https://doi.org/10.1016/j.rvsc.2022.06.004

CHAPTER 1

Understanding the voluntary intake of beef cows during gestation under grazing in tropical pastures

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ABSTRACT

Our objective was to understand and predict the voluntary intake of beef cows during gestation under grazing in tropical pastures. Thirty-seven multiparous Nellore cows, along their respective calves, with an average initial body weight (BW) of 517 ±15.7 kg, were used. Following pregnancy diagnosis, twenty-five got pregnant and were used in the subsequent intake trials. All cows were randomly assigned into eight paddocks with approximately 7.0 ha each, covered evenly with *Urochloa decumbens* signal grass. Intake and digestibility trials were carried out at approximately 4 days before the insemination, 30, 60, 90, 120, 150, 180, 210, 240, and 270 days of gestation. In order to understand dry matter intake (DMI) as gestation advances, we used a mixed linear regression approach. Models were adjusted considering the random effect of cow on the model parameters. We detected a quadratic pattern ($P < 0.05$) between week of pregnancy (WP) and DMI (g/kg BW), as described by the following model: $DMI = 20.978 \pm 0.9059 + 0.316 \pm 0.0947 \times WP - 0.0134 \pm 0.0023 \times WP^2$. According to the adjusted model, the DMI would be maximized at approximately 12 weeks of gestation, which would result in an estimated intake of 22.8 g/kg BW. Beyond 12 weeks of gestation, DMI progressively decreases as gestation advances. The cross-validation demonstrated that the model accurately predicted DMI, as the intercept and slope did not significantly differ from 0 and 1, respectively ($P \geq 0.78$). However, the model displayed relatively low precision, as evidenced by the low values of R^2 (0.333) and ρ (0.500). While there appears to be a direct effect of gestation on DMI, particularly in the late gestation, the inclusion of the week of gestation in the model accounted for variations in the chemical composition of the forage. Variations in forage composition appears to be the largest factor affecting voluntary intake. This finding reinforces the applicability of the model for predicting voluntary intake of pregnant beef cows in tropical production systems.

Keywords: Beef Cattle; Pregnancy; Grazing; Intake

INTRODUCTION

Predicting voluntary intake of ruminant animals is quite challenging due to the multifactorial nature of intake control (Forbes, 2007). This complexity is heightened in pregnant beef cows, as pregnancy induces several changes. For instance, physical, physiological, metabolic, and behavioral alterations can influence the voluntary intake of pregnant cows, making it difficult to incorporate all these factors into predictive equations (Gionbelli et al., 2023).

Furthermore, most research aimed at understanding and/or predicting the voluntary intake of pregnant beef cows has focused on *Bos taurus* cows mostly from pen-fed animals (Stanley et al., 1993; Ingvartsen, 1994; Coleman et al., 2014; Linder et al., 2014). Two main problems may arise regarding the applicability of these results in tropical conditions. First, physiological and metabolic differences between *Bos indicus* and *Bos taurus* cows are well-documented (Cardenas-Medina et al., 2010; Lopes et al., 2019; 2021). Such discrepancies could compromise the accuracy of these models in predicting the voluntary intake of Zebu cows. Additionally, models developed for fed-pen cows often fail to account for variations in voluntary intake throughout the cow's production cycle, including changes due to lactation and gestation, as well as differences in forage quality and availability. Consequently, the application of these models results in unrealistic predictions when applied to grazing beef cows (Coleman et al., 2014).

Conversely, there is a lack of data in both national and international literature on the voluntary intake of beef cows during gestation under grazing in tropical pastures. Most studies are carried out at specific time points during gestation, failing to account for variations throughout the cow's production cycle (Marquez et al., 2017; Ferreira et al., 2020; David et al., 2024). This data gap is primarily attributed to the difficulty in measuring the intake of ruminants under grazing conditions (Coleman et al., 2014). Moreover, due to the intensive management required during intake trials (e.g., restraint, marker infusion, sample collection), special care must be taken with pregnant cows to prevent any productive losses, such as difficulty conceiving and abortions. As a result, the variation in voluntary intake throughout gestation in beef cows and the factors affecting it still need to be understood. We hypothesized that dry matter intake of pregnant lactating cows can be predicted using weeks of lactation as the independent variable. .Hence, our objective was to understand and predict the voluntary intake of beef cows during gestation under grazing in tropical pastures.

MATERIAL AND METHODS

Ethical considerations, animals and experimental procedures

This study was carried out at the Extension, Teaching, and Research Unit in Beef Cattle at the Department of Animal Science of the Universidade Federal de Viçosa, Viçosa, MG, Brazil. All animal care and handling procedures followed protocols and standards approved by the Animal Care and Use Committee of the Universidade Federal de Viçosa (protocol 37-2023).

Thirty-seven multiparous Nellore cows, along their respective calves, with an average initial body weight (BW) of 517 ±15.7 kg and initial body condition score (BCS) of 5.0, were used. At approximately 66 days after parturition, cows were submitted to a fixed-time artificial insemination protocol using semen from the same sire in two attempts. Pregnancy diagnosis was carried out 30 days after insemination. Following pregnancy diagnosis, twenty-five got pregnant and were used in the subsequent intake trials.

All cows were randomly assigned into eight paddocks with approximately 7.0 ha each, covered evenly with *Urochloa decumbens* signal grass. Paddocks were equipped with waterers and covered feeders. Cows had access *ad libitum* to fresh water and mineral mixture throughout the experiment. To minimize any likely confounding effects between treatments and paddocks, cow-calf pairs were rotated among paddocks every 15 days, ensuring that each cow-calf pair spent equal time in each paddock and passed through all paddocks the same number of times.

Sampling and measurement

Intake and digestibility trials were carried out at approximately 4 days before the insemination (we considered this time-point as day 0 of gestation), 30, 60, 90, 120, 150, 180, 210, 240, and 270 days of gestation. To facilitate the modeling process, we subsequently transform these time-points into weeks of gestation.

Fecal output was estimated using chromic oxide (Cr_2O_3) as an external marker. Each trial lasted nine days, with five days to stabilize the marker’s fecal excretion and four days for sample collection (Sampaio et al., 2011). From the first to the eighth day of each trial, approximately 15 g of Cr_2O_3 was administered daily at 1000 h via the esophagus using a metal probe. Feces samples were collected immediately after defecation or directly from the rectum of the cows. Feces collections were carried out at 1800, 1400, 1000, and 0600 h on days 6, 7, 8 and 9 of each trial, respectively (Sampaio et al., 2011). On the ninth day of each trial, forage samples were taken from each paddock by hand-plucking method (De Vries, 1995).

Forage and feces samples were oven-dried at 55°C for 72 h and subsequently ground to pass through a 1-mm screen sieve. Feces samples were pooled per cow and trial (i.e., per each time-point relative to the week of gestation). Forage samples were pooled per paddock and trial. All samples were subsequently stored in plastic jars for further analysis.

At the end of each trial, all cows were weighed without prior fasting measure dry matter intake relative to average BW.

Laboratory Analysis

Samples of forage and feces were analyzed for DM (dried for 16 h at 105 ºC, method G-003/1), CP (Kjeldahl procedure, method N-001/2), and NDF (using heat-stable α-amylase and omitting sodium sulfite, method F-012/1), following the procedures described by the Brazilian National Institute of Science and Technology in Animal Science (INCT-CA, Detmann et al., 2021). The content of NDF was expressed with correction for contaminant ash and protein (apNDF). The iNDF content was obtained through a 288-h *in situ* incubation procedure (Valente et al., 2011). Fecal chromium concentration was analyzed using atomic absorption spectrophotometry (method M-005/2; Detmann et al., 2021).

Fecal output was obtained as the ratio between the quantity of Cr_2O_3 offered daily and the Cr_2O_3 concentration in feces, as follows:

$$FO = \frac{Cr_{prov.}}{Cr_{feces}} \tag{1}$$

Where FO is fecal output (g/day), Cr_{prov} is the quantity of Cr provided (g/day), and Cr_{feces} is the Cr concentration in the feces (g/g).

Forage intake was estimated using the indigestible neutral detergent fiber (iNDF) as an internal marker, as follows:

$$FI = \frac{\text{FO} \times iNDF_{feces}}{iNDF_{for}} \tag{2}$$

Where FI is the forage dry matter intake (kg/day), FO is the fecal output (kg/day), $iNDF_{feces}$ is the iNDF concentration in the feces (kg/kg DM), and $iNDF_{for}$ is the iNDF concentration in the forage (kg/kg DM).

Statistical analysis

Modeling process

Firstly, we performed an exploratory analysis to investigate the relationship between DMI and explanatory variables using graphical analyses and Pearson correlations. This analysis revealed that DMI was correlated with both week of gestation and forage quality characteristics. Furthermore, we found correlations between forage characteristics and week of gestation.

Based on these initial assessments, we decided to model DMI as a function of the week of gestation for several reasons. Firstly, this approach helps to avoid multicollinearity issues between the week of gestation and forage characteristics. Secondly, incorporating the week of gestation in the model accounts, at least partially, for variations in forage composition. Finally, information regarding forage composition is often not available in practical situations, which would limit the proposed model's applicability for prediction purposes. In order to understand DMI as gestation advances, we used a mixed linear regression approach. Models were adjusted considering the random effect of cow on the model parameters. The choice of the best (co)variance structure was based on the lowest value of the Akaike's information criterion with correction (AICC). All covariance parameters were estimated by the restricted maximum likelihood method and the degrees of freedom were estimated by the Kenward-Roger method.

The model including a quadratic term for the week of gestation provided a better fit compared to the model with only a linear term ($AICC_{linear}$ = 1342; $AICC_{quadratic}$ = 1324). All analyses were performed using the procedure MIXED in SAS 9.4. Significance for both fixed and random effects was declared at P<0.05.

Cross-validation

A leave-one-out cross-validation technique was employed to assess the adequacy of the proposed model. Briefly, each subject (i.e., cow) was sequentially excluded from the dataset, and the model was re-fitted to the remaining data. This process was repeated for each cow, ensuring that each one was left out once.

The actual and predicted DMI values were compared using a linear regression, as follows:

$$Y = \beta_o + \beta_1 \times X \quad (3)$$

Where Y is the actual DMI values; X is the predicted DMI values; β_o is the intercept; β_1 is the slope of the relationship between $\hat{Y}$ and X. The following hypothesis were evaluated (Neter et al., 1996):

$$H_0: \beta_o = 0 \qquad H_a: B_0 \neq 0 \quad (4)$$

$$H_0: \beta_1 = 1 \qquad H_a: \beta_1 \neq 1 \quad (5)$$

Accuracy and precision measures of the proposed model were obtained using mean square error of prediction (MSEP), concordance correlation coefficient (CCC), and coefficient of determination (R^2). The MSEP was decomposed into its components (Bibby and Toutenburg, 1977), as follows:

$$MSEP = MB + SB + RE = 1/n\sum_{i=1} (X_i - Y_i)^2 \quad (6)$$

$$MB = (\bar{X} - \bar{Y})^2 \quad (7)$$

$$SB = (s_X - rs_Y)^2 \quad (8)$$

$$RE = (1 - r^2)s^2_Y \quad (9)$$

Where X are the predicted DMI values; Y are the actual DMI values; MSEP is the mean squared error of prediction; MB is the mean bias (i.e., central tendency of deviation); SB is the systematic bias (i.e., deviation of the slope from 1); and RE is the random error (i.e., variation that is not explained by the regression); s_X and s_Y are the standard deviations of predicted and actual DMI values, respectively; s^2_Y is the variance of actual DMI values; r is the linear correlation coefficient between predicted and actual DMI values.

Furthermore, the concordance correlation coefficient (CCC) was decomposed into two components: the correlation coefficient estimate (ρ), which estimates model precision, and the bias correction factor (Cb), which estimates model accuracy. Both CCC, ρ, and Cb values range from 0 to 1, with values closer to 1 indicating more precise and accurate models (Tedeschi, 2006).

The adequacy evaluation analyses of the proposed model were performed using the Model Evaluation System (MES), version 3.2.7 (Tedeschi, 2006).

RESULTS

Dataset description

Descriptive statistics of the dataset are presented in Table 1. The dataset comprised 223 observations from twenty-five cows, with an average BW of 525 kg (±61.9 kg). The average DMI was 10.6 kg/day (±2.97 kg/day) and 20.2 g/kg BW (±5.52 g/kg BW). The average DMI during early (0-12 weeks), mid (16-25 weeks), and late (30-38 weeks) gestation was 21.8, 22.3, and 15.9 g/kg BW, respectively (Figure 1).

Likewise, the forage chemical composition showed considerable variation throughout the experiment (Figure 2). At the beginning of the experiment (week 0 of pregnancy), the forage had high quality, indicated by its high crude protein (CP) and low indigestible neutral detergent fiber (INDF) contents. However, forage quality tended to decline over the course of the experiment, reaching its lowest nutritional quality in late gestation, as evidenced by the decreased CP and increased INDF contents.

Correlations between DMI and explanatory variables

There was linear correlation among all variables ($P < 0.05$; Figure 2). Both DMI in kg/day and g/kg BW were negatively correlated ($P < 0.001$) with the week of pregnancy (r = -0.328 and r = -0.426, respectively). Similarly, DMI in g/kg BW was negatively correlated ($P < 0.001$) with both apFNDF (r = -0.286) and iFNDF (r = -0.537). Conversely, a positive correlation was found between DMI in g/kg BW and FCP (r = 0.443, $P < 0.001$). A similar pattern of correlations was observed for DMI in kg/day. The BW was positively correlated with DMI in kg/day (r = 0.237, $P < 0.001$) and negatively correlated with DMI in g/kg BW (r = -0.182, $P < 0.01$).

Additionally, forage composition characteristics were correlated with the week of pregnancy. A positive correlation ($P < 0.001$) was found between the week of pregnancy and both apFNDF (r = 0.765) and iFNDF (r = 0.878). Conversely, FCP was negatively correlated ($P < 0.001$) with the week of pregnancy (r = -0.738).

Relationship between DMI and week of gestation

We detected a quadratic pattern ($P < 0.05$; Figure 3) between week of pregnancy (WP) and dry matter intake (DMI, g/kg BW), as described by the following model:

$$DMI = 20.978_{\pm 0.9059} + 0.316_{\pm 0.0947} \times WP - 0.0134_{\pm 0.0023} \times WP^2$$

According to the adjusted model, the DMI would be maximized at approximately 11.8 weeks of gestation, which would result in an estimated intake of 22.8 g/kg BW. Beyond 11.8 weeks of gestation, DMI progressively decreases as gestation advances.

Adequacy model evaluation

The leave-one-out cross-validation demonstrated that the model accurately predicted DMI, as the intercept and slope did not significantly differ from 0 and 1, respectively ($P \geq 0.78$; Table 2). This was further confirmed by the decomposition of both MSEP and CCC. The decomposition of MSEP revealed that the prediction error due to the central tendency of deviation and deviation of the slope from 1 was negligible (0% and 0.032%, respectively). Most of the prediction error (99.968%) was attributed to random errors (i.e., errors not explained by the regression). The model also exhibited a Cb of 0.873, indicating high accuracy in predicting DMI. However, the model displayed relatively low precision, as evidenced by the low values of R^2 (0.333) and ρ (0.500).

DISCUSSION

Predicting the voluntary intake of pregnant beef cows is quite challenging due to numerous factors that are often not considered in models for other animal categories, including calf weight, hormonal regulation of pregnancy, and homeorhetic mechanisms (Gioenbelli et al., 2023). In the current study, we found that voluntary intake exhibited a quadratic relationship with the weeks of pregnancy. Analyzing the partial derivatives of the adjusted model, we observed that DMI increased by approximately 0.022 g DM/kg BW per day during the first 11 weeks of pregnancy. However, reductions in DMI were expected from 84 days of pregnancy (around 12 weeks), with a progressive decrease from that point onward. For instance, the model indicated an average decrease of approximately 0.084 g DM/kg BW per day during late gestation. Therefore, our data suggest that the reduction in DMI was most pronounced in late gestation, showing a 28% decrease compared to mid gestation.

Several authors have reported direct effects of pregnancy on voluntary intake (Linden et al., 2014; Rotta et al., 2015; Gionbelli et al., 2024). These effects are generally attributed to hormonal changes induced by pregnancy and, primarily, to the ruminal repletion effect caused by the increase in gestational tissues (e.g., gravid uterus) and fetal growth (Forbes, 2007). Research conducted in tropical regions has indicated that pregnant cows experience a decrease in ruminal volume due to compression by the gravid uterus, with this effect being more pronounced in late gestation (Moreira et al., 2023). These factors could partially explain, the marked reduction in DMI observed in late gestation.

Some authors have attempted to account for the effect of gestation week on the DMI of pregnant beef cows (Ingvartsen, 1992). For example, the current edition of the Brazilian system of Nutrient Requirement of Zebu and Crossbred Cattle (BR-CORTE, 2016) accounts for a linear reduction of 0.02 g DM/kg BW per day starting from 135 days of pregnancy for *Bos indicus* beef cows (Gionbelli et al., 2023). However, the model proposed by Gionbelli et al. (2023) has a major limitation: it was developed using non-lactating cows in pens, all receiving the same diet. Variations throughout the cow's production cycle, including physiological changes due to gestation and lactation, and fluctuations in forage quality, can directly impact the voluntary intake of grazing beef cows, often making predictions unrealistic (Coleman et al., 2014). Among the many influencing factors, the variation in forage quality and availability throughout the cow's production cycle should be particularly highlighted.

In the current study, we observed a significant decrease in forage quality throughout the weeks of pregnancy, as indicated by the increase iNDF and decreased FCP contents, particularly during the mid to late gestation. This trend is characteristic of cow-calf systems, as the breeding season is planned for cows to calve during the rainy season. Consequently, cows spend the mid to late gestation on lower-quality pastures.

Several studies have consistently demonstrated that forage quality, as indicated by its iNDF and CP contents, significantly affects the voluntary intake of grazing cattle (Detmann et al., 2014; Fernandes et al., 2021). Our data further support this, showing that voluntary intake was directly influenced by variations in forage quality throughout gestation. This relationship is evident in two keyways. First, the linear correlation analysis revealed a strong association between DMI and both CP and iNDF levels. This result indicates that forage quality varied sufficiently throughout the experiment to induce changes in DMI. Second, the decrease in DMI starting from 12 weeks of pregnancy coincided with a decline in pasture quality. Taken together, these findings suggest that forage quality directly affected DMI. Indeed, several authors have reported the impact of variations in forage quality on the DMI of beef cows grazing in both tropical (David et al., 2024) and non-tropical regions (Coleman et al., 2014).

CONCLUSION

We concluded that the voluntary intake can be adequately described as a function of the week of pregnancy according to the following model:

$DMI = 20.978_{\pm 0.9059} + 0.316_{\pm 0.0947} \times WP - 0.0134_{\pm 0.0023} \times WP^2$

While there appears to be a direct effect of gestation on voluntary intake, particularly in the late gestation, the inclusion of the week of gestation in the model accounted for variations in the chemical composition of the forage. Variations in forage composition appears to be the largest factor affecting voluntary intake. This finding reinforces the applicability of the model for predicting voluntary intake of pregnant beef cows in tropical production systems.

REFERENCES

Bibby, J., Toutenburg, H., 1977. Prediction and improved estimation in linear models. John Wiley &Sons, Berlin, Germany.

Cárdenas-Medina, V., Ku-Vera, J.C., Magaña-Monforte, J.G. Estimation of Metabolizable Energy Requirements for Maintenance and Energetic Efficiency of Weight Gain in Bos taurus and Bos indicus Cows in Tropical Mexico. J. Anim. Vet. Adv. 9, 421-428.

Coleman, S.W., Gunter, S.A., Sprinkle, J.E., Neel, J.P. 2014. Difficulties associated with predicting forage intake by grazing beef cows. J Anim Sci. 7, 2775-2784.

David, G.S.S., Matos, E.M.A., Domingos, B.R., Ebani, Y.C., Sousa, L.C.O., Leite, G.D.O., Pereira, P.H.B., Renno, L.N., Lopes, S.A., Valadares Filho, S.C., Paulino, M.F. 2024. Periparturient Changes in Voluntary Intake, Digestibility, and Performance of Grazing Zebu Beef Cows with or without Protein Supplementation. Animals. 14, 1710.

Detmann, E., Costa e Silva, L.F., Rocha, G.C., Palma, M.N.N., Rodrigues, J.P.P. 2021. Métodos para análise de alimentos. Second edition. Suprema, Visconde do Rio Branco, Minas Gerais.

Detmann, E., Valente, E.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. Livest. Sci. 162, 141-153.

De Vries, M.F.W. 1995. Estimating forage intake and quality in grazing cattle: A reconsideration of the hand-plucking method. J. Range Manag. 48, 370-375.

Ferreira M.F.L., Renno, L.N., Detmann, E., Paulino, M.F., Valadares Filho, S.C., Moreira, S.S., Martins, H.C., Oliveira, B.I.C., Marquez, J.A., Cidrine, I.P. 2020. Performance, metabolic and hormonal responses of grazing Nellore cows to an energy-protein supplementation during the pre-partum phase. BMC Vet Res. 1, 108.

Fernandes G.A., de Oliveira A.S., de Araújo, C.V., Couto, V.R.M., de Moraes, K.A.K., de Moraes, E.H.B.K. 2021. Prediction of pasture intake by beef cattle in tropical conditions. Trop. Anim. Health. Prod. 2021.

Fontes, P., Oosthuizen, N., Ciriaco, F., Sanford, C., Canal, L., Cooke, R., Pohler, K., Henry, D., Mercadante, V., Ealy, A.D., Johnson, S.E., DiLorenzo, N., Lamb, G.C. 2021. Effects of nutrient restriction on the metabolic profile of Bos indicus-influenced and B. taurus suckled beef cows. Animal. 3, 100166.

Fontes, P.L.P., Oosthuizen, N., Ciriaco, F.M., Sanford, C.D., Canal, L.B., Pohler, K.G., Henry, D. D., Mercadante, V.R.G., Timlin, C.L., Ealy, A.D., Johnson, S.E., Dilorenzo, N., Lamb, G.C., 2019. Impact of fetal vs. maternal contributions of Bos indicus and Bos taurus genetics on embryonic and fetal development. J. Anim. Sci. 97, 1645-1655.

Forbes, J.M. 2007. Voluntary Food Intake and Diet Selection in Farm Animals. Second edition. CABI Publishing, Wallingford, UK.

Gionbelli, M.P., Duarte, M.S., Valadares Filho, S.C., Gionbelli, T.R.S., Ramirez-Zamudio, G.D., Silva, L.H.P., Nascimento, K.B., Costa, T.C. 2024. Efect of pregnancy and feeding level on voluntary intake, digestion, and microbial nitrogen synthesis in Zebu beef cows. Trop. Anim. Health. Prod. 56, 41.

Gionbelli, M.P., Valadares Filho, S.C., Souza, M.D. Nutritional requirements for pregnant and non-pregnant beef cows. 2023. In: Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Suprema Grafica Ltda, Visconde do Rio Branco, 299-328.

Stanley, T.A., Cochran, R.C., Vanzant, E.S., Harmon, D., Corah, L. 1993. Periparturient changer in intake, ruminal capacity, and digestive characteristics in beef cows consuming alfalfa hay. J. Anim. Sci. 71, 788-795.

Gionbelli, M.P., Valadares Filho, S.C., Souza, M.D. Nutritional requirements for pregnant and non-pregnant beef cows. 2023. In: Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Suprema Grafica Ltda, Visconde do Rio Branco, 299-328.

Ingvartsen, L.K. 1994. Models of voluntary food intake in cattle. Livest. Prod. Sci. 39, 19-39.

Johnson, W.L., Trimberger, G.W., Wright, M.J., Van Vleck, L.D. and Henderson, C.R. 1966. Voluntary intake of forage by Holstein cows as influenced by lactation, gestation, body weight and frequency of feeding. J. Dairy Sci. 49, 856-864.

Linder, D.R., Tigemeyer, E.C., Olson, K.C., Anderson, D.E. Effects of gestation and lactation on forage intake, digestion, and passage rates of primiparous beef heifers and multiparous beef cows. J. Anim. Sci. 2014, 92, 2141–215.

Marquez, D.C., Paulino, M.F., Renno, L.N., Villadiego, F.C., Ortega, R.M., Moreno, D.S., Martins, L.S., De Almeida, D.M., Gionbelli, M.P., Manso, M.R., Melo, L.P., Moura, F.H., Duarte, M.S., 2017. Supplementation of grazing beef cows during gestation as a strategy to improve skeletal muscle development of the offspring. Animal, 11, 2184-2192.

Moreira, G.M., Aguiar, G.L., Meneses, J.A.M., Nascimento, K.B., Ramirez-Zamudio, G.D., Costa T.C., Duarte M.S., Gionbelli, M.P. 2023. Pregnancy afects maternal performance, feed intake, and digestion kinetics parameters in beef heifers. J. Anim. Sci.

Rotta, P.P., Valadares Filho, S.C., Gionbelli, T.R.S., Costa e Silva, L.F., Engle, T.E., Marcondes, M.I., Machado, F.S., Villadiego, F.A.C., Silva, L.H.R. 2015. Effects of day of gestation and feeding regimen in Holstein × Gyr cows: I. Apparent total-tract digestibility, nitrogen balance, and fat deposition. J. Dairy. Sci. 98, 3197-3210.

Sampaio, C.B., Detmann, E., Valente, T.N.P., Costa, V.A.C.; Valadares Filho, S.C.; Queiroz, A.C. 2011. Fecal excretion patterns and short-term bias of intestinal and external markers in a digestion assay with cattle. Rev. Bras. Zootc. 40, 657-665.

Tedeschi, L.O., 2006. Assessment of the adequacy of mathematical models. Agric. Syst. 89, 225-247.

Valente, T.N.P., Detmann, E., Queiroz, A.C., Valadares Filho, S.C., Gomes, D.I., Figueiras, J.F. 2011. Evaluation of ruminal degradation profiles of forage using bags made from different textiles. Rev. Bras. Zootc. 40, 1217-1240.

TABLES AND FIGURES

Table 1. Descriptive statistics of the variables used to compose the dataset

Item[a]	Statistics				
	n	Mean	Minimum	Maximum	s
Week of gestation	223	19.5	38	0	11.6
DMI, kg	223	10.6	4.47	19.5	2.97
BW, kg	223	525	415	680	61.9
DMIBW, g/kg BW	223	20.2	8.98	32.4	5.52
FCP, g/kg DM	223	54.7	21.8	104	17.1
apFNDF, g/kg DM	223	685	483	744	46.9
iFNDF, g/kg DM	223	201	108	301	47.1

[a] DMI, dry matter intake; DMIBW, dry matter intake per grams of body weight; BW, body weight; FCP, forage crude protein; apFNDF, forage neutral detergent fiber corrected for contaminant ash and protein; iFNDF, forage indigestible neutral detergent fiber.

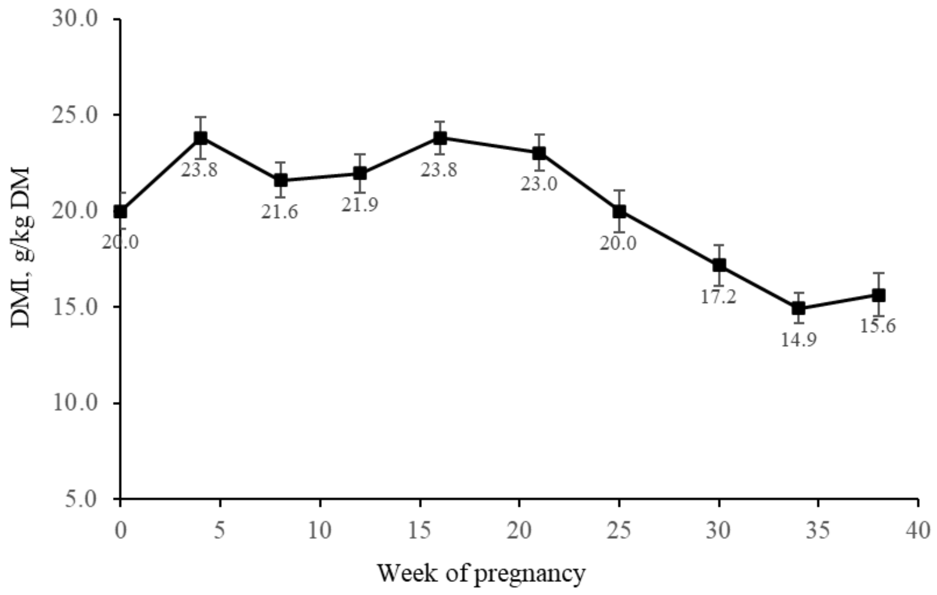

Figure 1. Variations in dry matter intake (DMI) of beef cows under grazing in tropical pastures throughout the week of pregnancy

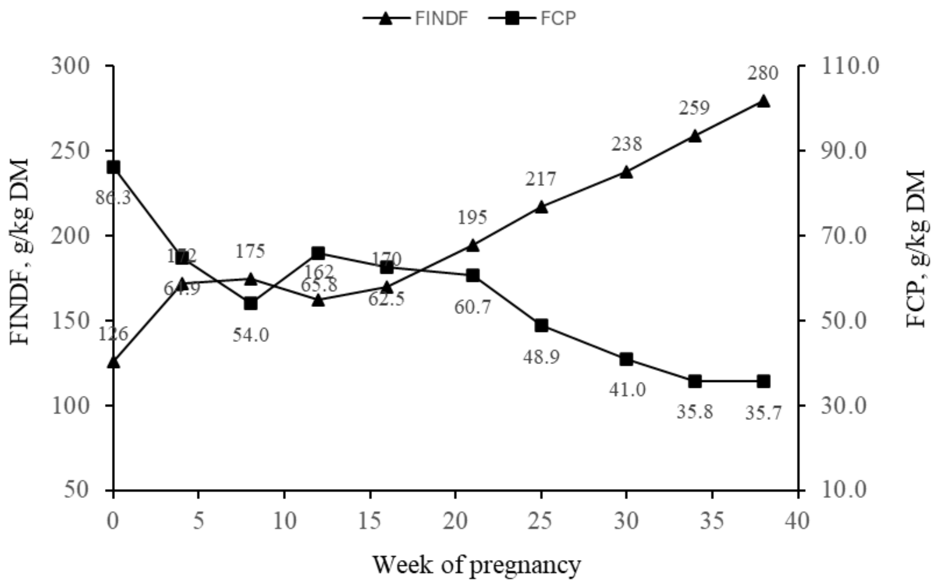

Figure 2. Variations in forage indigestible neutral detergent fiber (iFNDF) and forage crude protein (FCP) contents throughout the week of pregnancy

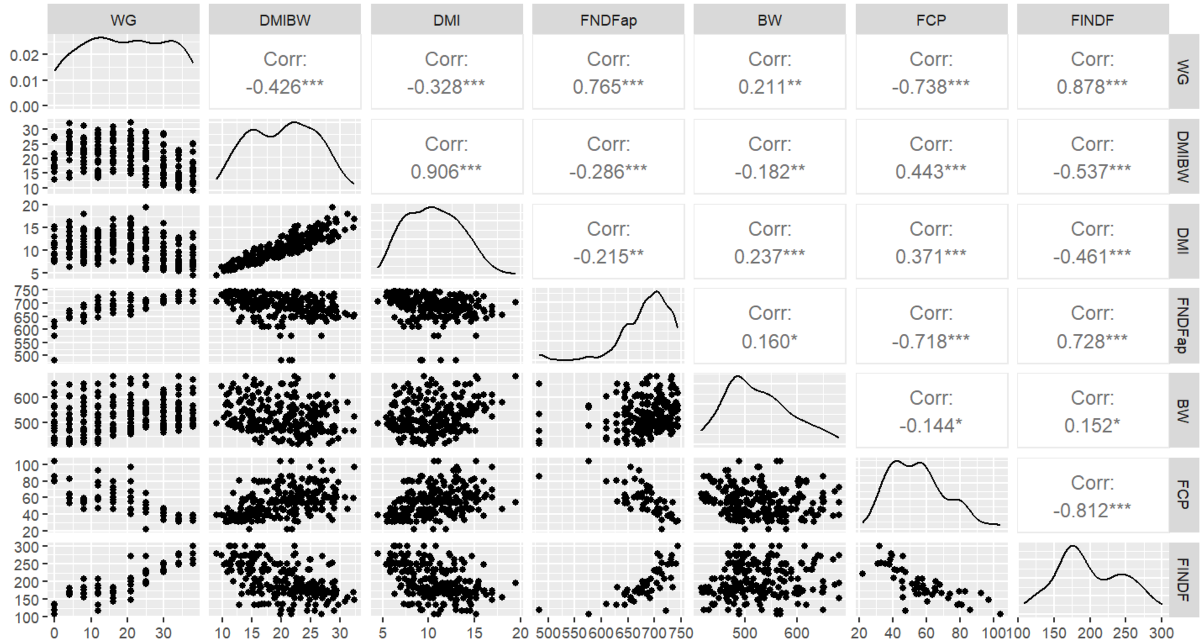

Figure 3. Main diagonal: densities of each variable (WP = week of pregnancy; DMIBW = dry matter intake, g/kg BW; DMI = dry matter intake, kg/day; apFNDF = forage neutral detergent fiber corrected for contaminant ash and protein, g/kg DM; BW = body weight, kg; FCP = forage crude protein, g/kg DM; iFNDF = forage indigestible neutral detergent fiber. Scatter plots below main diagonal: relationships among all variables. Values above main diagonal: Pearson correlation coefficients among variables. * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$.

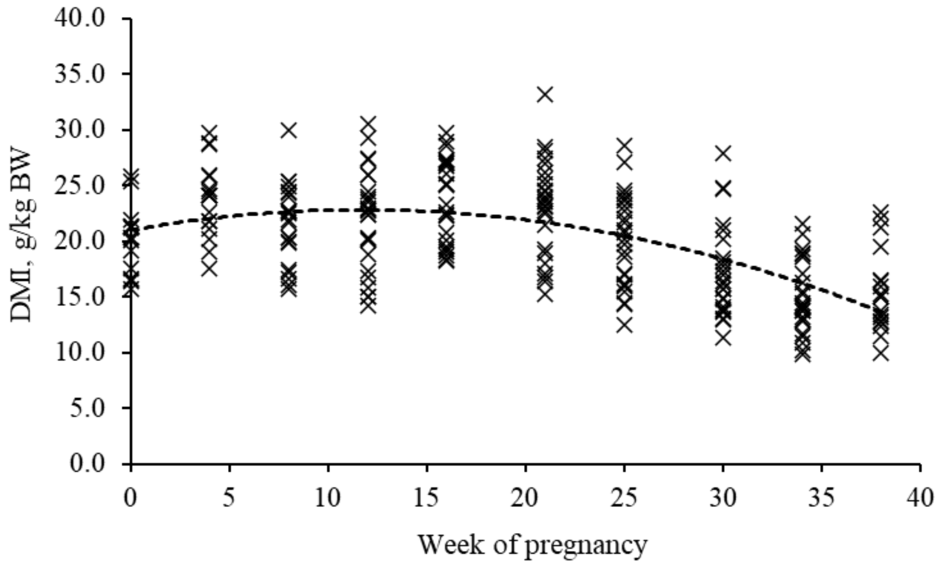

Figure 4. Relationship between week of pregnancy (WP) and dry matter intake (DMI, g/kg BW) of beef cows during gestation under grazing in tropical pastures. [DMI= $20.978_{\pm 0.9059}$ + $0.316_{\pm 0.0947}$ × WP – $0.0134_{\pm 0.0023}$ × WP^2]. The data points were adjusted for the random effect of cows.

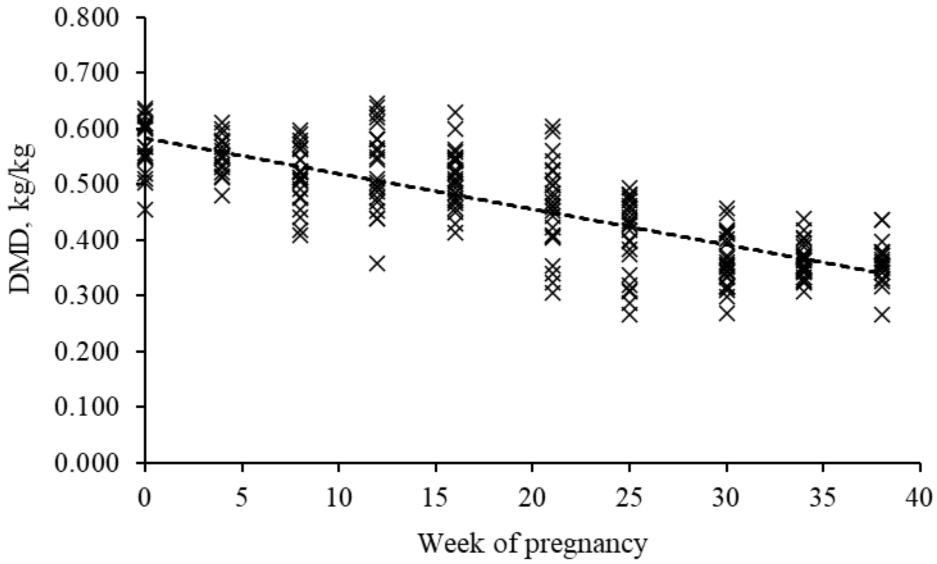

Figure 5. Relationship between week of pregnancy (WP) and dry matter digestibility (DMD, kg/kg) of beef cows during gestation under grazing in tropical pastures. [DMI = $0.5837_{\pm 0.0081} - 0.0064_{\pm 0.0003} \times$ WP]. The data points were adjusted for the random effect of cows.

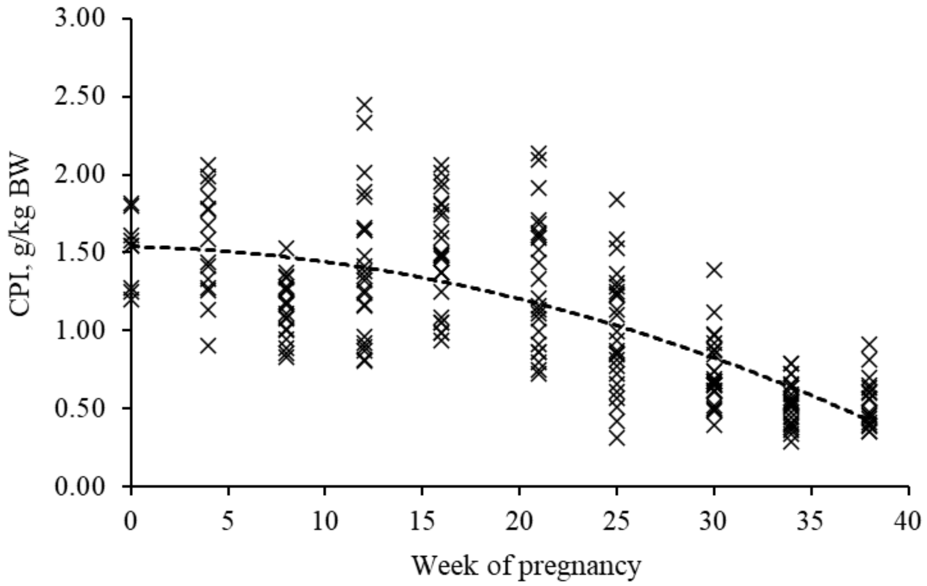

Figure 6. Relationship between week of pregnancy (WP) and crude protein intake (CPI, g/kg BW) of beef cows during gestation under grazing in tropical pastures. [DMI = $0.7600_{\pm 0.0391}$ + $0.0020_{\pm 0.0042}$ × WP – $0.0004_{\pm 0.0001}$ × WP^2]. The data points were adjusted for the random effect of cows.

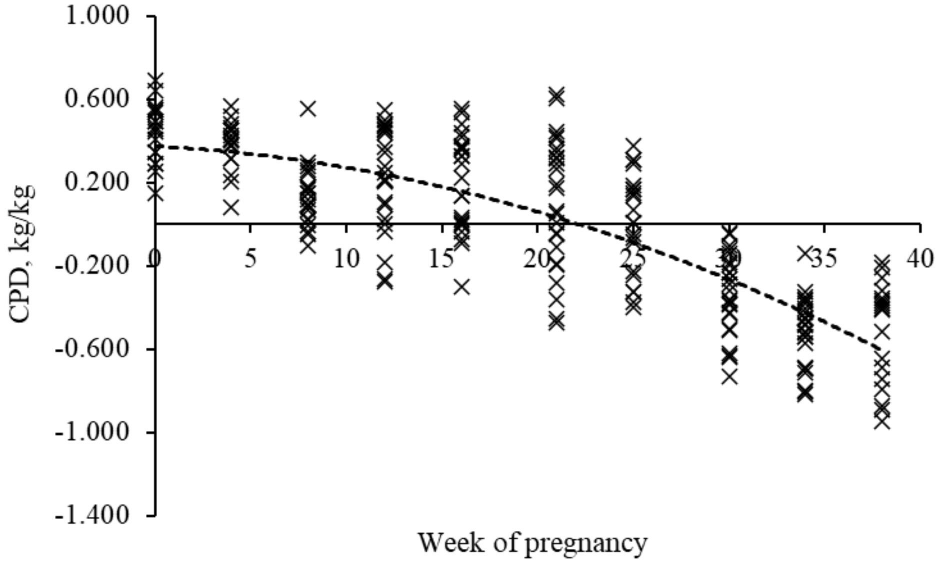

Figure 7. Relationship between week of pregnancy (WP) and crude protein digestibility (CPD, kg/kg) of beef cows during gestation under grazing in tropical pastures. [DMI= $1.5342_{\pm 0.0716}$ – $0.0023_{\pm 0.0079}$ × WP – $0.0007_{\pm 0.0002}$ × WP^2]. The data points were adjusted for the random effect of cows.

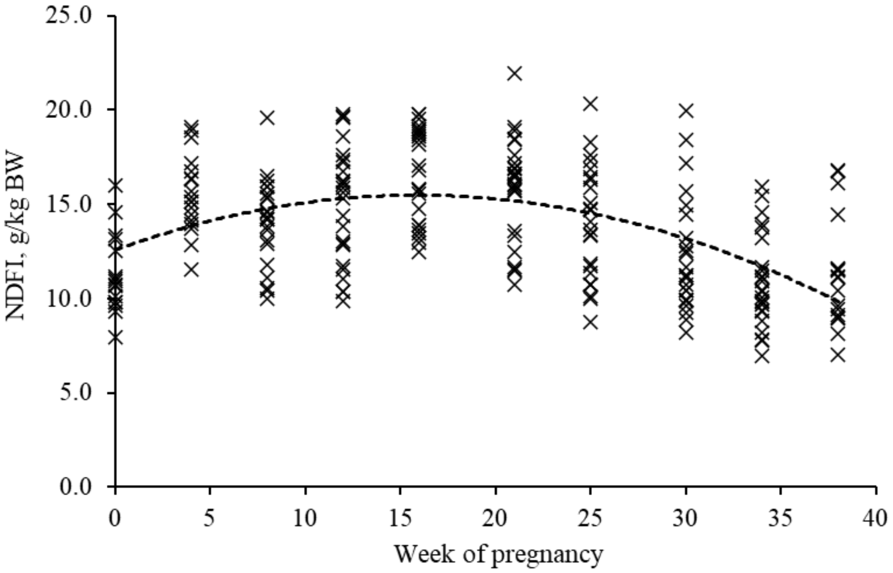

Figure 8. Relationship between week of pregnancy (WP) and neutral detergent fiber intake (iNDF, g/kg BW) of beef cows during gestation under grazing in tropical pastures. [DMI = $12.5529_{\pm 0.6128} + 0.3702_{\pm 0.0655} \times WP - 0.0116_{\pm 0.0016} \times WP^2$]. The data points were adjusted for the random effect of cows.

Table 2. Adequacy measures of the proposed model to predict the dry matter intake of beef cows during gestation under grazing in tropical pastures through leave-one out cross validation technique.

Item[a]	Actual[b]	Proposed model
Mean, g/kg BW	20.2	20.2
Maximum, g/kg BW	33.2	23.1
Minimum, g/kg BW	9.86	9.86
SD, g/kg BW	4.92	2.88
Regression		
Intercept		
Estimate ± SE	-	0.511±1.92
P-value (H_0: $\beta_0 = 0$)	-	0.79
Slope		
Estimate ± SE	-	0.975±0.094
P-value (H_0: $\beta_1 = 1$)	-	0.78
R^2	-	0.333
CCC	-	0.499
Cb	-	0.873
ρ	-	0.500
MSEP	-	16.2
MB	-	0.000
SB	-	0.00526
RE	-	16.1966

[a] R^2, coefficient of determination; CCC, concordance correlation coefficient; MSEP, mean squared error of prediction; MB, mean bias; SB, systematic bias; RE, random error.

[b] Actual, actual DMI values

CHAPTER 2

Effects of dried distiller's grains supplementation on performance and digestion characteristics of suckling beef calves in tropical pastures

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ABSTRACT

Thirty-seven Red Angus × Nellore male calves (162±4.1 kg BW; 120±30 days old) were used to evaluate the effects of dried distiller's grains (DDG) supplementation on performance and digestion characteristics of suckling beef calves on tropical pastures. Cow-calf pairs (n = 37) were randomly assigned into eight paddocks formed by *Urochloa decumbens* pastures. Animals were subjected to a 14-d adaptation period followed by a 108-d performance and digestibility period. Four treatments were evaluated: Control (mineral premix, n = 8), supplement without inclusion of DDG (n = 10), or supplements including 375 (n = 9) and 750 g/kg DDG (n = 10) on a dry matter (DM) basis. A specific contrast was applied to compare control (mineral mixture only) vs. supplemented calves, and linear and quadratic effects were used to compare 0, 375, and 750 g/kg DDG inclusion. Supplementation increased ($P \leq 0.01$) voluntary intake, although it showed no effect ($P \geq 0.46$) on neutral detergent fiber corrected for ash and protein (apNDF) or indigestible neutral detergent fiber (iNDF) intakes. Additionally, supplementation did not affect ($P < 0.42$) calves' forage intake. Conversely, the addition of DDG in the supplements tended to have a quadratic effect ($P < 0.07$) on the calves' forage intake, with the greatest intake for calves receiving a supplement containing 375 g DDG/kg DM. Supplementation improved linearly ($P \leq 0.04$) DM and crude protein (CP) digestibilities, and dietary digested organic matter (DOM). Otherwise, CP and OM digestibilities decreased linearly ($P \leq 0.03$) as the addition of DDG in the supplement increased. Additionally, adding DDG in the supplement decreased linearly ($P < 0.01$) the dietary DOM. On the other hand, a quadratic effect of DDG inclusion was detected on CP digestibility, with the lowest CP digestibility for calves receiving supplements with 375 g DDG/kg DM. Supplementation enhanced (P <0 .01) average daily gain (ADG) when compared to control calves. Conversely, ADG decreased linearly as the DDG inclusion in the supplement increased ($P \leq 0.04$). We did not detect any effect ($P \geq 0.12$) of treatment on milk yield and composition of the cows. Supplementing beef calves through creep-feeding system enhances calf performance. Conversely, including 375 or 750 g DDG/kg DM in supplements decreases digestion characteristics and performance of suckling beef calves in tropical pastures.

Keywords: Beef cattle; Byproducts; Creep feeding; Ethanol; Weight gain.

INTRODUCTION

Creep-feeding systems have been utilized successfully in cow-calf operations worldwide. Numerous studies have shown that supplementing suckling calves leads to increased weaning weights (Viñoles et al., 2013; Moriel et al., 2017; Lopes et al., 2017). Thus, creep-feeding can improve profitability in cow-calf operations. Additionally, it is widely recognized that early-life nutrient supplementation can trigger permanent changes in skeletal muscle development, leading to long-term effects on performance and meat quality of the offspring (Du et al., 2013; 2015). For instance, several studies have reported that early-life supplementation can enhance the development and differentiation of intramuscular adipogenic cells, resulting in higher marbling scores at slaughter (Shike et al., 2007; Scheffler et al., 2014; Santos et al., 2023).

Despite the numerous benefits of supplementing suckling beef calves, the use of concentrate feeds increase production costs and can impact profitability in cow-calf systems. In this light, using industry co-products to replace common feed ingredients in calf supplements may reduce feed costs while maintaining production goals in intensive cow-calf production systems.

Dried distillers grains (DDG) have been used extensively in diets of growing and finishing cattle due to their high energy and rumen undegradable protein content (RUP; Martin et al., 2007; Kopflestein et al., 2008; Buttrey et al.,2012). Overall, DDG shows significant variation in chemical composition, primarily due to differences in the type of grain and processing methods used in the ethanol production (Boever et al., 2014; Nkomba et al., 2016; Böttger et al., 2018). This variability is a key factor contributing to the wide range of performance outcomes observed in cattle fed diets containing DDG (Klopfenstein et al., 2008; Griffin et al., 2012; Torres et al., 2022).

In tropical conditions, DDG has been utilized in cattle supplements as a strategy to increase the flow of amino acids to the intestine, thereby increasing the efficiency of nitrogen

(N) utilization in cattle under grazing in tropical pastures (Ferrari et al., 2021). Several studies have shown that DDG can completely replace common protein sources (e.g., cottonseed meal) without negatively affecting the performance of grazing beef cattle (Araújo et al., 2021; Hoffmann et al., 2021; Dias et al., 2024)

However, most research on the addition of DDG in cattle diets has focused on growing and finishing beef cattle. To the best of our knowledge, no studies have investigated the use of DDG in supplements for suckling beef calves in pastures. Consequently, the effects of DDG on the performance and digestion characteristics of suckling beef calves remain to be understood. We hypothesized that the addition of DDG in supplements does not affect the performance and digestion characteristics of suckling beef calves on tropical pastures. Our objective was to evaluate the effect of DDG on the performance and digestion characteristics of suckling beef calves on tropical pastures.

MATERIAL AND METHODS

Animals, treatments, and experimental design

This study was carried out at the Extension, Teaching, and Research Unit in Beef Cattle at the Department of Animal Science of the Universidade Federal de Viçosa, Viçosa, MG, Brazil. All animal care and handling procedures followed protocols and standards approved by the Animal Care and Use Committee of the Universidade Federal de Viçosa (protocol 37-2023).

Thirty-seven Nellore × Red Angus male calves (162±4.1 kg BW; 120±30 days old) were used in this study. Cow-calf pairs (n = 37) were randomly assigned into eight paddocks with approximately 7.0 ha each, covered evenly with *Urochloa decumbens* signal grass. All paddocks were equipped with waterers and covered feeders. Both cows and calves had access *ad libitum* to fresh water and mineral mixture throughout the experiment. To minimize any likely confounding effects between treatments and paddocks, cow-calf pairs were rotated among paddocks every 15 days, ensuring that each cow-calf pair spent equal time in each paddock and passed through all paddocks the same number of times. All calves were subjected to a 14-day adaptation period and a further period of 108 days for performance evaluation.

Treatments were assigned to experimental units (calves) according to a completely randomized design. Four treatments were evaluated, as follows: control (mineral mixture only; n = 8), supplement without DDG (n = 10), or supplements including 375 (n = 9) and 750 g/kg DDG (n = 10) on a dry matter (DM) basis. The supplements were composed by ground corn, soybean meal, urea, and DDG (Table 1), and formulated to contain 300 g CP/kg DM (Table 2). The supplements were provided daily in the amount of 7.0 g/kg BW (as fed) (Carvalho et al., 2019) at 1000 h in collective troughs designated exclusively for the calves (creep feeders) positioned next to the cows' troughs. All calves were weighted every 15 days in order to adjust the amount of supplement to be provided.

Sampling and measurements

Performance characteristics were evaluated by recording calves' BW at the beginning and end of the experiment without prior fasting. Intake and digestibility trials were carried out on days 30-39, 60-69, and 90-99 of the evaluation period. Fecal output was estimated using chromic oxide (Cr_2O_3) as an external marker. Each trial lasted nine days, with five days to stabilize the marker's fecal excretion and four days for sample collection (Sampaio et al., 2011). From the first to the eighth day of each period, the Cr_2O_3 (10 g per calf) was administered daily at 1000 h via the esophagus using a metal probe. Individual supplement intake was estimated utilizing titanium dioxide (TiO_2) as an external marker, being mixed daily to the supplement at a dosage of 10 g per calf (Titgemeyer et al., 2001). Fecal collections were carried out at 1800, 1400, 1000, and 0600 h on days 6, 7, 8 and 9 of each trial, respectively (Sampaio et al/., 2011). Fecal samples were collected immediately after defecation or directly from the rectum of the calves. On the ninth day of each trial, forage samples were taken from each paddock by hand-plucking sampling (De Vries, 1995).

Samples of forage and feces were oven-dried at 55°C for 72 h and then ground to pass through a 1-mm screen sieve. Fecal samples were pooled per animal and trial (i.e., 30, 60, and 90 days of experiment). Forage samples were pooled per paddock and trial. All samples were subsequently stored in plastic jars for further analysis.

To estimate milk yield, cows were milked during the second trial (i.e, at 60 days into the experiment). To drain the udders of the cows, the calves were separated from their mothers from 1500 to 1745h when they were reunited with their mothers so that they could suckle and then drain the milk from their udders. At 1800h, the calves were separated from the cows again until the next morning, during which time they were housed in a pen with access to water. Cows were milked by a milking machine at 0500h of the following day immediately after the application of 2 mL of Oxytocin (10 IU/mL, Ocitopec®, Biovet, São Paulo, Brazil) in the mammary vein. Milk was weighed and a subsample of 30 mL was collected to evaluate nutrient composition. The exact order and time that each cow was milked was recorded. Calves remained separated from their mothers, and a new milk collection was performed at 1800h to obtain a 24-h milk yield.

Laboratory Analysis

Pooled samples of forage, concentrate ingredients, and feces were analyzed for DM (dried for 16 h at 105 ºC, method G-003/1), ash (complete combustion in a muffle oven at 550ºC, method M-001/2), CP (Kjeldahl procedure, method N-001/2), and NDF (using heat-stable α-amylase and omitting sodium sulfite, method F-012/1), following the procedures described by the Brazilian National Institute of Science and Technology in Animal Science (INCT-CA, Detmann et al., 2021). The content of NDF was expressed with correction for contaminant ash and protein (apNDF) according to equations proposed by Valadares Filho et al. (2024, in review/correction of Animal feed Science and Technology). The iNDF content was obtained through a 288-h *in situ* incubation procedure (Valente et al., 2011).

Fecal concentration of chromium and TiO_2 were determined via atomic absorption spectrophotometry (Detmann et al., 2021 methods M-005/2 and M-007/2, respectively). Milk samples were analyzed for protein, fat, lactose, and total solids by spectroscopy method (Foss MilkoScan FT120, Hillerød, Denmark).

Calculations

Fecal output was obtained as the ratio between the quantity of Cr_2O_3 offered daily and the Cr_2O_3 concentration in feces.

Individual supplement intake was calculated using the following equation:

$$SI = \frac{\mathrm{FO} \times T_iO_{2feces}}{T_iO_{2sup}} \quad (1)$$

Where SI is the supplement dry matter intake (kg/day), FO is the fecal output (kg/day), T_iO_{2feces} is the T_iO_2 concentration in the feces (kg/kg DM), T_iO_{2sup} is the T_iO_2 concentration in the supplement (kg/kg DM).

Forage intake was estimated using the indigestible neutral detergent fiber (iNDF) as an internal marker, as follows:

$$FI = \frac{[(\mathrm{FO} \times iNDF_{feces}) - (\mathrm{SI} \times iNDF_{sup})]}{iNDF_{for}} \quad (2)$$

Where FI is the forage dry matter intake (kg/day), FO is the fecal output (kg/day), $iNDF_{feces}$ is the iNDF concentration in the feces (kg/kg DM), SI is the supplement dry matter intake (kg/day), $iNDF_{sup}$ is the iNDF concentration in the supplement (kg/kg DM), and $iNDF_{for}$ is the iNDF concentration in the forage (kg/kg DM).

The milk yield was corrected for 4 % of fat using the following equation (NRC, 2001):

$$FMC = 0.4 \times MY + 15 \times FY \quad (3)$$

Where FCM is the 4% fat-corrected milk yield (kg/day), MY is the milk yield (kg/day), and FY is the fat yield (kg/day).

Statistical analysis

The data were analyzed according to the following model:

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

Where Y_{ij} is the response measured on the calf j submitted to treatment i, μ is the overall constant, T_i is the fixed effect of treatments, and $\varepsilon_{(i)j}$ is the random error, assumed ~ iid N(0, σ_ε^2). When pertinent, calves' initial BW was used as a concomitant variable in the model. If the effect of this variable was found to be non-significant, the model was reparameterized by excluding it.

Comparisons between treatments were performed by decomposing its sum of squares into orthogonal effects, as follows: 1) control (mineral mixture only) versus supplemented calves; 2) evaluation of the linear and quadratic order effects of the inclusion of DDG in the supplements. All analyses were performed utilizing the MIXED procedure of SAS 9.4 considering 0.05 as the critical level of probability for the occurrence of type I error.

RESULTS

Intake and digestibility

There was no effect ($P \geq 0.30$) of treatment on calves' milk intake (Table 3). The supplement intake averaged 1.50, 1.52, and 0.91 kg/day for calves receiving supplements with 0, 375, and 750 g DDG/kg DM, respectively. Supplementation increased ($P = 0.01$) voluntary intake, although it showed no effect ($P \geq 0.58$) on apNDF or iNDF intakes. Additionally, supplementation did not affect ($P > 0.42$) calves' forage intake.

Conversely, addition of DDG in the supplements tended to have a quadratic effect ($P < 0.07$) on forage intake. This effect indicated a greater forage intake in calves fed a supplement containing 375 g DDG/kg DM. A similar pattern ($P \leq 0.04$) was observed for the DM, OM, CP, and apNDF total intakes. On the other hand, the addition of DDG in the supplements did not affect ($P \geq 0.10$) digested organic matter (DOM) or iNDF intakes.

Supplementation improved linearly ($P \leq 0.04$) DM and CP digestibilities, and dietary DOM (Table 4). Otherwise, organic matter digestibilities decreased linearly ($P \leq 0.03$) as the addition of DDG in the supplement increased. Similarly, adding DDG in the supplement decreased linearly ($P < 0.01$) the dietary DOM. On the other hand, a quadratic effect ($P = 0.01$) of DDG inclusion was detected on CP digestibility. This pattern showed lower CP digestibility in calves receiving supplements with 375 g DDG/kg DM. The apNDF digestibility was not affected ($P \geq 0.45$) by supplementation or level of inclusion of DDG in the supplement.

Performance

Supplementation enhanced ($P \leq 0.02$) average daily gain (ADG) and final body weight (FBW) when compared with calves receiving only mineral mixture (Table 5). On average, ADG was increased from 0.96 to 1.07 kg/day for control and supplemented calves, respectively. Conversely, ADG and FBW decreased ($P \leq 0.04$) linearly as the level of inclusion of DDG in the supplement increased. The differential ADG of calves that received supplements with 375 and 750 g/kg DDG when compared to calves that did not receive DDG was -0.060 and -0.090 kg/day, respectively.

Milk yield and composition

We did not detect any effect ($P \geq 0.12$) of treatment on milk yield and composition (Table 6). On average, milk yield and 4%-fat corrected milk yield were 5.95 and 7.74 kg/day, respectively.

DISCUSSION

Supplementation increased total DM intake, without affecting forage or milk intakes. This indicates that the increase in total DM intake is directly due to the supplement provided. These results are supported by assessing intake of other dietary fractions. Supplementation was able to increase the intake of all dietary fractions except for insoluble fiber. Such as results confirm that supplementation had no negative impact on forage intake.

Several authors have reported that supplementation for suckling beef calves decreases forage intake, despite an overall increase in energy intake primarily due to the supplement supply (Lopes et al., 2017; Almeida et al., 2018; Carvalho et al., 2019). Such effects have been attributed to negative feedback signals regulating voluntary intake due to either excess energy or protein (Carvalho et al., 2019). Under these conditions, the animal may change its eating habits in an attempt to maintain body homeostasis (Forbes et al., 2007). Indeed, studies conducted in tropical conditions have demonstrated that supplementation for beef calves reduces grazing time, consequently leading to a decrease in forage intake (Valente et al., 2013; Lopes et al. 2017; Martins et al., 2017). To our knowledge, a likely reason for the lack of effect of supplementation on forage intake in our study could be attributed to the genetic group used.

There is a lack of comparative data in the literature regarding the nutrient requirements of Angus x Nellore calves versus purebred Nellore calves in tropical pastures. However, the present study indicates that the crossbred calves had DM and CP intakes that were 54% and 23% higher, respectively, than the recommendations provided by the BR-CORTE for Nellore calves in the same body weight, weight gain, week of lactation, and dam’s milk yield (Lopes et al., 2023). This suggests that crossbred beef calves may have higher protein and energy requirements compared to Zebu breeds, consistent with the higher energy and protein needs observed in growing and finishing crossbred cattle (Valadares Filho et al., 2023). Thus, it is reasonable to assume that the diet may have provided nutrients and energy that did not exceed calves' demands, mitigating any negative effects of supplementation on forage intake.

Overall, we observed that supplementation increased OM apparent digestibility. This increase contributed, at least partially, to the greater energy content in the diet, as indicated by the increase in dietary DOM of the supplemented calves. Conversely, we did not observe any effect of supplementation on insoluble neutral detergent fiber digestibility, suggesting that supplementation did not impact forage utilization. Hence, the increase in OM digestibility and dietary energy content can be linked solely to the supplement supply. To our knowledge, there are two main reasons for this. Firstly, supplement supply increased the proportion of highly digestible fractions in the diet (e.g., CP and NFC) resulting in an overall increase in OM digestibility. Secondly, the intake of non-fibrous fractions is positively correlated with the apparent digestibility of these fractions. This is due to the dilution of the fecal metabolic fraction as intake increases, resulting in increased apparent digestibility (Van Soest, 1994). These arguments also apply to the increase in CP digestibility with supplementation.

The aforementioned results on the improvement of nutritional characteristics with supplementation are supported by evaluating calves' performance. Overall, supplementation enhanced performance compared with calves receiving only mineral mixture. On average, supplementation was able to provide an additional 110 g/day of ADG compared with the calves receiving only mineral mixture. Indeed, the increase in performance is a direct effect of the increased protein and energy intake promoted by supplementation. Conversely, meta-analytical evaluations conducted in tropical conditions have shown that the supplementation for suckling male beef calves is able to provide an additional ADG of up to 200 g/day compared to calves that did not receive supplements (Carvalho et al., 2019). Several factors can explain the greater ADG reported by Carvalho et al. (2019), including supplement amount and composition, forage quality, breed, sex, and milk yield (Lopes et al., 2023). However, a likely explanation for the lower ADG observed herein compared to the aforementioned metanalysis study, may be attributed to the decreased performance of calves receiving supplements containing DDG. Ultimately, the reduced performance of calves receiving DDG led to a lower average performance of the supplemented calves (110 g/d) when compared to literature data (Carvalho et al., 2019).

Most research utilizing DDG in cattle diets has been focusing on growing and finishing cattle. To the best of our knowledge, there are no studies that have investigated the use of DDG for suckling beef calves in tropical pastures. Hence, our main objective was to evaluate the effectiveness of DDG in supplements for suckling beef calves in tropical conditions. In this light, our data indicate that the addition of high DDG levels in the supplements linearly decreased calf performance.

In feedlot settings, positive responses to the addition of DDG in terms of performance have been attributed to the increased supply of both protein and metabolizable energy (Klopfenstein et al., 2008; Griffin et al., 2012). This is primarily due to the greater levels of RUP and fat content in diets containing DDG. However, it is important to note that the DDG used in the present study had a low- fat content. Thus, some contribution of fat to enhancing feeding value in DDG diets would be small. Furthermore, the supplements had similar protein contents. Therefore, the observed results can be attributed to variations in protein availability and the content of other dietary fractions, such as insoluble fiber and non-fiber carbohydrates.

In this context, we observed that the addition of DDG linearly decreased the OM apparent digestibility. Such result can be attributed, at least partially, to the decrease in starch content and the increase in insoluble fiber content with the addition of DDG in the supplements. Accordingly, studies have demonstrated that the addition of DDG to cattle diets decreases the OM apparent digestibility (Alhadas et al., 2021; 2022) and increases the excretion of gross energy in feces (Hales et al., 2013). Similarly, it has been reported that the addition of DDG to cattle diets can decrease CP digestibility when compared to diets without DDG (Alhadas et al., 2021; 2022). Several authors have shown a significant variation in both ruminal degradability (De Boever et al., 2014; Böttger and Südekum, 2017) and intestine digestibility of DDG protein (Kleinschmit et al., 2007; Corrigan et al., 2009). Grains characteristics used and variations in processing methods between and within industries can influence the availability of DDG protein (De Boever et al., 2014). Generally, reduced DDG protein digestibility has been associated with excessive heating during processing and drying (Böttger and Südekum, 2018). Excessive heat can induce non-enzymatic reactions, such as the Maillard reaction, which involve a series of chemical reactions between AA and reducing sugars. The final products of these reactions are typically insoluble and indigestible, thereby reducing the digestibility of AA (Van Soest, 1994). Furthermore, some authors have also suggested that various steps in the ethanol production process, such as heat treatment prior to drying, blending of product streams, addition of chemicals, and milling of cereal grains, can affect protein availability (Böttger and Südekum, 2018).

Conversely, we observed a quadratic pattern in CP digestibility in response to the addition of DDG to supplements, with lower CP digestibility for calves receiving supplements containing 375 g DDG/kg DM. Interestingly, CP digestibility appeared to increase at the 750 g DDG/kg DM inclusion level, which was unexpected. It is important to note that this pattern was not influenced by the CP content of the supplements, as all supplements were formulated to contain similar CP content. A likely explanation for this pattern can be found by examining the calves' intake patterns.

We observed that animals fed supplement containing 375 g DDG/kg DM tended to have a greater forage intake compared to DDG0 and DDG750 calves. Even though a solid explanation cannot be drawn from this experiment, we speculate two likely causes for this effect. First, the addition of DDG to cattle diets increases AA flow to the intestine via increasing the RUP content in the diet (Montano et al., 2023). Some authors have reported that, under conditions of marginal nitrogen (N) deficiency, an increased supply of N in the intestine may stimulate voluntary intake, likely as an attempt to mitigate N deficiency in metabolism (Egan, 1965; Carvalho et al., 2020). This hypothesis seems plausible for young calves with a high rate of weight gain. Additionally, the inclusion of DDG decreased the starch content in the supplements. The lower starch content in supplements containing DDG could reduce the negative effects of excess energy, as observed in supplemented calves in tropical pastures (Lopes et al., 2017; Carvalho et al., 2019).

Logically, the increased forage intake in calves receiving supplements with 375 g DDG/kg DM resulted in a greater total DM intake, which in turn increased the intake of other dietary fractions, including CP. While we cannot determine the exact contribution of the protein fraction from forage and supplement to the CP apparent digestibility, it is reasonable to assume that the increased CP intake due to greater forage intake may have led to lower CP digestibility, explaining the observed quadratic pattern. Thus, the higher CP digestibility for calves receiving 750 g DDG/kg DM might indicate a decrease in the CP fraction derived from forage, rather than a direct effect of the CP from DDG.

Taken together, our data indicate that the addition of DDG to the supplements decreased OM digestibility due to lower starch content and increased fiber content, as well as reduced CP digestibility in DDG-fed calves. These findings are supported by the linear decrease in the DOM content of the diet with the addition of DDG, thereby implying reduced energy availability for animal metabolism. Accordingly, Alhadas et al. (2021) reported that the addition of low-fat DDG to finishing cattle diets reduced energy intake and negatively impacted animal performance

Additionally, one point must be highlighted here. By design, we defined the same amount of supplement for all treatments. However, we observed that calves fed supplements containing

750 g DDG/kg DM refused the supplement, resulting in lower supplement average intake. In addition to the forementioned arguments, the lower supplement intake also helps to explain the reduced performance of calves receiving supplements with 750 g DDG/kg DM. To the best of our knowledge, no data in the literature reports a reduction in supplement intake with added DDG. Some authors have noted a reduction in DDG intake when supplied above 11 g/kg BW to grazing calves (Gustad et al., 2006). However, in that study, DDG was provided alone, which was not the case in our study. Some studies suggest that increased sulfur and fat levels can reduce voluntary intake when high levels of DDG are included in cattle diets (Sarturi et al., 2013; Drewnoski et al., 2014; NASEM, 2016). In our study, any effect of fat on supplement intake is unlikely, as we utilized low-fat DDG. Unfortunately, we do not have information about the sulfur content of the DDG used in this study. Additionally, we speculate that excessive heating during milling processing may have resulted in a DDG with low palatability for calves, though this hypothesis requires further investigation.

Finally, we did not observe any effects of supplementation or level of inclusion of DDG on cows' milk yield and composition. The effects of creep-feeding on cows' milk production are quite inconsistent. Some authors have reported that creep-feeding could reduce cows' milk yield due to a decrease in suckling stimulation (Fordyce et al., 1996). However, a meta-analytic evaluation of a large dataset from tropical pastures indicates that creep-feeding supplementation does not affect dam's milk yield (Lopes et al., 2016). These authors suggest that calves appear to have a feed preference hierarchy, as follows: milk, concentrate feed, and forage. Hence, as observed in the present study, the effects of creep-feeding supplementation on cow's milk yield are likely minimal.

Collectively, our results indicate that supplementation improved performance and nutritional characteristics of suckling beef calves in tropical pastures. However, the addition of up to 375 g DDG/kg DM in the supplement appears to stimulate forage intake in suckling beef calves. Despite this increase in forage intake, DDG decreased both energy intake and overall performance of the calves. Notably, calves receiving 750 g DDG/kg DM had lower supplement intake. Therefore, further studies evaluating different types of DDG with varying inclusion levels for suckling beef calves are encouraged.

CONCLUSION

Supplementing beef calves through creep-feeding system enhances animal performance. Conversely, including 375 or 750 g DDG/kg DM in supplements decreases digestion characteristics and performance of suckling beef calves in tropical pastures.

REFERENCES

Alhadas, H.M., Valadares Filho, S.C., Tedeschi, L.O., Vilela, R.S., Souza, G.A., Lage, B.C., Silva, B.C., Rennó, L.N. and Paulino, M.F., 2021. Impact of different levels of low-fat dried distillers grains on performance of young Nellore bulls during the finishing phase. Anim. Sci. J., 92 3623

Alhadas, H.M., Valadares Filho, S.C., Tedeschi, L.O., Vilela, R.S., Souza, G.A., Lage, B.C., Silva, B.C., Rennó, L.N. and Paulino, M.F., 2022. Effect of inclusion levels of low-fat dried distillers grains in finishing diets on protein and energy intake and retention and estimation of protein and energy requirements of young Nellore bulls fed with high concentrate diets. Anim. Sci. J, 93. 13678.

Böttger, C. and Südekum, K.H., 2017. Within plant variation of distillers dried grains with solubles (DDGS) produced from multiple raw materials in varying proportions: Chemical composition and in vitro evaluation of feeding value for ruminants. Anim. Feed Sci. Technol., 229, 79-90.

Böttger, C. and Südekum, K.H., 2018. Protein value of distillers dried grains with solubles (DDGS) in animal nutrition as affected by the ethanol production process. Anim. Feed Sci. Technol., 244, 11-17.

Buckner, C.D., Wilken, M.F., Benton, J.R., Vanness, S.J., Bremer, V.R., Klopfenstein, T.J., Kononoff, P.J., Erickson, G.E., 2011. Nutrient variability for distillers grains plus solubles and dry matter determination of ethanol by-products. Prof. Anim. Sci. 27, 57–64.

Buttrey, E.K., McCollum III, F.T., Jenkins, K.H., Patterson, J.M., Clark, B.E., Luebbe, M.K., Lawrence, T.E. and MacDonald, J.C., 2012. Use of dried distillers grains throughout a beef production system: Effects on stocker and finishing performance, carcass characteristics, and fatty acid composition of beef. J. Anim. Sci., 90, 2381-2393.

Carvalho, V.V., Paulino, M.F., Detmann, E., Valadares Filho, S.C., Lopes, S.A., Rennó, L.N., Sampaio, C.B. and 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. Livest. Sci., 227, 175-182.

Corrigan, M.E., Klopfenstein, T.J., Erickson, G.E., Meyer, N.F., Vander Pol, K.J., Greenquist, M.A., Luebbe, M.K., Karges, K.K. and Gibson, M.L., 2009. Effects of level of condensed distillers solubles in corn dried distillers grains on intake, daily body weight gain, and digestibility in growing steers fed forage diets. J. Anim. Sci., 87, 4073-4081.

De Araújo, T.L.D.R., da Silva, W.L., Berça, A.S., Cardoso, A.D.S., Barbero, R.P., Romanzini, E.P. and Reis, R.A., 2021. Effects of replacing cottonseed meal with corn dried distillers' grain on ruminal parameters, performance, and enteric methane emissions in young nellore bulls reared in tropical pastures. Animals, 11, 2959.

De Carvalho P. C., Doelman, I; Martín-Tereso, J., 2020. Post-ruminal non-protein nitrogen supplementation as a strategy to improve fibre digestion and N efficiency in the ruminant. J. Anim. Physiol. Anim. Nutr., 104, 64-75.

De Boever, J.A., Blok, M.C., Millet, S., Vanacker, J. and De Campeneere, S., 2014. The energy and protein value of wheat, maize and blend DDGS for cattle and evaluation of prediction methods. Animal, 8, 1839-1850.

De Vries, M.F.W. 1995. Estimating forage intake and quality in grazing cattle: A reconsideration of the hand-plucking method. J. Range Manag. 48, 370-375.

Depenbusch, B.E., Coleman, C.M., Higgins, J.J. and Drouillard, J.S., 2009. Effects of increasing levels of dried corn distillers grains with solubles on growth performance, carcass characteristics, and meat quality of yearling heifers. J. Anim. Sci., 87, 2653-2663.

Dias, M.R., Moraes, K.A.K.D., Oliveira, A.S.D., Batista, E.D., Salomão, A.M.R., Zambenedetti, A., Petrenko, N.B., Sousa, J.N., Ortelam, J.C., Ickert, A. and Chaves, C.S.,

2024. Nutritional Performance of Grazing Beef Cattle Supplemented with High-Protein Distillers' Dried Grain. Animals, 14, 1209.

Du, M., Huang, Y., Das, A. K., Yang, Q., Duarte, M. S., Dodson, M. V., & Zhu, M. J. (2013). Meat science and muscle biology symposium: Manipulating mesenchymal progenitor cell differentiation to optimize performance and carcass value of beef cattle. J. Anim. Sci., 91, 1419–1427.

Du M, Wang B, Fu X, Yang Q, Zhu MJ. 2015. Fetal programming in meat production. Meat Sci. 109, 40-7.

Drewnoski ME, Pogge DJ, Hansen SL. 2014. High-sulfur in beef cattle diets: a review. J Anim Sci. 92, 3763-80.

Egan, A.R., 1965. Nutritional status and intake regulation in sheep. II. The influence of sustained duodenal infusions of casein or urea upon voluntary intake of low-protein roughages by sheep. Aust. J. Agric. Res., 16, 451-462.

Ferrari, A.C., Leite, R.G., Fonseca, N.V., Romanzini, E.P., Cardoso, A.D.S., Barbero, R.P., Costa, D.F., Ruggieri, A.C. and Reis, R.A., 2021. Performance, nutrient use, and methanogenesis of Nellore cattle on a continuous grazing system of *Urochloa brizantha* and fed supplement types varying on protein and energy sources. Livest. Sci., 253, 104716.

Fordyce, J., Cooper, N.J. and Kendall, I.E. 1996. Creep feeding and prepartum supplementation effects on growth and fertility of Brahman-cross cattle in the dry tropics. Aust. J. Exp. Agric., 36, 389-395.

Griffin, W.A., Bremer, V.R., Klopfenstein, T.J., Stalker, L.A., Lomas, L.W., Moyer, J.L. and Erickson, G.E., 2012. A meta-analysis evaluation of supplementing dried distillers grains plus solubles to cattle consuming forage-based diets. Prof. Anim. Sci., 28, 306-312.

Gustad, K.H., Klopfenstein, T.J., Erickson, G.E., Vander Pol, K.J., MacDonald, J.C. and Greenquist, M.A., 2006. Dried distillers grains supplementation of calves grazing corn residue. Nebraska Beef Cattle Reports, 114.

Hales, K.E., Cole, N.A. and MacDonald, J.C., 2013. Effects of increasing concentrations of wet distillers grains with solubles in steam-flaked, corn-based diets on energy metabolism, carbon-nitrogen balance, and methane emissions of cattle. J. Anim. Sci., 91, 819-828.

Hoffmann, A., Berça, A.S., Cardoso, A.D.S., Fonseca, N.V.B., Silva, M.L.C., Leite, R.G., Ruggieri, A.C. and Reis, R.A., 2021. Does the effect of replacing cottonseed meal with dried distiller's grains on Nellore bulls finishing phase vary between pasture and feedlot? Animals, 11, 85.

Kleinschmit, D.H., Anderson, J.L., Schingoethe, D.J., Kalscheur, K.F. and Hippen, A.R., 2007. Ruminal and intestinal degradability of distillers grains plus solubles varies by source. J. Dairy Sci., 90, 2909-2918.

Klopfenstein, T.J., Erickson, G.E. and Bremer, V.R., 2008. Board-invited review: Use of distillers by-products in the beef cattle feeding industry. J. Anim Sci., 86, 1223-1231.

Lopes, S.A., Paulino, M.F., Detmann, E., Valadares Filho, S. C., Valente, É.E.L., Barros, L.V., Cardenas, J.E.G., Almeida, D.M., Martins, L.S. and Silva, A.G., 2014. Supplementation of suckling beef calves with different levels of crude protein on tropical pasture. Trop. Anim. Health Prod, 46, 379-384.

Lopes, S.A., Paulino, M.F., Detmann, E., Valente, É.E.L., de Barros, L.V., Rennó, L.N., de Campos Valadares Filho, S. and Martins, L.S., 2016. Does supplementation of beef calves by creep feeding systems influence milk production and body condition of the dams? Trop. Anim. Health Prod., 48, 1241-1246.

Lopes, S.A., Paulino, M.F., Detmann, E., Valente, É.E.L., Rennó, L.N., Valadares, R.F.D., Cardenas, J.E.G., Almeida, D.M.D., Moura, F.H.D. and Oliveira, C.A.S., 2017. Avaliação de planos de suplementação para bezerros de corte lactentes manejados em pasto tropical. Semina: Cienc. Agrar., 38, 1027-1040.

Lopes, S.A., Costa, L.F., Silva Filho, S.D.C.V., de Lana Ferreira, M.F., Saraiva, D.T., Matos, E. M. A., Paulino, P. V. R.., Paulino, M.F., Siqueira, G.R., Exigências nutricionais de vacas de corte lactantes e seus bezerros. 2023. In: Valadares Filho, S. de C., Saraiva, D. T., Benedeti. P.B., Silva, F. A. S., Chizzotti, M. L. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Suprema Grafica Ltda, Visconde do Rio Branco, 299-328.

Martin, J.L., Cupp, A.S., Rasby, R.J., Hall, Z.C. and Funston, R.N., 2007. Utilization of dried distillers grains for developing beef heifers. J. Anim Sci., 85, 2298-2303.

Martins, L.S., Paulino, M.F., Rennó, L.N., Detmann, E., de Almeida, D.M., Ortega, R.M. Moreno, D.P.S. and Cárdenas, J.E.G., 2017. Creep feeding effects on male Nellore calves influencing behavior and performance of their dams. Trop. Anim. Health Prod, 49, 1669-1676.

Moriel, P., Vendramini, J.M., Arthington, J.D., Aguiar, A.D., Caputti, G., 2017. Effects of crude protein level and degradability of limited creep-feeding supplements on performance of beef cow-calf pairs grazing limpograss pastures. Liv. Sci., 200, 1–5.

Montano M. F., Carvalho P. H. V., Ferraz Junior M. V. C., Latack B. C., Zinn R. A. 2023. Influence of level of dried distillers grains plus solubles substitution for steam-flaked corn on characteristics of growth performance, and dietary energetics of calf-fed Holstein steers during the initial 16-week growing phase: metabolizable protein versus metabolizable amino acids. Transl Anim Sci. 7, 24.

NASEM. 2016. Nutritional Requirements of Beef Cattle, Eighth revised edition. Washinton, D. C. National Academies Press.

Nkomba, E. Y., E. van Rensburg, A. F. A. Chimphango, and J. F. Gorgens. 2016. The influence of sorghum grain decortication on bioethanol production and quality of the distillers dried grains with solubles using cold and conventional warm starch processing. Bioresour. Technol. 203, 181–189.

Sampaio, C.B., Detmann, E., Valente, T.N.P., Costa, V.A.C., Valadares Filho, S.D.C. and Queiroz, A.C.D., 2011. Fecal excretion patterns and short-term bias of internal and external markers in a digestion assay with cattle. Rev. Bras. Zootec., 40, 657-665.

Santos, M. M., Costa, T. C., Silva, W., Pistillo, L.Z., Valente Junior D.T., Verardo, L.L., Paulino, P.V.R., Sampaio, C.B., Gionbelli, M.P., Du, M., Duarte, M.S. 2023. Nutrient supplementation of beef female calves at pre-weaning enhances the commitment of fibro-adipogenic progenitor cells to preadipocytes. Meat Sci., 204, 109286.

Sarturi, J.O., Erickson, G.E., Klopfenstein, T.J., Vasconcelos, J.T., Griffin, W.A., Rolfe, K.M., Benton, J.R., Bremer, V.R. 2013. Effect of sulfur content in wet or dry distillers grains fed at several inclusions on cattle growth performance, ruminal parameters, and hydrogen sulfide. J. Anim. Sci. 91.

Scheffler, J. M., McCann M. A, Greiner, S. P., Jiang H., Hanigan, M. D., Bridges, G. A., Lake, S. L., Gerrard, D. E.. 2014. Early metabolic imprinting events increase marbling scores in fed cattle. J. Anim. Sci. 92, 320-4.

Shike, D.W., Faulkner, D.B., Cecava, M.J., Parrett, D.F. and Ireland, F.A.,2007. Effects of weaning age, creep feeding, and type of creep on steer performance, carcass traits, and economics. Prof. Anim. Sci, 23, 325-332.

Spiehs, M.J., Whitney, M.H. and Shurson, G.C., 2002. Nutrient database for distiller's dried grains with solubles produced from new ethanol plants in Minnesota and South Dakota. J. Anim. Sci., 80, 2639-2645.

Titgemeyer, E.C., Armendariz, C.K., Bindel, D.J., Greenwood, R.H. and Löest, C.A., 2001. Evaluation of titanium dioxide as a digestibility marker for cattle. J. Anim. Sci, 79, 1059-1063.

Torres, R.N.S.; Coelho, L.M., Paschoaloto, J.R., Ghedini, C.P., Neto, O.R.M., Chardulo, L.A.L., Almeida, M.T.C. 2022. Does distillers' grains supplementation affect beef cattle performance, carcass characteristics, and meat quality? A meta-analysis study. Res. Vet. Sci., 149, 21–35.

Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. 2023. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Third revised edition. Suprema Grafica Ltda, Visconde do Rio Branco, 466.

Valente, T.N.P., Detmann, E., Queiroz, A.C., Valadares Filho, S.C., Gomes, D.I., Figueiras, J.F. 2011. Evaluation of ruminal degradation profiles of forages using bags made from different textiles. Rev. Bras. Zootec., 40, 2565–2573.

Valente, É.E.L., Paulino, M.F., Detmann, E., Valadares Filho, S.C.V., Barros, L.V., Paula, N.F., Lopes, S.A., Almeida, D.M., Martins, L.S., 2013. Effect of calves' supplementation on performance, nutritional and behavioral characteristics of their dams. Trop. Anim. Health Prod., 45, 487-495.

Van Soest, P.J., 1994. Nutritional ecology of the ruminant, Second edition. Cornell University Press, Ithaca, New York.

Viñoles, C., Jaurena, M., De Barbieri, I., Do Carmo, M., Montossi, F., 2013. Effect of creep feeding and stocking rate on the productivity of beef cattle grazing grasslands. New Zeal. J. Agric. Res. 56, 279–287.

TABLES

Table 1

Chemical composition of supplements ingredients.

Item (g/kg DM)[a]	Ground corn	Soybean meal	DDG	Urea/SA[b]
DM, g/kg as fed	855	874	890	980
OM	984	929	978	998
CP	87.6	520	338	2,537
EE	34.0	17.0	46.8	-
apNDF	124	155	432	-
iNDF	23.4	20.6	56.3	-
NFC	738	237	-	-

[a]DM, dry matter, OM, organic matter, CP, crude protein, EE, ether extract, apNDF, neutral detergent fiber corrected for contaminant ash and protein; iNDF, indigestible neutral detergent fiber; NFC, non-fibrous carbohydrates.

[b]Urea/SA, urea and ammonium sulfate (9:1).

Table 2

Ingredients and chemical composition of supplements and forage

Item (g/kg DM)[a]	Addition of DDG in the supplement (g/kg DM)			*Urochloa Decumbens*[b]		
	0	375	750	30-39 d	60-69 d	90-99 d
Ingredients						
Ground corn	562	385	213			
Soybean meal	417	211	11			
DDG	0	383	757			
Urea/AS	20	20	20			
Chemical composition						
DM	865	875	884	301±1.07	351±1.29	415±1.55
OM	961	970	979	908±0.69	915± 0.39	915±0.44
CP	318	325	331	68.3±0.42	65.2±0.62	47.2±0.42
EE	26.2	34.6	42.8	14.5±0.16	19.9±0.21	15.2±0.18
apNDF	135	246	355	696±0.67	679±0.66	700±0.79
iNDF	21.8	35.0	47.8	171±0.38	175±0.83	207±1.04
NFC	514	396	281	129±0.71	151±0.73	153±0.63

[a] DDG, dry distiller grains, DM, dry matter, OM, organic matter, CP, crude protein, EE, ether extract, apNDF, neutral detergent fiber corrected for contaminant ash and protein; iNDF,

indigestible neutral detergent fiber; NFC, non-fibrous carbohydrates, Urea/AS, urea and ammonium sulfate (9:1). [b]Hand-plucked samples, days relative to digestibility.

Table 3

Effects of supplementation and dried distillers grains on voluntary intake of suckling beef calves in tropical pastures

Item[a]	Treatments				P-value[b]		
	Control	DDG0	DDG375	DDG750	C vs. S	L	Q
kg/day							
TDMI	4.66±0.32	5.46±0.28	6.11±0.30	5.19±0.28	0.014	0.50	0.035
FDMI	3.62±0.26	3.11±0.23	3.73±0.24	3.29±0.23	0.42	0.59	0.078
SDMI	-	1.50±0.15	1.52±0.16	0.91±0.15	-	-	-
MDMI	1.05±0.13	0.84±0.11	0.86±0.12	0.97±0.12	0.30	0.38	0.70
OM	4.28±0.29	5.08±0.26	5.66±0.27	4.81±0.26	0.009	0.46	0.035
CP	0.45±0.05	0.86±0.05	0.95±0.05	0.76±0.05	<0.001	0.17	0.032
apNDF	2.39±0.18	2.23±0.17	2.83±0.18	2.46±0.17	0.58	0.32	0.030
DOM	2.81±0.20	3.65±0.18	3.73±0.19	3.23±0.18	0.002	0.10	0.19
iNDF	0.69±0.05	0.62±0.04	0.73±0.05	0.68±0.04	0.88	0.34	0.18
g/kg BW							
TDMI	21.5±1.46	24.2±1.30	26.4±1.37	23.2±1.30	0.065	0.62	0.11
FDMI	16.8±1.27	13.8±1.19	16.0±1.19	14.8±1.13	0.20	0.51	0.23
apNDF	11.1±0.86	9.87±0.77	12.1±0.81	11.1±0.77	0.89	0.27	0.099
iNDF	3.17±0.23	2.73±0.21	3.12±0.22	3.05±0.23	0.45	0.28	0.37

[a] TDMI, total dry matter intake, FDMI, forage dry matter intake, SDMI, supplement dry matter intake, MDMI, milk dry matter intake, OM, organic matter, CP, crude protein, apNDF, neutral detergent fiber corrected for contaminant ash and protein; DOM, digested organic matter; iNDF, indigestible neutral detergent fiber. [b]C vs. S, calves receiving only mineral mixture (control) versus supplemented calves, L, Q = linear and quadratic effects of the addition of DDG in the supplements.

Table 4

Effects of supplementation and dried distillers grains on digestibility in suckling beef calves in tropical pastures

Item[a]	Treatments				P-value[b]		
	Control	DDG0	DDG375	DDG750	C vs. S	L	Q
g/kg							
DM	624±16.1	692±14.3	650±15.1	647±14.3	0.035	0.034	0.28
OM	660±15.9	727±14.2	676±15.0	675±14.2	0.076	0.015	0.17
CP	557±23.3	732±20.8	636±22.0	676±20.8	<0.001	0.066	0.014
apNDF	683±8.6	690±7.7	683±8.1	681±7.7	0.85	0.45	0.80
DOM	607±14.8	676±13.2	626±13.9	625±13.2	0.041	0.011	0.15

[a] DM, dry matter, OM, organic matter, CP, crude protein, apNDF, neutral detergent fiber corrected for contaminant ash and protein; DOM, digested organic matter content in the diet.

[b]C vs. S, calves receiving only mineral mixture (control) versus supplemented calves, L, Q = linear and quadratic effects of the addition of DDG in the supplements.

Table 5

Effects of supplementation and dried distillers grains on performance of suckling beef calves in tropical pastures

Item[a]	Treatments				P-value[b]		
	Control	DDG0	DDG375	DDG750	C vs. S	L	Q
IBW, kg	162±9.1	159±8.1	164±8.6	163±8.1	0.96	0.77	0.74
FBW, kg	281±5.0	301±3.9	293±4.3	290±3.9	0.019	0.045	0.60
ADG, kg/day	0.96±0.04	1.12±0.03	1.06±0.04	1.03±0.03	0.015	0.038	0.62
aADG, kg/day	-	0.160	0.100	0.070	-	-	-
dADG, kg/day	-	-	-0.060	-0.090	-	-	-

[a]IBW, initial body weight, FBW, final body weight, ADG, average daily gain, aADG, additional weight gain in relation to animals without supplementation (control); dADG, differential weight gain in relation to animals without addition of DDG in the supplement.

[b]C vs. S, calves receiving only mineral mixture (control) versus supplemented calves, L, Q = linear and quadratic effects of the addition of DDG in the supplements.

Table 6

Effects of supplementation and dried distillers grains for suckling beef calves on milk yield and composition of their dams in tropical pastures

Item[a]	Treatments				P-value[b]		
	Control	DDG0	DDG375	DDG750	C vs. S	L	Q
kg/day							
Milk	6.77±0.81	5.46±0.73	5.34±0.77	6.21±0.73	0.23	0.47	0.59
[a]FCM	8.54±1.07	6.99±0.95	6.90±1.01	8.55±0.95	0.38	0.25	0.47
%							
Protein	3.88±0.15	4.00±0.14	4.22±0.14	3.92±0.15	0.34	0.69	0.14
Fat	5.74±0.48	5.70±0.45	5.86±0.47	6.69±0.45	0.54	0.12	0.55
Lactose	4.63±0.24	4.32±0.22	4.18±0.23	4.09±0.22	0.12	0.46	0.91
Total solids	15.4±0.68	15.2±0.61	16.1±0.64	15.9±0.61	0.65	0.47	0.38

[a]FMC, 4 % fat-corrected milk yield.

[b]C vs. S, calves receiving only mineral mixture (control) versus supplemented calves, L, Q = linear and quadratic effects of the addition of DDG in the supplements.

CHAPTER 3

Effect of including sugarcane molasses on performance, digestion, and rumen epithelium cell characteristics in finishing beef heifers

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ABSTRACT

Our objective was to evaluate whether the inclusion of molasses affect voluntary intake, performance, digestive characteristics, and rumen development in finishing beef heifers. Twelve newly weaned Red Angus × Nellore heifers averaging 238 ± 5.1 kg, were used. The basal diet consisted of corn silage, sorghum silage, ground corn, soybean meal, urea, and mineral mixture, with a forage-to-concentrate ratio of 50:50. All heifers were fed *ad libitum* twice daily at 0700 and 1600 h, allowing up to 10% in orts (as fed). The following treatments were evaluated: control (no addition of powdered molasses) or addition of 50 g/kg of diet (DM basis) of powdered molasses. There was no effect ($P \geq 0.15$) of molasses inclusion on any of the intake variables. Similarly, molasses addition had no effect ($P \geq 0.06$) on the total-tract apparent digestibility or total digestible nutrient (TDN) content of the diet. The inclusion of molasses in the diet did not affect ($P \geq 0.19$) nitrogen (N) intake or N excretion in feces and urine. Consequently, no significant effects ($P \geq 0.32$) were observed for N retention or utilization efficiency with molasses addition. Providing molasses increased rumen pH compared to control ($P < 0.032$). Molar proportion of both acetate ($P<0.013$) and butyrate ($P < 0.012$) were higher in heifers fed molasses than control. On the other hand, dietary molasses addition decreased the propionate molar proportion ($P < 0.001$). Molasses addition had no impact ($P \geq 0.59$) on papillae length or width. Likewise, there was no effect ($P > 0.82$) of molasses on the number of ruminal papillae per cm^2. There was no ($P \geq 0.72$) effect of molasses on performance variables. Similarly, dietary molasses addition did not affect hot carcass weight ($P > 0.15$) or cold carcass weight ($P > 0.19$. No effect ($P > 0.87$) of molasses was detected on ribeye area. Conversely, the addition of molasses to the diet increased ($P < 0.024$) subcutaneous fat thickness compared to the control group. Providing sugarcane molasses does not affect the intake, digestion characteristics, and performance of young beef finishing heifers. While the inclusion of molasses increases the molar proportion of butyrate, it does not promote

development of rumen epithelium. However, the addition of molasses can affect carcass fat thickness.

Keywords: Byproducts; Beef Cattle; Nutrition; Butyrate

INTRODUCTION

Fattening young cattle is a widely adopted practice designed to enhance meat quality and enable producers to access more demanding markets, including those with stringent export standards. In such systems, animals are confined immediately after weaning and subjected to various management practices such as vaccination, deworming, and batch formation prior to the finishing phase. However, the period immediately following weaning poses significant challenges during the early stages of calf feedlot. During this time, calves are exposed to multiple stressors, including the loss of maternal bond, sudden changes in diet and environment, and the necessity of establishing a new social hierarchy. These stressors can significantly reduce voluntary feed intake, leading to a decline in performance, particularly during the critical adaptation period (Silva et al., 2023). To address these challenges and minimize both stress and productivity losses, various strategies have been developed, with nutritional management being particularly prominent.

Molasses is a widely used feed in the diets of production animals, especially during early life in order to stimulate feed intake. In cattle, the use of molasses has benefits related to improved voluntary intake and increased production of volatile fatty acids (VFA), especially butyrate (Brown et al., 1987; Lesmeister, et al., 2005). Butyrate is closely related to gastrointestinal tract (GIT) health, as it contributes to the development and maintenance of GIT integrity, especially mucosal epithelial cells (Górka, et al., 2017). In theory, this could increase the absorption area of the ruminal epithelium and improve the utilization of diet components, resulting in enhanced voluntary intake and immune response capacity in stressful situations.

Additionally, incorporating molasses into the diets of finishing cattle can enhance immune system resilience, making the animals less susceptible to potential challenges upon arrival at feedlot, such as immune system decline, respiratory diseases, and ruminal environment imbalances. Furthermore, the inclusion of molasses can help stabilize feed intake throughout the entire feedlot period, thereby ensuring consistently satisfactory production levels (Reinhardt et al., 2015; Kos et al., 2023).

Hence, we hypothesized that providing molasses in the diet enhances voluntary intake and performance in young beef finishing heifers. Our objective was to evaluate whether the effects of molasses inclusion on voluntary intake, performance, digestive characteristics, and the absorption area of the ruminal epithelium in young beef finishing heifers.

MATERIAL AND METHODS

Animal management, experimental design, and diets

This experiment was carried out at the Experimental Feedlot at the Department of Animal Science of the Universidade Federal de Viçosa, Minas Gerais, Brazil. All animal care and handling procedures followed protocols and standards approved by the Animal Care and Use Committee of the Universidade Federal de Viçosa (protocol 02-2024).

Twelve newly weaned Red Angus × Nellore heifers, averaging 238±5.14 kg and 8±1 months old, were used. The heifers originated from the Extension, Teaching, and Research Unit in Beef Cattle of the Department of Animal Science, located about 1.4 km from the experimental feedlot. Following weaning, all heifers were housed in individual pens with concrete floors, covered feeders, and drinkers. Heifers were randomly assigned to 2 dietary treatments: control (no addition of powdered molasses; n = 6) or addition of 50 g/kg of diet (DM basis) of powdered molasses (n = 6). The experiment lasted for a total of 79 days, including a 14-day adaptation period.

The basal diet (50:50 forage-to-concentrate ratio) consisted of corn silage, sorghum silage, and a concentrate formulated with ground corn, soybean meal, urea, and mineral mixture (Table 1). The diet was formulated to achieve an average daily gain (ADG) of 1.2 kg/day (Valadares Filho et al., 2023). All heifers were fed *ad libitum* twice daily at 0700 and 1600 h, allowing up to 10% in orts (as fed). The molasses was mixed with the concentrate at the time of feed supply.

Sample collections and measurements

Feed intake was measured daily throughout the experiment. Samples of orts and forage were collected daily, stored in plastic bags, and kept at -20°C. Samples of concentrate ingredients were collected directly at the feed mill. Samples of forage and orts were pooled per animal over approximately 21-day periods, oven-dried a 55ºC for 72 h and then processed in a knife mill to pass through a 1-mm screen sieve. After grinding, samples were stored in plastic jars for further analysis.

On day 55 of the experiment, a digestibility trial was carried out over a 5-d period. Briefly, fecal collections were carried out at 0600, 0900, 1200, 1500, and 1800 h on days 1, 2, 3, 4, and 5 of the trial, respectively. Fecal samples were collected immediately after defecation or directly from the rectum of the animals. Samples were placed in aluminum trays, oven-dried at 55°C for 72 h and then processed to pass through a 1-mm screen sieve. Fecal samples were then pooled per animal and stored as previously described.

On days 3 and 5 of the digestibility trial, urine samples were collected by stimulating the sub vulvar region at the same times as the previously described fecal collection. An aliquot of approximately 50 mL was filtered through four layers of cheesecloth and divided into two samples. The first one consisted of approximately 10 mL of urine diluted in 40 mL of 0.036 N H_2SO_4 for the analysis of purine derivatives. The second one consisted of approximately 40 mL of urine without the addition of acid for the quantification of nitrogen (N) content. Both samples were stored at -20°C for subsequent analysis.

On days 68 and 76 of the experiment, ruminal fluid was collected before feeding and 4 hours after feeding using an oral probe with the aid of a vacuum pump (BP-600.4; Milan Scientific Equipment, Colombo, PR, Brazil). The initial jets of ruminal fluid were discarded to minimize saliva contamination. The ruminal fluid was then filtered through four layers of cheesecloth, and the pH was immediately measured using a digital potentiometer. Subsequently, approximately 2 mL of ruminal fluid was transferred to microfuge tubes and stored at -80°C for VFA analysis.

All heifers were weighed at the beginning and end of the experiment, after 16-hour solids fasting evaluate performance characteristics. Subsequently, the heifers were slaughtered at the slaughterhouse of the Department of Animal Science of UFV, following Normative

Instruction No. 9,013/MAPA 2017. Briefly, the slaughter was carried out via captive bolt stunning followed by exsanguination. The non-carcass components were separated and weighted. The gastrointestinal tract (i.e., rumen, reticulum, omasum, abomasum, and small and large intestines) was emptied and washed. The carcass was then split into two halves along the spine, and the hot carcass weight (HCW) was recorded. The halves were chilled at 4°C for 24h. After 24h, both halves were weighted again obtain the cold carcass weight (CCW). The ribeye area (REA) was measured at 12^{th} rib by using a geometric method. The subcutaneous fat thickness (SFT) was measured using an electronic caliper.

Rumen samples of approximately 5×5 cm were excised from the *Saccus ventralis*, thoroughly washed with 0.9% NaCl solution, and fixed in 10% formalin. After 24 h, the samples were transferred to tubes plastics containing 70% ethanol and then stored until further analysis. For the rumen analysis, a fragment of rumen tissue from the section that was preserved in ethanol of approximately 1 cm^2 in size taken. The number of papillae per cm^2 was manually counted. The rumen samples were then photographed at 0.3x magnification using a stereomicroscope (Discovery V.20, Zeiss, Germany). The height (from base to apex) and width (measured at the median region) of the papillae were measured using ImageJ software (National Institutes of Health, Bethesda, MD, USA).

Laboratory analysis and calculations

Samples of forage, concentrate feeds, and feces, previously processed to pass through a 1-mm sieve, were analyzed for DM (dried for 16 h at 105 ºC, method G-003/1), ash (complete combustion in a muffle oven at 550ºC, method M-001/2), CP (Kjeldahl procedure, method N-001/2), and NDF (using heat-stable α-amylase and omitting sodium sulfite, method F-012/1), following the procedures described by the Brazilian National Institute of Science and Technology in Animal Science (INCT-CA, Detmann et al., 2021). The indigestible neutral detergent (iNDF) content was obtained through a 288-h *in situ* incubation procedure (Valente et al., 2011).

Ruminal fluid samples were centrifuged (12,000 × g, 10 min, 4◦C) and then the supernatants were processed according to Siegfried et al. (1984). The ruminal volatile fatty acid (VFA) concentration was analyzed via high performance liquid chromatography (HPLC) using a Dionex Ultimate 3000 Dual detector HPLC (Dionex Corporation, Sunnyvale, CA, USA) coupled to a refractive index Shodex RI-101. Acetate, propionate, and butyrate values were expressed as molar proportion of the total VFA concentration. Uric acid and creatinine concentrations were analyzed using an automatic biochemical analyzer (Minudray, model BS200E, Shenzhen, China). Urine allantoin concentration was measured using a colorimetric method according to Chen and Gomes (1992).

Fecal output was obtained as the ratio between the iNDF intake and the iNDF concentration in feces, as follows:

$$FO = \frac{iNDF_{cons.}}{iNDF_{feces}} \quad (1)$$

Where FO is fecal output (kg/day), $iNDF_{cons.}$ is the iNDF intake (kg/day), and $iNDF_{feces}$ is the iNDF concentration in the feces (kg/kg DM).

Urine creatinine excretion was estimated according to the equation proposed by Santos et al. (2023), as follows:

$$UCE = 66.2158 \times BW^{0.8384} \quad (3)$$

Where UCE is urinary creatinine excretion (mg/day), and BW is body weight (kg). Urinary volume was estimated by dividing daily creatinine excretion by urinary creatinine concentration.

Urine N excretion was estimated according to an updated equation proposed by BR CORTE (2023, unpublished data), as follows:

$$NU = 22.62 \times (exp^{0.00669}NI) \quad (2)$$

Where NU is N excreted in the urine (g/day) and NI is N intake (g/day).

Urinary excretion of allantoin and uric acid was measured by the product of the concentration of these compounds in urine and the estimated urinary volume. Total excretion of purine derivatives was estimated by the sum of the urinary excretions of allantoin and uric acid. Absorbed purines were calculated from the excretion of purine derivatives according to Barbosa et al. (2011), as follows:

$$AP = \frac{[\mathrm{PD} - (0.301 \times BW^{0.75})}{0.80} \quad (4)$$

Where AP is the absorbed purines (mmol/d) and PD is the excretion of purine derivatives (mmol/day).

The microbial N compound synthesis was also estimated according to the equation proposed by Barbosa et al. (2011), as follows:

$$NMIC = \frac{70 \times \mathrm{AP}}{0.93 \times 1{,}000 \times 0{,}137} \quad (5)$$

Where NMIC is N compound synthesis (g/day) and AP is the absorbed purines (mmol/d). The NMIC was multiplied by 6.25 to obtain microbial protein synthesis (MPS, g/day). The MPS efficiency was calculated by the ratio between the MCP and digested OM (DOM) and TDN intakes.

Statistical analysis

The data were analyzed according to the following model:

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

Where Y_{ij} is the response measured on the heifer j submitted to treatment i, μ is the overall constant, T_i is the fixed effect of treatments, and $\varepsilon_{(i)j}$ is the random error, assumed ~ iid N(0, σ_ε^2). When pertinent, heifers' initial BW was used as a concomitant variable in the model. If the effect of this variable was found to be non-significant, the model was reparameterized by excluding it.

All variables were tested for assumptions of normal error distribution and homoscedasticity of error variances between treatments using the Shapiro-Wilk and Levene tests, respectively. All analyses were performed using the MIXED procedure of SAS 9.4 considering 0.05 as the critical level of probability for the occurrence of type I error.

RESULTS

Intake and digestibility

There was no effect ($P \geq 0.15$) of molasses inclusion on any of the intake variables (Table 2). The average dry matter intake (DMI) was 6.18 kg/day for the control group and 6.20 kg/day for heifers fed molasses. Similarly, molasses addition had no effect ($P \geq 0.06$) on the total-tract apparent digestibility or total digestible nutrient (TDN) content of the diet (Table 3).

Nitrogen balance and microbial protein synthesis

The inclusion of molasses in the diet did not affect ($P \geq 0.57$) N intake or N excretion in feces and urine (Table 4). Consequently, no significant effects ($P \geq 0.77$) were observed for N retention or utilization efficiency with molasses addition. Similarly, molasses inclusion had no effect ($P \geq 0.48$) on MPS or MPS efficiency.

Rumen characteristics

Providing molasses increased rumen pH compared to control ($P < 0.032$). The addition of molasses did not affect VFA concentration ($P > 0.90$). Conversely, we detected an effect of molasses on VFA molar proportion. Molar proportion of both acetate ($P < 0.013$) and butyrate ($P < 0.012$) were greater in heifers fed molasses than control. On the other hand, dietary molasses addition decreased the propionate molar proportion ($P < 0.001$). This resulted in an increased acetate-to-propionate ratio in heifers fed diets containing molasses ($P < 0.002$).

Molasses addition had no impact ($P \geq 0.59$) on papillae length or width. Likewise, there was no effect ($P > 0.82$) of molasses on the number of ruminal papillae per cm^2.

Performance and carcass characteristics

There was no effect of molasses on final BW ($P > 0.78$), final SBW ($P > 0.71$), or ADG ($P > 0.72$) of the heifers (Table 6). Additionally, no differences ($P > 0.69$) were observed between treatments for feed efficiency. Similarly, dietary molasses addition did not affect hot carcass weight ($P > 0.15$) or cold carcass weight ($P > 0.19$). However, carcass yield tended to be greater (*P* ≤ 0.084) in heifers fed molasses. No effect (*P* > 0.87) of molasses was detected on ribeye area. Conversely, the addition of molasses to the diet increased (*P* < 0.024) subcutaneous fat thickness by 63.4% compared to the control group.

DISCUSSION

In our study, dietary molasses addition did not affect the voluntary intake of the heifers. This finding contrasts with several reports that suggest adding molasses to cattle diets generally increases voluntary intake (Pate, 1983; Ciriaco et al., 2015; Silva et al., 2023). Such results are primarily due to its high palatability (Murphy et al., 1997). However, the impact of molasses on voluntary intake seems to be typically more pronounced when it is added to low-quality forages (Bowman et al., 1995; Titgemeyer et al., 2004). Under such conditions, molasses may enhance voluntary intake through two primary mechanisms. First, by improving the palatability of low-quality fibrous feeds, molasses can make these feeds more acceptable to the animals (Mordenti et al., 2021). Second, molasses can promote microbial growth on the forage fibrous fraction by providing soluble sugars as substrates, which in turn enhances the digestibility of insoluble fiber and thus increases voluntary intake (Oba et al., 2011; Havekes et al., 2021). These effects are often amplified when molasses is used in combination with non-protein nitrogen sources as urea (Moriel et al., 2018; Lopes et al., 2019; Abreu et al., 2022). However, the diets in our study included moderate levels of corn and sorghum silage, which are of greater quality compared to the forages used in the aforementioned studies. This difference in forage quality may explain why molasses did not affect voluntary intake. Additionally, our results are consistent with other studies where molasses addition did not affect the voluntary intake of beef steers in feedlot (Felix et al., 2018).

In agreement with the intake results, we did not observe any effect of molasses addition on diet digestibility. The impact of molasses on digestibility in cattle diets is quite variable, with results differing across studies (Broderick et al., 2004; Abreu et al., 2022; Torres et al., 2023). These variations can largely be attributed to the level of molasses inclusion, its combination with other ingredients, and the characteristics of the basal diet. For instance, some studies have shown that molasses supplementation alone can reduce fiber digestibility in cattle diets, especially when forage-based diets are used (Kalmbacher et al., 1995; Torres et al., 2023). This effect is often linked to decreased ruminal pH and increased competition between fibrolytic and aminolytic bacteria (Royes et al., 2001). Conversely, other studies have demonstrated that adding molasses to cattle diets in combination with non-protein nitrogen sources can improve the digestibility of insoluble fiber, particularly in low-quality forages (Bowman et al., 1995; Ciriaco et al., 2015). In our study, the likely of molasses affecting the digestion of the fibrous fraction was low, as the diets provided sufficient nutrients and nutritional attributes to support optimal microbial growth and ruminal degradation.

Overall, we did not observe any effect of molasses addition on N balance or microbial protein synthesis. Adding molasses to cattle diets can enhance N retention and improve the efficiency of N utilization (Broderick et al., 2004). This improvement is believed to result from better N uptake by ruminal microorganisms, achieved by optimizing the synchronization between carbohydrates and protein. This synchronization could potentially reduce N excretion in urine and feces, thereby increasing N retention and efficiency (Broderick et al., 2004; Hristov et al., 2005; Brito et al., 2017).

However, improvements in N retention are closely linked to N intake. In our study, molasses did not affect N intake. This could explain the lack of impact on N retention and efficiency of N utilization. It is worth noting that heifers in both groups exhibited basically the same efficiency of N utilization. In other words, the molasses inclusion did not impact the efficiency, it is possible that the diet and genetic group utilized in our study contributed to these results. Since they are highly efficient animals, the inclusion of molasses alone was not enough to show a difference in N use in any group.

Similar to the results for N retention and utilization efficiency, the addition of molasses did not impact microbial protein synthesis or its efficiency. We expected that molasses supplementation would enhance microbial protein synthesis by increasing N uptake by rumen microorganisms. Indeed, some studies have suggested that adding molasses to cattle diets boosts microbial protein synthesis and its flow to the intestine (Huhtanen, 1988; Broderick et al., 2004). However, these studies also reported an increase in voluntary intake, which likely contributed, at least partially, to the observed rise in microbial protein synthesis.

In our study, two main factors could explain the lack of effect of molasses on microbial protein synthesis and efficiency. First, molasses did not alter voluntary intake, a major factor influencing microbial protein synthesis. Second, the absence of an effect on microbial protein production suggests that the diets already provided sufficient basal conditions for substrate assimilation by the microorganisms. This is often the case with medium- to high-quality forages, which typically meet the degradable protein requirements of ruminal microorganisms, thereby limiting the response to additional concentrate ingredients (Titgemeyer et al., 2004).

One of the primary objectives of this study was to enhance butyrate production through the addition of molasses to the diet, with the aim of improving rumen epithelial cells in newly weaned heifers. In line with this goal, we observed that molasses supplementation increased the proportions of acetate and butyrate while decreasing the proportion of propionate. This shift in fermentation profile was further supported by an increase in ruminal pH among the heifers fed molasses. These findings are consistent with numerous studies showing that molasses supplementation increases ruminal butyrate concentrations in beef cattle (Ciriaco et al., 2015; Stierwalt et al., 2017; Silva et al., 2023).

On the other hand, we did not observe any effect of molasses addition on the length, width, or number of ruminal papillae. This result was somewhat unexpected, as increased butyrate production is typically associated with enhanced papillae development in cattle (Górka et al., 2009; Fukumori et al., 2022) and sheep (Liu et al., 2019). A likely explanation for the lack of effect on rumen development may be related to the level of stress experienced by the animals. Generally, the benefits of molasses or butyrate supplementation on gastrointestinal tract development are more pronounced in animals under high-stress conditions (Silva et al., 2023), such as during long transportation, receiving protocols, or dietary changes. In this study, however, the heifers were likely not exposed to significant stress. This can be attributed to the short transportation distance, proper handling during the receiving process, and the fact that the heifers came from the same herd, where social hierarchies were already established. These factors may have contributed to the absence of a measurable effect on ruminal papillae development.

Corroborating the previous findings, we did not observe any effect of molasses on performance variables. The impact of molasses on animal performance is generally inconsistent, with some studies reporting improvements (Silva et al., 2023; Felix et al., 2018), no change (Salinas-Chavira et al., 2018), or even a decrease in performance (Torres et al., 2023) when molasses is added to cattle diets. Typically, improvements in animal performance with molasses supplementation have been attributed to increases in DMI, diet digestibility, and microbial protein synthesis (Mordenti et al., 2021). However, as highlighted earlier, we did not observe any significant effects of molasses on these characteristics, which likely explains the lack of impact on overall performance. It is important to note that the effects of molasses on animal performance are more pronounced in diets based on low-quality forages (Royes et al., 2000; Ciriaco et al., 2015), a scenario that differs significantly from the conditions of the present study.

Finally, we observed that the addition of molasses to the diet increased subcutaneous fat thickness in the carcass compared to the control group. There is generally limited data on the effects of molasses on carcass characteristics in cattle. A recent meta-analysis found that adding up to 150 g/kg DM of molasses to the diet had no significant effect on carcass characteristics (Torres et al., 2023). To our knowledge, the increased subcutaneous fat thickness may be directly linked to the higher molar proportion of acetate in heifers receiving molasses. This increase in subcutaneous fat could offer practical benefits, such as better carcass protection during freezing, potentially improving meat quality. Nonetheless, further research is needed to fully understand the effects of molasses on carcass and meat characteristics.

In summary, our data suggest that the addition of molasses to the diets of newly weaned beef heifers in the finishing phase did not affect intake or performance characteristics. While molasses supplementation did increase the molar proportion of butyrate, this increase did not translate into positive effects on rumen epithelial cells. However, the addition of molasses appears to have influenced carcass fat thickness, which could enhance carcass preservation during freezing.

CONCLUSÃO

Providing sugarcane molasses does not affect the intake, digestion characteristics, and performance of young beef finishing heifers fed with medium quality diets. While the inclusion of molasses increases the molar proportion of butyrate, it does not promote rumen epithelial cell development. However, the addition of molasses appears to affect carcass fat thickness, which could enhance carcass preservation during process freezing.

REFERENCES

Abreu, D., Dubeux, J.C.B., Jr., Queiroz, L.D., Jaramillo, D., Da Silva Santos, E.R., Van Cleef, F., Vela-Garcia, C., DiLorenzo, N., Ruiz-Moreno, M. 2022. Supplementation of Molasses-Based Liquid Feed for Cattle Fed on Limpograss Hay. Animals. 12, 2227.

Barbosa, A.M., Valadares, R.F.D., Valadares Filho, S.C., Pina, D.S., Detmann, E., Leão, M.I., 2011. Endogenous fraction and urinary recovery of purine derivatives obtained by different methods in Nellore cattle. J. Anim. Sci. 89, 510-519.

Bowman, J.G.P., Sowell, B.F., Paterson, J.A. Liquid supplementation for ruminants fed low quality forage diets: A review. Anim. Feed Sci. Technol. 55, 105-138.

Brito, A.F., Soder, K.J., Chouinard, P.Y., Reis, S.F., Ross, S., Rubano, M.D., Casler, M.D. 2017. Production performance and milk fatty acid profile in grazing dairy cows offered ground corn or liquid molasses as the sole supplemental nonstructural carbohydrate source. J. Dairy Sci. 100, 8146-8160.

Broderick, G.A., Radloff, W.J. 2004. Effect of Molasses Supplementation on the Production of Lactating Dairy Cows Fed Diets Based on Alfalfa and Corn Silage. J. Dairy Sci. 87, 2997-3009.

Brown, W. F.; Johnson, D. D. 1991. Effects of energy and protein supplementation of ammoniated tropical grass hay on the growth and carcass characteristics of cull cows. J. Anim. Sci., 69, 348-357.

Chen, X.B., Gomes, M.J. 1992. Estim. Microb. Protein Supply Sheep Cattle Based Urin. Excretion Purine Deriv. Overv. Tech. Details 20.

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. 20.

Ciriaco, F.M., Henry, D.D., Mercadante, V.R., Schulmeister, T., Ruiz-Moreno M., Lamb G. C., DiLorenzo, N. 2015. Effects of different levels of supplementation of a 50:50 mixture of molasses:crude glycerol on performance, Bermuda grass hay intake, and nutrient digestibility of beef cattle. J. Anim. Sci. 93:2428-2438.

Detmann, E., Costa e Silva, L.F., Rocha, G.C., Palma, M.N.N., Rodrigues, J.P.P. 2021. Métodos para análise de alimentos. Second edition. Suprema, Visconde do Rio Branco, Minas Gerais.

Felix T.L., Long, C.J., Stierwalt, M.R., Carvalho, P.H.V., Blalock, H.M. 2018. Effects of increasing dietary level of a commercial liquid supplement on growth performance and carcass characteristics in feedlot steers. Transl. Anim Sci. 18, 216-222.

Górka, P., B. Śliwiński, J., Flaga, J., Wieczorek, M.M., Godlewski, E., Wierzchoś, R., Zabielski., Kowalski, Z.M. 2017. Effect of butyrate infusion into the rumen on butyrate flow to the duodenum, selected gene expression in the duodenum epithelium, and nutrient digestion in sheep. J. Anim. Sci. 95, 2144-2155.

Gorka, P., Kowalski, Z.M., Pietrzak, P., Kotunia, A., Kiljanczyk, R., Flaga, J., Holst, J.J., Guilloteau, P., Zabielski, R. 2009. Effect of sodium butyrate supplementation in milk replacer and starter diet on rumen development in calves. J. Physiol. Pharmacol. 2009. 60, 47-53.

Havekes, C., Duffield, T., Carpenter, A., DeVries, T. 2020. Effects of Molasses-Based Liquid Feed Supplementation to A High-Straw Dry Cow Diet on Feed Intake, Health, And Performance of Dairy Cows Across the Transition Period. J. Dairy Sci. 103, 5070-5089.

Hristov, A.N., Ropp, J.K., Grandeen, K.L., Abedi, S., Etter, R.P., Melgar, A., Foley, A.E. 2005. Effect of carbohydrate source on ammonia utilization in lactating dairy cows. J. Anim. Sci. 83, 408-421.

Huhtanen, P. 1988. The effect of barley, unmolassed sugar-beet pulp and molasses supplements on organic matter, nitrogen and fibre digestion in the rumen of cattle given a silage diet. Anim. Feed Sci. Technol. 20, 259-278.

Kalmbacher, R.S., Brown, W.F., Pate, F.M. 1995. Effect of molasses-based liquid supplements on digestibility of creeping bluestem and performance of mature cows on winter range. J. Anim. Sci. 73:853-860

Kos, D., Schreiner, B., Thiessen, S., McAllister, T., Jelinski, M., Ruzzini, A. 2023. Insight into antimicrobial resistance at a new beef cattle feedlot in western Canada. Msphere. 8, 00317-23.

Lesmeister, K. E.; Heinrichs, A. J. 2005. Effects of adding extra molasses to a texturized calf starter on rumen development, growth characteristics, and blood parameters in neonatal dairy calves. J. Dairy Sci. 88, 411-418.

Liu, L. Sun, D, Mao, S., Zhu, W., Liu, J. 2019. Infusion of sodium butyrate promotes rumen papillae growth and enhances expression of genes related to rumen epithelial VFA uptake and metabolism in neonatal twin lambs. J. Anim. Sci. 97, 909-921.

Lopes, H.M., Lima, M.L.P., Zilião, G.C., Negrão, J.A., Simili, F.F., Pires, B.V. and El Faro, L., 2019. Performance of Nellore females in tropical pastures during the rainy season when supplemented with molasses blocks. Semina: Cienc. Agrar. 40, 1991-2006.

Mordenti, A.L., Giaretta, E., Campidonico, L., Parazza, P., Formigoni, A. 2021. A Review Regarding the Use of Molasses in Animal Nutrition. Animals, 11, 115.

Moriel, P., Vendramini, J.M., Carnelos, C., Piccolo, M.B. and da Silva, H.M., 2019. Effects of monensin on growth performance of beef heifers consuming warm-season perennial grass and supplemented with sugarcane molasses. Trop. Anim. Health Prod. 51, 339-344.

Murphy, J.J. 1999. The effects of increasing the proportion of molasses in the diet of milking dairy cows on milk production and composition. Anim. Feed Sci. Technol. 78, 189-198.

Oba, M. 2011. Review: Effects of Feeding Sugars on Productivity of Lactating Dairy Cows. Can. J. Anim. Sci. 91, 37-46.

Pate, F. M. 1983. Molasses in beef nutrition. Molasses in animal nutrition. West Des Moines (IA): National Feed Ingredients Association; p. 1-57.

Reinhardt, C., Thomson, D.U. 2015. Nutrition of newly received feedlot cattle. Vet. Clinics: Food Anim. Pract. 31, 283-294.

Royes, J.B., Brown, W.F., Martin, F. G., Bates, D.B. 2001. Source and level of energy supplementation for yearling cattle fed ammoniated hay. J. Anim. Sci. 79:1313-1321.

Salinas-Chavira, J., Montano-Gomez, M.F., Navarrete, J.D., Raymundo, C., Torrentera, N.G., Zinn, R.A. 2018. Influence of mixing supplements (dry or liquid) with monensin or soluble protein on the feeding value of finishing diets for feedlot cattle. J. Appl. Anim. Res. 46:734-739

Santos, S.A., Rotta, P.P., Costa e Silva, L.F., Menezes, A.C.B., Valadares Filho, S.C. Ruminal protein degradation of feeds and microbial protein synthesis. 2023. In: Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Suprema Grafica Ltda, Visconde do Rio Branco, 299-328.

Siegfried, B.R., Ruckemann, H., Stumpf, G. 1984. Method for the determination of organic acids in silage by high performance liquid chromatography. Landwirtsch. Forsch. 37, 298-304.

Silva, B.C., Godoi, L.A., Supapong, C., Bitsie, B., Valadares Filho, S.C., Schoonmaker, J.P. 2023. Effect of a molasses-based liquid supplement on gastrointestinal tract barrier function, inflammation, and performance of newly received feedlot cattle before and after a transport stress. J. Anim. Science. 101, 1-11.

Stierwalt, M.R., Blalock, H.M., Felix, T.L. 2017. Effects of the interaction of forage and supplement type on digestibility and ruminal fermentation of beef cattle. J. Anim. Sci. 95, 1-9.

Titgemeyer, E.C., Drouillard, J.S., Greenwood, R.H., Ringler, J.W., Bindel, D.J, Hunter, R.D., Nutsch, T. 2004. Effect of forage quality on digestion and performance responses of cattle to supplementation with cooked molasses blocks. J Anim. Sci. 2004 82, 487-494.

Torres N.S.R., Bertoco, J.P.A., de Arruda, M.C.G., Coelho, L. M., Paschoaloto, J.R., Neto, O.R.M., Ezequiel, J.M.B., Almeida, M.T.C., Chardulo, L.A.L., Baldassini, W.A. 2023. Effects of molasses supplementation on animal performance and carcass parameters of beef cattle: a meta-analysis. Trop. Anim. Health Prod. 55, 143.

Valadares Filho, S.C., Saraiva, D.T., Benedeti. P.B., Silva, F.A.S., Chizzotti, M.L. 2023. Nutrient requirements of Zebu and crossbred cattle - BR-CORTE. Third revised edition. Suprema Grafica Ltda, Visconde do Rio Branco, Minas Gerais.

Valente, T.N.P., Detmann, E., Queiroz, A.C., Valadares Filho, S.C., Gomes, D.I., Figueiras, J.F. 2011. Evaluation of ruminal degradation profiles of forages using bags made from different textiles. Rev. Bras. Zootec. 40, 2565–2573.

TABLES

Table 1. Ingredients and composition of the experimental diets

Item[a]	Control	Molasses
Ingredients		
Corn silage	269	268
Sorghum silage	226	226
Corn ground	445	401
Soybean meal	25.9	25.8
Molasses	-	53.4
Urea	12.7	14.2
Mineral mixture	11.6	11.6
Chemical composition		
Dry matter, g/kg as fed	425	427
Organic matter	949	940
Crude protein	113	114
NDF	336	357
iNDF	118	116

[a] NDF, neutral detergent fiber; iNDF, indigestible neutral detergent fiber.

Table 2. Effects of sugar cane molasses on voluntary intake of young beef finishing heifers.

Item[a]	Treatments[b]		SEM	P-value
	Control	Molasses		
Intake, kg/day				
Dry matter	6.18	6.20	0.170	0.95
Organic matter	5.87	5.83	0.160	0.86
Crude protein	0.708	0.712	0.0188	0.90
Ether extract	0.170	0.161	0.0044	0.15
NDF	2.28	2.26	0.064	0.84
NDFi	0.727	0.730	0.0210	0.91
NFC	2.84	2.87	0.077	0.78
DOM	4.26	4.44	0.132	0.34
TDN	4.58	4.80	0.142	0.28
Intake, g/kg BW				
Dry matter	22.7	22.5	0.65	0.84
Crude protein	2.60	2.59	0.073	0.93
NDF	8.38	8.22	0.242	0.65
iNDF	2.68	2.65	0.080	0.80

[a] [ap]NDF, neutral detergent fiber; iNDF, indigestible neutral detergent fiber; NFC, non-fibrous carbohydrates; DOM, digested organic matter; TDN, total digestible nutrients. [b] Control, no addition of sugar cane molasses to the diet; Molasses, addition of 50 g/kg DM sugar cane molasses to the diet.

Table 3. Effects of sugar cane molasses on apparent digestibility and diet energy content in young beef finishing heifers

Item[a]	Treatments[b]		SEM	P-value
	Control	Molasses		
g/kg				
Dry matter	671	689	9.9	0.23
Organic matter	701	725	10.4	0.13
Crude protein	653	646	12.6	0.73
NDF	443	474	15.3	0.18
NFC	915	939	8.2	0.060
TDN	716	738	9.6	0.14

[ap] NDF, neutral detergent fiber, NFC, non-fibrous carbohydrates; TDN, total digestible nutrients.

[b] Control, no addition of sugar cane molasses to the diet; Molasses, addition of 50 g/kg DM sugar cane molasses to the diet.

Table 4. Effects of sugar cane molasses on nitrogen balance and microbial protein synthesis in young beef finishing heifers

Item[a]	Treatments[b]		SEM	P-value
	Control	Molasses		
N intake, g/d	118	120	3.5	0.60
Fecal N, g/d	41.1	42.5	2.11	0.63
Urine N, g/d	49.8	50.8	1.20	0.57
Retained N, g/d	26.9	27	1.64	0.92
EUN, g/g N intake	0.230	0.224	0.0123	0.77
MPS, g/d	279	270	18.7	0.72
MPS efficiency, g MPS/ kg DOM	65.5	61.1	4.59	0.51
MPS efficiency, g MPS/ kg TDN	61.0	56.6	4.29	0.48

[a] EUN, efficiency of N utilization; MPS, microbial protein synthesis, DOM, digested organic matter.

[b] Control, no addition of sugar cane molasses to the diet; Molasses, addition of 50 g/kg DM sugar cane molasses to the diet.

Table 5. Effects of sugar cane molasses on rumen characteristics in young beef finishing heifers

Item[a]	Treatments[b]		SEM	P-value
	Control	Molasses		
Papillae characteristics				
Number of papillae	67.8	70.3	7.82	0.82
Length papillae, mm	2.58	2.51	0.527	0.92
Width papillae, mm	0.980	0.913	0.0862	0.59
Fermentation characteristics				
Rumen pH	7.09	7.21	0.035	0.032
Total VFA, mmol/L	36.1	35.8	1.42	0.90
VFA, mmol/mmol				
Acetate	0.546	0.572	0.0061	0.013
Propionate	0.352	0.314	0.0057	0.001
Butyrate	0.103	0.115	0.0029	0.012
Acetate:propionate	1.55	1.83	0.049	0.002

[a] VFA, volatile fatty acids.

[b] Control, no addition of sugar cane molasses to the diet; Molasses, addition of 50 g/kg DM sugar cane molasses to the diet.

Table 6. Effects of sugar cane molasses on performance of young beef finishing heifers

Item[a]	Treatments[b]		SEM	P-value
	Control	Molasses		
Performance				
Initial BW, kg	237	239	7.62	0.88
Final BW, kg	310	311	3.75	0.78
Final EBW, kg	263	264	3.2	0.71
ADG, kg/day	1.12	1.15	0.056	0.72
Gain:feed intake, kg/kg	0.182	0.185	0.0056	0.69
Carcass characteristics				
Hot carcass weight, kg	161	165	2.3	0.24
Cold carcass weight, kg	152	157	2.1	0.11
Hot carcass yield, kg/kg	0.518	0.534	0.0065	0.084
Cold carcass yield, kg/kg	0.488	0.509	0.0069	0.057
Ribeye area, cm^2	26.5	26.3	0.98	0.87
SFT, mm	3.98	6.28	0.614	0.024

[a] BW, body weight; EBW, empty body weight; ADG, average daily gain; SFT, subcutaneous fat thickness.

[b] Control, no addition of sugar cane molasses to the diet; Molasses, addition of 50 g/kg DM sugar cane molasses to the diet.