SIDNEI ANTÔNIO LOPES

ESTRATÉGIAS PARA OTIMIZAÇÃO DA PERFORMANCE DE BOVINOS EM PASTAGENS TROPICAIS

Tese apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Zootecnia, para obtenção do título de Doctor Scientiae.

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Aos meus familiares,
Aos meus amigos

Dedico.

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SUMÁRIO

RESUMO

LOPES, Sidnei Antônio, D.Sc., Universidade Federal de Viçosa, Outubro de 2015. **Estratégias para otimização da performance de bovinos em pastagens tropicais.** Orientador: Mário Fonseca Paulino. Coorientadores: Edenio Detmann, Sebastião de Campos Valadares Filho e Luciana Navajas Rennó.

Foram elaborados três artigos científicos envolvendo avaliações nutricionais e produtivas de bovinos de corte em pastejo, submetidos a diferentes estratégias de suplementação. No primeiro artigo, objetivou-se avaliar os efeitos da suplementação com diferentes teores de proteína sobre desempenho nutricional e produtivo de vacas de corte em pastejo durante o pós-parto. Foram utilizadas 36 vacas de corte com idade e peso corporal médio de 5 anos e 490 ± 17,9 kg, respectivamente. O delineamento experimental foi inteiramente casualizado. Os tratamentos foram: controle = vacas receberam somente mistura mineral ad libitum; suplementados = vacas receberam 1 kg/dia de suplemento contendo 80, 200 ou 320 g de proteína bruta (PB)/kg. Não houve efeito ($P \geq 0,16$) da suplementação sobre consumo voluntario. Entre os animais suplementados, o consumo de PB aumentou linearmente ($P < 0,02$) com teor de PB no suplemento. A suplementação não afetou ($P \geq 0,20$) digestibilidade total da matéria orgânica, fibra em detergente neutro corrigida para cinza e proteína (FDNcp) e da PB. Entre os animais suplementados, houve efeito linear positivo ($P < 0,01$) dos teores de PB nos suplementos sobre a digestibilidade da PB. O fluxo intestinal de compostos nitrogenados microbianos e a eficiência de síntese de proteína microbiana não foram afetados ($P \geq 0,18$) pelos tratamentos. O desempenho, produção e composição do leite não foram afetados ($P \geq 0,11$) pelos tratamentos. A suplementação não afetou ($P \geq 0,10$) as concentrações séricas de ácidos graxos esterificados, ureia e progesterona. Conclui-se que a suplementação de vacas de corte em pastejo durante pós-parto não afeta o consumo e o desempenho produtivo. No segundo artigo, objetivou-se avaliar os efeitos de diferentes quantidades de suplemento sobre o desempenho nutricional e produtivo, e características comportamentais de bezerros de corte lactentes, e o desempenho produtivo de suas mães em pastagem tropical. Foram utilizados 44 bezerros de corte machos da raça Nelore com idade e peso corporal inicial médio de 120 dias e 145 ±

3,7 kg respectivamente, e suas respectivas mães, com peso corporal médio de 449 ± 6,9 kg. O delineamento experimental foi inteiramente casualizado. As quantidades de suplemento avaliadas foram as seguintes: 0 = bezerros receberam somente mistura mineral ad libitum; 3, 6 e 9 = bezerros receberam 3, 6 ou 9 g/kg PC de suplemento, contendo 25 g de PB/kg. Foi observado efeito linear positivo para o consumo de MS e MO ($P < 0,01$). O consumo de FDNcp apresentou comportamento cúbico ($P < 0,07$). Houve diferença ($P < 0,06$) na digestibilidade total da FDNcp. No tocante ao desempenho, efeito cúbico foi observado para o desempenho dos bezerros ($P < 0,01$). Contudo, a suplementação dos bezerros não afetou ($P \geq 0,21$) a produção de leite e o desempenho de suas mães. A suplementação diminuiu o tempo de pastejo ($P < 0,01$), mas não influenciou o tempo de amamentação ($P \geq 0,59$) dos bezerros. Recomenda-se o fornecimento de suplemento contendo 25 g PB/kg na quantidade de 6 g/kg PC para bezerros de corte lactentes manejados em pastagem tropical. A suplementação de bezerros de corte lactentes aumenta o consumo de matéria seca, diminui o tempo de pastejo e o consumo de forragem. Contudo, não influencia o tempo de amamentação e o desempenho produtivo de suas mães. No terceiro artigo, objetivou-se avaliar os efeitos da suplementação de bezerros de corte em sistema de creep feeding sobre a produção de leite, PC, ECC de matrizes de corte em pastagem tropical usando uma abordagem meta-analítica. O banco de dados foi obtido a partir de 11 experimentos conduzidos entre 2009 e 2014 no Brasil, totalizando 485 observações. O banco de dados foi composto por 273 vacas Nelore e 212 mestiças ($^{7}/_{8}$ Nelore × $^{1}/_{8}$ holandês). Todos os experimentos foram conduzidos na fase de amamentação dos três aos oito meses de idade durante a fase de transição chuva-secas (fevereiro a junho de cada ano). Os dados foram analisados por meio de meta-análise, utilizando os procedimentos do MIXED, considerando aleatórias as variações entre experimentos. Todas as análises estatísticas foram realizadas utilizando 0,05 para a ocorrência do erro tipo I. A suplementação ($P \geq 0.59$) e o sexo ($P \geq 0.48$) dos bezerros não afetaram a produção de leite das vacas. A produção média de leite foi de 6,71 e 6,83 kg/dia para vacas que tiveram suas crias suplementadas e não suplementadas, respectivamente. Foram observadas diferenças ($P < 0.0001$) na produção de leite devido ao grupo genético, onde vacas mestiças apresentaram maior produção de leite (7,37 kg/dia) comparada com vacas Nelore

(6,17 kg/dia). Não houve efeito da suplementação sobre o PC ($P \geq 0.11$) e ECC ($P \geq 0.23$) das vacas. Conclui-se que a suplementação de bezerros de corte utilizando creep feeding em pastagem tropical, não afeta a produção de leite, desempenho e a condição corporal de suas mães.

ABSTRACT

LOPES, Sidnei Antônio, D.Sc., Universidade Federal de Viçosa, October, 2015. **Strategies to optimization of performance of cattle on the tropical pasture.** Adviser: Mário Fonseca Paulino. Co-advisers: Edenio Detmann, Sebastião de Campos Valadares Filho and Luciana Navajas Rennó.

It was elaborated three manuscripts relate to nutritional and productive evaluations of beef cattle on grazing, submitted to different supplementation strategies. In the first manuscript the effects of supplementation with different crude protein contents on nutritional and productive performance of grazing beef cows during post-calving were assessed. Thirty-six beef cows, with age and average body weight of 5 years and 490 ± 17.9 kg, respectively, were used. The experimental design was completely randomized. The treatments were: control = cows received only mineral mixture ad libitum; supplemented = cows received 1 kg/d of supplement containing 80, 200, or 320 g crude protein (CP)/ kg. There was no effect ($P \geq 0.16$) of supplementation on voluntary intake. A linear effect ($P < 0.02$) of the CP content in the supplements was observed among supplemented cows, only for the CP intake. Supplementation did not affect ($P \geq 0.20$) the total digestibility of organic matter, neutral detergent fiber corrected for ash and protein, and CP. Among supplemented cows, a positive linear effect ($P < 0.01$) of the CP content in the supplement was observed for the CP digestibility. Intestinal flow of microbial nitrogen compounds and efficiency of microbial synthesis were not affected ($P \geq 0.18$) by treatments. Performance, milk yield and composition were not also affected ($P \geq 0.11$) by treatments. Supplementation did not affect ($P \geq 0.52$) non-esterified fatty acids, urea nitrogen and progesterone serum concentrations. It is concluded that supplementation of grazing beef cows during post-calving does not affect nutritional and productive performance. In the second manuscript, the effects of different amounts of supplement on the productive performance and behavioral characteristics of suckling beef calves, as well as the productive performance of their dams on tropical pastures were assessed. Forty-four male Nellore beef calves with an average age of 120 days and an initial average body weight (BW) of 145 ± 3.7 kg and their respective dams,

with an average BW of 449 ± 6.9 kg, were used. The amounts of supplement evaluated were as follows: 0 = calves received only mineral mixture ad libitum; 3, 6, and 9 = calves received 3, 6, or 9 g/kg BW of supplement, respectively, containing 250 g CP/kg. The experimental design was completely randomized. A linear effect ($P < 0.01$) was observed in the intakes of dry matter and organic matter, and a cubic effect ($P < 0.07$) was observed for the intake of neutral detergent fiber corrected for ash and protein. There was difference ($P < 0.06$) in total digestibility only for neutral detergent fiber. A cubic effect ($P < 0.01$) was observed for the calves' performance. However, the calves' supplementation did not affect the milk yield and performance ($P \geq 0.21$) of their dams. The supplementation decreased grazing time ($P < 0.01$) but did not influence suckling time ($P \geq 0.59$). It is recommended supplying of supplement containing 250 g CP/kg in amount 6 g/kg BW to suckling beef calves managed in tropical pasture. Supplementation of suckling beef calves increases the intake of dry matter, decreases grazing time and forage intake. However, it does not affect the suckling time and productive performance of their dams. In the third manuscript, aim was to evaluate the effects of beef calves' supplementation in creep feeding system on milk yield, body weight and body condition score of their dams on tropical pastures using a meta-analytical approach. The database was obtained from 11 experiments conducted between 2009 and 2014 in Brazil, totaling 485 observations. The database was composed by 273 Nellore and 212 crossbred ($^{7}/_{8}$ Nellore × $^{1}/_{8}$ Holstein) cows. All experiments were carried out in the suckling phase (from 3 to 8 months of age of calves) during transition phase between rainy and dry seasons from February to June of each year. The data was analyzed using meta-analysis using the mixed procedures, considering random variations among experiments. All statistical evaluations were performed using 0.05 as the critical level for the occurrence of type I error. Calves' supplementation ($P \geq 0.59$) and the calf' sex ($P \geq 0.48$) did not affect milk yield of cows. Average fat corrected milk (FCM) yield was 6.71 and 6.83 kg/d for cows that had their calves supplemented and not supplemented, respectively. Differences were observed ($P < 0.0001$) for milk yield due to the genetic group where crossbred cows presented greater FCM yield (7.37 kg/d) compared to Nellore cows (6.17 kg/d). There was no effect of the calves' supplementation on variation of body weight ($P \geq 0.11$) and variation of body

condition score ($P \geq 0.23$) of the cows. Therefore, we concluded that supplementation of beef calves using creep feeding systems in tropical pastures, does not affect milk yield, performance and body condition of their dams.

INTRODUÇÃO GERAL

Os pastos tropicais constituem a principal fonte de nutrientes para a bovinocultura de corte no Brasil, destacando-se dos demais meios de alimentação pelo baixo custo de produção e alta praticidade (Paulino et al., 2008). Segundo o Anuário da Pecuária Brasileira (Anualpec, 2014), 91% dos animais abatidos são provenientes de sistemas baseados em pastagens. Sendo assim, deve-se enfatizar maneiras para aumentar a proporção de energia proveniente da pastagem que pode ser convertida em produto animal.

As forragens pastejadas devem ser entendidas como recurso nutricional basal de elevada complexidade, uma vez que sua capacidade de fornecimento de substratos para a produção animal varia qualitativa e quantitativamente ao longo do ano em função, principalmente, da influência de variáveis climáticas (Detmann et al., 2010). Logo, os pastos tropicais raramente constituem uma dieta balanceada no sentido que seus constituintes orgânicos e inorgânicos estejam presentes nas concentrações e proporções que satisfaçam as necessidades dos animais. Portanto, os bovinos geralmente sofrem carências múltiplas de proteína, energia, minerais e vitaminas (Paulino et al., 2014). Desta forma, para um programa de produção contínua de carne eficiente e competitivo, torna-se essencial minimizar ou mesmo eliminar os efeitos das carências nutricionais múltiplas no desenvolvimento animal, proporcionando aos mesmos, condições para expressar todo seu mérito genético, durante todo o ano, a fim de alcançar as metas produtivas estabelecidas dentro do sistema.

Neste contexto, os programas de suplementação constituem ferramentas para o provimento de recursos suplementares visando à redução ou eliminação de entraves

nutricionais e/ou metabólicos (meta primária) e o alcance de metas de produção animal (meta secundária) (Detmann et al., 2014). Em síntese, a suplementação de bovinos em pastejo permite inserir uma fonte adicional de nutrientes, refletindo em mudanças no consumo de forragem, na disponibilidade de energia dietética e, consequentemente, no desempenho animal (Paulino et al., 2010).

Entretanto, verifica-se que ainda existem questionamentos sobre a viabilidade técnica e econômica da suplementação nas diversas fases de produção, sobre a quantidade adequada de suplemento a ser utilizado em cada fase e mesmo sobre a resposta produtiva possível de ser alcançada.

A intervenção técnica sempre foi focada no processo final de engorda, de maior visibilidade e de resultado palpável e imediato (Paulino et al., 2014). Contudo, o sucesso dessa etapa depende da qualidade dos animais produzidos na fase de cria. No entanto, até a produção de um bezerro as matrizes de corte demandam grande investimento, sendo que, cabe ao bezerro, o produto desta categoria, gerar o retorno econômico. Assim, por não representar uma forma imediata de geração de renda, a nutrição desta categoria é frequentemente negligenciada, refletindo em baixos índices reprodutivos no rebanho.

De modo geral, os índices reprodutivos da pecuária de corte brasileira ainda são muito baixos. A alta proporção de vacas em anestro no início da estação de monta é uma condição comum em rebanhos de corte e afeta negativamente a eficiência reprodutiva, causando sérios prejuízos à pecuária brasileira (Ribeiro et al., 2009). Nos sistemas extensivos de criação de bovinos de corte, observa-se que cerca de 50% das matrizes não recebem manejo que contribua com melhores índices de fertilidade (Madureira et al., 2006).

Entretanto, para o processo de cria ser considerado eficiente, as vacas precisam produzir um bezerro a cada 12-13 meses (Torres Jr. et al., 2009). Para isto, o retorno da vaca à ciclicidade deve ocorrer até 60 dias pós-parto. Deste modo, a implantação de práticas que possibilitem elevar o número de animais em produção no rebanho de cria assume grande importância para o estabelecimento de bovinocultura de alto desempenho competitiva e sustentável.

A nutrição influencia diretamente a fertilidade dos ruminantes pelo suprimento de nutrientes específicos requeridos nos processos de ovulação, fertilização, sobrevivência embrionária e gestação; e indiretamente, pelo impacto na circulação de hormônios e metabólitos que são requeridos nesses processos (Robinson et al., 2006). A baixa ingestão de proteína durante os períodos pré e pós-parto afeta negativamente o desempenho reprodutivo de vacas de corte (Peixoto & Osório, 2007), sendo fundamental a ingestão de níveis adequados de proteína no final da gestação e no início da lactação.

A avaliação do estado nutricional dos ruminantes de interesse zootécnico por meio da observação da condição corporal constitui medida subjetiva baseada na avaliação visual, classificando-os em função da cobertura muscular e da massa de gordura. Embora, seja considerada medida subjetiva, a avaliação da condição corporal é correlacionada com diversos eventos reprodutivos como: intervalo entre partos, serviços por concepção, produção de leite, peso à desmama, dificuldade de parto e sobrevivência do bezerro (Bischoff et al., 2011). O NRC (1996) recomendou a escala de pontuação de escores de condição corporal (ECC) de 1 a 9, em que 1 corresponde a animais extremamente magros e 9 a animais extremamente gordos, sendo desejáveis vacas com escore mínimo de 5 ao parto.

Adicionalmente, a avaliação do perfil metabólico em vacas de corte constitui abordagem que vem despertando o interesse de pesquisadores. Segundo Contreras (2000), o perfil metabólico pode colaborar no estudo do balanço nutricional dos rebanhos, uma vez que, em algumas situações, os desbalanços nutricionais podem influenciar nas concentrações sanguíneas de alguns metabólitos. Entre os metabólitos mais usados para a avaliação do status nutricional estão a glicose, o beta-hidroxibutirato (BHB), os ácidos graxos não esterificados ou livres (AGL), o nitrogênio uréico e a albumina sérica.

Os níveis sanguíneos de AGL são bastante significativos para avaliar o estado de energia em ruminantes, respondendo rapidamente a mudanças no consumo de alimentos (Peixoto & Osório, 2007). Quando as vacas estão em balanço energético negativo (situação comum durante o pós-parto) aumentam as concentrações sanguíneas de AGL, ureia e β-hidroxibutirato (Sartori & Guardieiro, 2010). Assim, a suplementação estratégica poderia minimizar os efeitos do balanço energético negativo e contribuir para acelerar o reinício da ciclicidade nas vacas pós-parto.

Por outro lado, além da eficiência reprodutiva discutida anteriormente, a exploração da bovinocultura de corte intensiva, caracteriza-se pelo acasalamento das novilhas aos 14-16 meses de idade, e o abate dos machos entre 11 e 16 meses de idade. Assim, ganhos de peso contínuo do nascimento a desmama são fundamentais para o sucesso do sistema (Paulino et al., 2012).

A fase de cria dos bezerros constitui etapa altamente responsiva a práticas de manejo e alimentação. Evidências apontam que na fase pré-desmame verifica-se as maiores taxas de ganho de peso, podendo alcançar em apenas sete meses de vida, de 25 a 50% do peso corporal adulto. Durante as primeiras semanas de vida, o leite é a

principal fonte de energia e nutrientes para os bezerros de corte. Estima-se que para cada 1 kg de ganho de peso corporal seja necessário que o bezerro consuma 6,9 kg de leite (Forster et al., 2010). Porém, a relação entre esses dois fatores diminui bastante de intensidade por volta de 16 semanas de vida (Silva 2000). Mesmo que o aumento na produção de leite permita aumentar o ganho de peso a desmama dos bezerros, não se pode negligenciar que o nível nutricional na maior parte dos sistemas baseados em pastagens é limitante para dar suporte a níveis elevados de produção de leite (Paulino et al., 2012).

Por outro lado, à medida que o animal se desenvolve, as exigências nutricionais aumentam e o consumo de leite diminui. Nos sistemas de produção de bovinos de corte, o leite produzido pelas matrizes não é suficiente para atender as exigências nutricionais dos bezerros a partir dos três meses de idade (Henriques et al., 2011). Adicionalmente, entre o 3º e 4º mês de idade, ocorrem mudanças consideráveis no trato digestório do bezerro e este se transforma efetivamente em animal ruminante (Porto et al., 2009). A partir desta idade, o bezerro se torna cada vez mais dependente do pasto. Entretanto, na maioria dos sistemas de produção brasileiros, estes processos acontecem durante o período de transição águas-seca, período em que ocorre a diminuição na qualidade e quantidade de forragem disponível para o pastejo. Consequentemente, a diferença entre as exigências nutricionais do bezerro e a quantidade de nutrientes supridos pelo leite e pelo pasto tendem a aumentar (Oliveira et al., 2006; Rasby & Niemeyer, 2011), colocando o bezerro em situação desfavorável no tocante ao equilíbrio nutricional.

Assim, nos sistemas intensivos de produção de bovinos, em que se exige maior aporte nutricional, visualiza-se a suplementação dos animais lactentes sob

sistema de creep feeding, que consiste em fornecimento do alimento em local cujo acesso é restrito aos bezerros (Paulino et al., 2012). Tal técnica, ao corrigir e enriquecer a dieta do bezerro pode otimizar o desempenho de cada indivíduo, o que resultaria em maior peso corporal ao desmame ensejando em redução no ciclo de produção.

Ademais, poucos estudos têm avaliado os efeitos do creep feeding sobre a matriz de corte. Souza et al. (2007) observaram que vacas cujos bezerros foram suplementados em creep feeding, tiveram maior peso corporal e escore de condição corporal à desmama comparadas àquelas vacas cujos bezerros não foram suplementados. Tais autores atribuíram essa observação ao aumento do número de mamadas diárias, o que estimulou a maior produção de leite, ensejando em redução da condição corporal das vacas cujos bezerros não foram suplementados. Em contraste, Sampaio et al. (2010) observaram que vacas de corte cujos bezerros receberam suplementação apresentaram maior perda de peso. Por outro lado, Porto et al. (2009) e Valente et al. (2013) não observaram diferenças na produção de leite, escore de condição corporal e peso corporal à desmama das vacas devido à suplementação dos bezerros.

Estudos, portanto, são necessários para avaliar as respostas de bovinos de corte de diferentes categorias submetidos a diferentes planos de suplementação. Desta forma, objetivou-se com este estudo avaliar inter-relações entre planos de suplementação e as respostas de bovinos de corte manejados em pastagens tropicais.

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Evaluation of grazing beef cows receiving supplements with different protein contents

Abstract The aim of this study was to evaluate the effects of supplementation with different crude protein contents on nutritional and productive performance, of grazing beef cows during post-calving. Thirty-six beef cows, with age and average body weight of 5 years and 490 ± 17.9 kg, respectively, were used. The experimental design was completely randomized. The treatments were: control = cows received only mineral mixture ad libitum; supplemented = cows received 1 kg/d of supplement containing 80, 200, or 320 g crude protein (CP)/ kg. There was no effect ($P \geq 0.16$) of supplementation on voluntary intake. A linear effect ($P < 0.02$) of the CP content in the supplements was observed among supplemented cows, only for the CP intake. Supplementation did not affect ($P \geq 0.20$) the total digestibility of organic matter, neutral detergent fiber corrected for ash and protein, and CP. Among supplemented cows, a positive linear effect ($P < 0.01$) of the CP content in the supplement was observed for the CP digestibility. Intestinal flow of microbial nitrogen compounds and efficiency of synthesis microbial were not affected ($P \geq 0.18$) by treatments. Performance, milk yield and composition were not also affected ($P \geq 0.11$) by treatments. Supplementation did not affect ($P \geq 0.52$) non-esterified fatty acids, urea nitrogen and progesterone serum concentrations. It is concluded that supplementation of grazing beef cows during post-calving does not affect nutritional and productive performance.

Keywords: beef cows, intake, Nellore, non-esterified fatty acids, supplementation

Introduction

For an efficient production of beef cattle on pastures, cows need to produce a calf every 12-13 months. However, the cow/calf system is normally conducted using low-quality forages and supplementation of beef cows is still uncommon practice in Brazil (Cabral et al., 2012).

Nutrition influences directly the fertility of ruminants as the supply of specific nutrients is required for the ovulation process, fertilization, embryo survival and development; and indirectly through the impact on the circulation of hormones and metabolites required for those processes (Robinson et al., 2006).

Cows poorly nourished and with low-body condition score will be inefficient in the following breeding season (Cabral et al., 2012). According to Peixoto and Osório (2007), protein intake below the requirements during peripartum period would affect negatively the productive performance of beef cows.

The strategic supplementation of this animal category during certain months of the year could contribute to maintain body condition of beef cows in the post-calving and consequently, improve productive and reproductive performance.

Therefore, the objective of this study was to evaluate the effects of supply supplements with different crude protein contents on nutritional and productive performance of grazing beef cows during post-calving.

Material and methods

All procedures involving animals were approved by the Institutional Committee of Universidade Federal de Viçosa for animal care and use experimentation, process UFV number 43/2014.

Animals, experimental design, and diets

The experiment was conducted at the Universidade Federal de Viçosa, MG, Brazil, (20° 45′ S, 42° 52′ W), between September and November 2012, during transition phase between dry and rainy seasons. The experimental area was located in a hilly region, at altitude of 670 m. Over days measurements, the average minimum and maximum temperatures were, respectively, 13.6°C and 27.8°C in September, 16.0°C and 29.6°C in October, and 18.3°C and 26.5°C in November. The amounts of rainfall were 46.9 mm in September, 98.9 mm in October and 225.3 mm in November.

Thirty-six Nellore beef cows, averaging 5 years-old and 490 ± 17.9 kg of body weight (BW) were used. The treatments were distributed randomly to the cows at calving occurrence. The experimental treatments were: control = cows received only mineral mixture ad libitum; supplemented = cows received 1 kg of supplement containing 80, 200, or 320 g of crude protein (CP)/kg (Table 1), fed once a day at 11h00.

The evaluations started from the first day post-calving. The period between the first and last calving lasted 25 days, consequently, the experiment lasted 85 days. Before calving, all cows were managed into a 30-ha paddock with Brachiaria decumbens where they receiving only mineral mixture ad libitum. After calving, cow-calf pairs were managed into a 40-ha with Brachiaria decumbens, divided in four paddocks of 10-ha each, where there were drinkers and shaded feeders in each paddock and cows received one of the treatments assessed.

To minimize the possible effects of the paddock on the experimental treatments, the cow/calf groups were rotated among the four paddocks every seven days, so that each group stayed on each paddock for the same period.

Experimental procedures and sampling

For performance evaluation, the cows were weighed on the first, thirtieth, and sixtieth days post-calving (always in the morning at 6h30), and the BCS of the cows was evaluated using a scale from 1 to 9, as recommended by the NRC (1996); all evaluations were performed by the same four evaluators.

Forage samples were collected every 28 days for the evaluation of forage availability. In each paddock, four samples of forage were randomly selected using a metallic square (0.5×0.5 m), and forage was cut approximately 1 cm above the ground. Sampling for the qualitative assessment of the forage consumed by the animals was obtained every fourteen days by the hand-plucking method. All the samples were oven-dried (60°C /72 hours) and ground in a Wiley mill (model 3, Arthur H. Thomas, Philadelphia, PA) to pass through a 2-mm screen. After that, half of each ground sample was ground again to pass through a 1-mm screen.

Twenty days after the last calving, a nine-day assay was performed to evaluate voluntary intake and digestibility. Chromium oxide (Cr_2O_3), used to estimate fecal excretion, was packaged in paper cartridges in the amount of 20 g per cow/d, and was introduced (11h00) into the esophagus via a rubber tube; titanium dioxide (TiO_2), used to estimate individual supplement intake, was mixed with the supplement distributed to the cows in an amount equal to 15 g per animal/d. The first six days were used to stabilize the flow of markers in gastrointestinal tract of the

96 animals, while the last three days were used for feces collection at 16h00 on day 7, at
97 11h00 on day 8, and at 6h00 on day 9. The fecal samples were oven-dried (60°C /72
98 hours) and proportionally pooled per animal, then ground in a Wiley mill (model 3,
99 Arthur H. Thomas, Philadelphia, PA) to pass through a 2-mm screen. After that, half
100 of each ground sample was ground again to pass through a 1-mm screen. In the fifth
101 day of the digestibility assay, a sample of forage in each paddock was obtained by
102 the hand-plucking method, to estimate voluntary intake and digestibility.

103 To evaluate the microbial protein production of cows, spot urine samples (10
104 mL) were collected from spontaneous micturition 4 hours after supply of supplement
105 in the 9th day of the digestibility assay. Urine samples were diluted in 40 mL of
106 H_2SO_4 (0.036 N) and frozen (-20°C).

107 To estimate milk yield, cows were milked at days 32 and 62 of the
108 experiment. Cows were separated from their calves at 18h00. At 6h00 of the
109 following day, cows were injected with 2-mL oxytocin (10 IU/mL; Ocitovet®,
110 Brazil) in the mammary artery and immediately milked. The exact time when each
111 cow was milked was recorded, and the milk produced was proportionally converted
112 into a 24-h based production. The milk produced was corrected to 4 % of fat (FCM)
113 according to NRC (2001):

114 $$FCM = 0.4 \times \text{milk yield (kg/d)} + 15 \times \text{fat yield (kg/d)} \quad (1)$$

115 Blood samples were collected on the thirtieth and sixtieth days post-calving
116 via jugular vein puncture, using vacuum tubes with separator gel (BD Vacutainer ®
117 SST II Advance) and were centrifuged at 3000 × g for 15 minutes; the serum was
118 frozen (-20°C) for later analysis.

119

120 Chemical analysis

121 Samples of forage, feces, and supplement processed to pass through 1-mm
122 screen sieves were analyzed according to the standard analytical procedures of the
123 Brazilian National Institute of Science and Technology in Animal Science (INCT-
124 CA; Detmann et al., 2012) for dry matter (DM; INCT-CA method G-003/1), ash
125 (INCT-CA method M-001/1), crude protein (CP; INCT-CA method N-001/1), ether
126 extract (EE; INCT-CA method G-004/1), neutral detergent fiber (NDFap; INCT-CA
127 method F-002/1), using alpha thermostable amylase without addition of sodium
128 sulfite and corrected for ash and protein (NDIP; INCT-CA method N-004/1).
129 Indigestible neutral detergent fiber (iNDF; INCT-CA method F-009/1) was
130 quantified by in situ incubation procedures with F57 bags (Ankom®) for 288 hours
131 in samples processed at 2-mm. Fecal samples were evaluated for the contents of
132 chromium (INCT-CA method M-005/1) and titanium (INCT-CA method M-007/1).
133 Milk samples were analyzed with regards as protein, fat, lactose, and total solids
134 content using infrared spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark).

135 The percentage of potentially digestible DM (DMpd) in the forage samples
136 obtained for evaluation of forage availability was estimated according to Paulino et
137 al. (2008):

138 $$DMpd = 0.98 X (100 - NDF) + (NDF - iNDF) \quad (2)$$

139 where: DMpd = forage content of potentially digestible DM (DM %); 0.98 = true
140 digestible coefficient of cell content; and NDF and iNDF = forage content of NDF
141 and iNDF, respectively (DM %).

142 Individual DM intake of supplement (DMS) was estimated by relation of
143 excretion of TiO_2 in feces and marker concentration in the supplement.

144 The voluntary intake of from forage (DMF) was estimated using an internal
145 iNDF, according to Detmann et al. (2001), using the following equation:

146 $$DMF = \left[(FE \times iNDFfeces) - DMSi \times iNDF\ sup\right] \div iNDFforage \quad (3)$$

147 where FE = fecal excretion (kg/d), iNDFfeces = concentration of iNDF in the feces
148 (kg/kg), DMSi = dry matter supplement intake (kg/day), iNDFSup = concentration of
149 iNDF in the supplement (kg/kg), and iNDF forage = concentration of iNDF in the
150 forage (kg/kg).

151 The total DM intake was calculated by the sum of DMF intake and DMS
152 intake.

153 Samples of serum were analyzed for concentrations of progesterone (P4),
154 non-esterified fatty acids (NEFA), and serum urea nitrogen (SUN) by
155 chemiluminescence, enzymatic spectrophotometry, and kinetic fixed time methods,
156 respectively. The analyses were conducted at clinical analyses Laboratory
157 (Viçosalab, Viçosa, MG). Progesterone concentrations higher than 1 ng/ml (P4>1
158 ng/ml) were considered an indicator of ovarian activity (Nogueira et al., 1993).

159 In the urine samples, analyses were performed for creatinine, uric acid, and
160 urea by colorimetric kinetic, enzymatic colorimetric and kinetic fixed time methods,
161 respectively, using automatic biochemical analyzer (Mindray, BS200E model).

162 Daily urinary volume was calculated using the relationship between the daily
163 creatinine excretion (CE), taking as reference the equation proposed by Costa e Silva
164 et al. (2012), and its concentration in the spot samples:

165 $$CE(g/d) = 0.0345 x SBW^{0.9491} \quad (4)$$

166 where: SBW = shrunk body weight

Allantoin was analyzed by the colorimetric method as described by Chen & Gomes (1992). Total excretion of purine derivatives was calculated by the sum of the amounts of allantoin and uric acid excreted in urine.

The purines absorbed were calculated from the excretion of purine derivatives according the equation Barbosa et al. (2011):

$$AP = \frac{(PD - 0.301 x BW^{0,75})}{0.80} \quad (5)$$

where: AP = absorbed purines (mmol/d); PD = excretion of purine derivative (mmol/day); 0.301 = endogenous excretion of purine derivative in the urine (mmol) per unit of metabolic weight ($BW^{0.75}$); and 0.80 = recovery of purine absorbed as purine derivative in the urine (mmol/mmol).

Ruminal synthesis of microbial nitrogen compounds was calculated as a function of AP using the equation described by Barbosa et al. (2011).

$$Nmic = \frac{(70 x AP)}{(0.93 x R x 1000)} \quad (6)$$

where: NMIC = intestinal flow of microbial nitrogen compounds (g/d); 70 = N content in purines (mg N/mol); 0.93 = digestibility of microbial purines and R = 0.134 = N purine: total N in the bacteria according to Valadares et al. (1999).

Statistical analysis

The experiment was carried out according to a completely randomized design, including the fixed effects of treatments and using the BW of the cows at calving as a covariate. The comparisons among treatments were performed using a set of orthogonal contrasts which encompassed a comparison between the control

treatment and the treatments with supplementation, and the linear and quadratic effects of the CP contents in supplements. All statistical procedures were performed adopting 0.10 as the critical level of probability for the type I error and the MIXED procedure of the Statistical Analysis System 9.4 (SAS Institute, Inc.).

Results

Average availability of DM and DMpd during the experiment was 5.3 t/ha and 3.1 t/ha, respectively, which corresponded to the momentary average availability of 203.1 and 119.0 g/kg BW. Forage sampled by hand-plucking had an average CP content of 72 g/kg DM (Table 2).

The average milk yield and composition were not affected ($P \geq 0.11$) by the treatments (Table 3).

Overall, there was not effect ($P \geq 0.16$) of supplementation on voluntary intake (Table 4). A linear effect ($P < 0.02$) of the CP content in the supplements was observed among supplemented cows only for the CP intake (Table 4).

The average CP contents in the diet, calculated from the ratio of the CP intake on total DM intake, were 69, 70, 80, and 82 g/kg for the control, and supplements with 80, 200, and 320 g CP/kg DM, respectively.

Supplementation did not affect ($P \geq 0.20$) the total digestibility of OM, NDFap, and CP (Table 5). However, among supplemented cows, a positive linear effect ($P < 0.01$) of the CP content in the supplement was observed for the CP digestibility (Table 5).

The efficiency of microbial protein synthesis (EMS) and NMIC were not affected ($P \geq 0.18$) by treatments. Nevertheless, NMIC in relation to nitrogen intake

(NMIC/NI) was lower in supplemented cows compared to the control cows. Among supplemented cows, a negative linear effect ($P < 0.01$) of the CP content in the supplements was observed for NMIC/NI (Table 5).

There was not ($P \geq 0.14$) effect of supplementation or of the CP content in the supplements on performance productive (Table 6). Supplementation did not affect ($P \geq 0.52$) NEFA, SUN and progesterone serum concentrations (Table 7). However, among supplemented cows, a positive linear effect ($P < 0.03$) of the CP content in the supplements was observed for SUN concentrations at 30 days post-calving (Table 7).

Discussion

The forage mass available was not supposed being a limiting factor for feed intake in this study. According to Paulino et al. (2008), the interpretation of forage available for grazing as baseline nutritional resource should be conducted from the perspective of the fraction potentially convertible into animal product which can be achieved by applying the concept of DMpd, as it integrates the quantity and quality regardless of season. The average mass of DMpd (119 g/kg BW) was higher than those recommended by Paulino et al. (2004) from 40 to 50 g/kg BW for satisfactory performance in a grazing system. Thus, the availability of forage could be considered non-restrictive, giving to animals the possibility of a high selective grazing and choose by the best-quality forage parts.

On the other hand, if there is a minimum quantity of forage mass to supply animal demand, canopy structure, and nutritive value are more important than forage mass to pasture intake (Valente et al., 2013). In tropical pastures, protein supply is the major limiting nutrient to production. Average CP content of forage during the

current experiment was approximately 72 g CP/kg DM; in addition, approximately 360 g/kg CP was associated to fiber, being slowly available to ruminal microorganisms (Table 2). According to observations obtained in tropical conditions, additional supplying of nitrogen compounds to animals consuming low quality forage would favor fibrolytic bacteria growth increasing ruminal NDF degradation, voluntary forage intake and energy extraction from fibrous carbohydrates (Paulino et al., 2008; Detmann et al., 2010).

However, positive effects on voluntary intake (Table 4) and fiber digestibility (Table 5) were not observed in this study. The absence of effect on digestible neutral detergent fiber (DNDF) intake indicates that no change was caused by supplementation or by CP contents in the supplement on forage intake and digestibility. According to Detmann et al. (2014a), positive responses on fiber degradation has been observed with increased dietary CP levels for concentrations close to 100 g/kg DM, in addition, the voluntary intake of forage has been stimulated with the establishment of concentrations close to 145 g/kg DM (Detmann et al., 2014b). In this study, there was not difference in protein intake between supplemented and control cows (Table 4). Although it has been observed a linear increase on protein intake among supplemented animals, dietary crude protein content increased slightly, remaining below the suggested by the authors cited above, resulting in deficiency of nitrogen compounds to synthesis of microbial enzymes responsible for the degradation of the fibrous forage compounds (Detmann et al., 2009), resulting in absence of changes in digestion and forage intake.

The absence of effect on voluntary intake reflected on animal performance. Thus, BW did not differ among treatments. In contrast, other studies aiming to

evaluate supplementation of grazing beef cows during post-calving in the tropics found improved weight in supplemented animals (Ruas et al., 2000; Godoy et al., 2004). However, Oliveira et al. (2006) highlighted that the evaluation of BCS is more efficient than BW, because it takes into account the accumulation of body reserves, which the female has to mobilize during suckling period. Moreover, two animals can be marked difference in BW and similar BCS. Nevertheless, there was no effect on BCS. It observed BCS of the cows was lower than the minimum body condition score at parturition (5.0) recommended by the NRC (2000) so that females will have a good reproductive performance in the next breeding season.

Overall, effects of supplementation on the digestibility was focused on increase the CP digestibility with the increase of CP contents in the supplements (Table 5), which may be due to the effect of lower proportion of metabolic fecal fraction in relation to ingested nutrients (Barros et al., 2011).

There was no effect of supplementation or CP contents in the supplements, on NMIC and EMS (Table 5). The absence effect on NMIC has also been observed in others studies using beef cattle grazing in tropical conditions (Cabral et al., 2012; Barros et al., 2014). This behavior could be attributed to the fact that in situations where there lack of nitrogen compounds in diet, there would be a net gain of nitrogen to rumen in the system via recycling to support the rumen microbial growth that is first order demand (Detmann et al., 2014b).

On the other hand, NMIC/NI was affected by supplementation and CP levels in the supplements. According to Detmann et al. (2010) estimates of NMIC/NI higher than one indicates that intestinal flow of microbial nitrogen is higher than ingested nitrogen. These cases, there will be greater reliance on recycling events to

provide adequate supply of N in the rumen (Detmann et al., 2014b). In this study, it may observe that average estimates were close to one, in the control animals, and became lower when supplemental CP was provided. This observation again indicates the occurrence of protein deficit in the animals' diet.

During early lactation, the partitioning mechanisms of nutrients give priority for the milk yield in relation to other functions; thus, increasing feeding levels by supplementation during post-calving could improve milk yield. However, there was no effect of supplementation or CP contents in the supplements on milk yield. Similar results were reported by Ruas et al. (2000), who provided 1 or 2 kg of supplement for Nellore cows during post-calving. The absence of effects on milk yield could be attributed, at least in part, to an increased mobilization of body reserves by cows without supplementation. However, the lack of difference on BCS and serum concentrations of NEFA among supplemented and control cows does not support such argument. Thus, the absence of answer on total intake (Table 4) as a function of supplementation, it seems to more plausibly explain this fact.

In post-calving condition, nutritional requirements of beef cows increase above which pastures can normally provide, and cows usually begin negative energy balance (NEB). In this situation, there is fat mobilization and a subsequent increase in NEFA serum concentrations (Sartori & Guardieiro, 2010). The serum levels of NEFA are quite significant for evaluating the energy state in ruminants, responding quickly to changing feed intake (Peixoto et al., 2007). Thus, adding supplements to the animals' diet, we would expect higher intake and improved energy status of the animal, which could be reflected in decreasing serum concentrations of NEFA. However, serum concentrations of NEFA did not differ between treatments (Table

7). Similar results were reported by Mulliniks et al. (2013), who worked with beef cows grazing on native range during post-calving.

Few studies indicate what threshold of serum NEFA would be considered wherein there is a high lipid mobilization, but according to Oetzel (2004), values higher than 0.40 mmol/L already indicate problems concerning energy balance. The values of NEFA serum (Table 7) suggests that the cows were mobilizing body reserves to supply nutritional deficit. Thus, as previously discussed, supplementation failed to increase energy intake of animals and reduce the NEB.

These observations together would justify, at least in part, the absence of return to ovarian activity, as evidenced by low the concentration of P4 for all treatments (Table 7). Overall, the results suggest that supplementation was no enough for minimize energy balance negative and induce to reproductive response. According to Santos et al. (2009), calved cows are more likely to reconception when presenting weight gain or weight maintenance in the critical period of reproduction.

Conclusions

It is concluded that supplementation of grazing beef cows during post-calving does not affect nutritional and productive performance.

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Tables

Table 1 Supplement composition (g/kg as fed)

Ingredient	Crude protein content		
	80	200	320
Ground corn	470	305	140
Ground sorghum	470	305	140
Soybean meal	0	330	660
Mineral mixture[a]	60	60	60

[a]Composition: dicalcium phosphate (500.0 g/kg), sodium chloride (471.9 g/kg), zinc sulfate (15.0 g/kg), copper sulfate (7.0 g/kg), cobalt sulfate (0.5 g/kg), potassium iodide (0.5 g/kg), sodium selenite (0.1 g/kg), and manganese sulfate (5.0 g/kg).

441 **Table 2** Chemical composition of the supplements and forage

Item	Crude protein content (g/kg DM)			Brachiaria decumbens	
	80	200	320	Forage[d]	Forage[e]
DM[a]	955	952	957	326±0.6	348±0.3
OM[b]	939	921	903	921±0.2	932±0.4
CP[b]	77	209	329	72±0.8	70±0.5
EE[b]	11	13	14	9±0.2	8±0.6
NDFap[b]	138	150	155	658±1.1	698±0.5
NFC[b]	713	549	405	187±1.1	157±0.8
iNDF[b]	5	6	6	261±1.4	266±0.3
NDIN[c]	184	176	137	360±3.3	380±2.3

442 DM = dry matter; OM = organic matter; CP = crude protein; EE = ether extract; NDFap = neutral
443 detergent fiber correct for ash and protein; NFC = non-fiber carbohydrate; iNDF = indigestible neutral
444 detergent fiber; NIDN = insoluble neutral detergent nitrogen.
445 [a]/ g/kg as fed
446 [b]/ g/kg DM
447 [c]/ g/kg total nitrogen
448 [d] / Mean ± standard error of the mean (hand-plucked samples collected throughout study)
449 [e]/ Mean ± standard error of the mean (hand-plucked samples collected during digestibility trial)
450

Table 3 Least squares means, standard error of the mean (SEM) and significance indicative for milk yield and composition

Item	Crude protein content (g/kg DM)				P value[a]		
	Control	80	200	320	SEM	CONT	L	Q
	kg/d							
Milk	8.55	8.94	9.21	9.26	0.87	0.370	0.296	0.720
FCM[b]	8.99	8.91	9.13	9.34	0.95	0.904	0.755	0.994
	g/kg							
Fat	43.4	39.7	39.0	34.0	0.33	0.150	0.301	0.438
Protein	29.9	29.5	30.5	30.4	0.06	0.724	0.361	0.511
Lactose	45.0	46.3	46.0	47.4	0.08	0.109	0.346	0.383
Total solids	128.7	125.4	126.9	122.6	0.33	0.340	0.557	0.477

[a]/ CONT = contrast between supplemented and non-supplemented; L, and Q = linear and quadratic effects regarding to CP content in the supplements: 80, 200 and 320 g/kg

[b]/ FCM = 4 % fat-corrected milk yield

457 **Table 4** Least squares means, standard error of the mean (SEM) and significance
458 indicative for voluntary intake

Item	Crude protein content (g/kg DM)					P value [a]		
	Control	80	200	320	SEM	CONT	L	Q
	kg/d							
DM	12.01	11.27	12.00	12.23	0.84	0.860	0.431	0.812
DMF	12.01	10.43	11.10	11.42	0.77	0.260	0.373	0.860
OM	11.19	10.52	11.18	11.34	0.78	0.852	0.445	0.813
CP	0.83	0.79	0.96	1.06	0.07	0.812	0.012	0.688
NDFap	8.20	7.48	7.89	8.10	0.55	0.359	0.375	0.845
iNDF	3.20	2.78	2.96	3.05	0.20	0.269	0.372	0.859
DOM	6.31	5.85	6.23	6.17	0.44	0.653	0.613	0.687
DNDF	5.09	4.40	4.70	4.78	0.32	0.161	0.403	0.804
	g/kg BW							
DM	24.6	24.2	24.9	24.9	1.43	0.945	0.749	0.836
DMF	24.6	22.3	23.3	23.8	1.33	0.596	0.437	0.871
OM	22.6	22.4	23.3	23.6	1.34	0.739	0.532	0.842
NDFap	16.8	16.1	16.4	16.7	0.94	0.724	0.675	0.979

459 DM = dry matter, DMF = forage dry matter, OM = organic matter, CP = crude protein, NDFap =
460 neutral detergent fiber corrected for ash and protein, iNDF = indigestible neutral detergent fiber, DOM
461 = digestible organic matter, DNDF = digestible neutral detergent fiber
462 [a]/ CONT = contrast between supplemented and non-supplemented; L and Q = linear and quadratic
463 effects regarding to CP content in the supplements: 80, 200 and 320 g/kg
464

Table 5 Least squares means, standard error of the mean (SEM) and significance indicative for total digestibility and synthesis of microbial nitrogen compounds

Item	Crude protein content (g/kg DM)					P value[a]		
	Control	80	200	320	SEM	CONT	L	Q
OM	55.71	55.21	55.98	55.05	0.54	0.647	0.840	0.211
CP	49.36	45.32	48.27	50.26	1.00	0.236	0.002	0.702
NDFap	61.05	60.08	60.05	60.01	0.66	0.205	0.951	0.993
DOM	525.17	518.91	519.65	504.37	5.44	0.072	0.051	0.205
NMIC	127.36	111.65	115.30	103.06	10.51	0.181	0.577	0.543
NMIC/NI	0.95	0.88	0.75	0.65	0.06	0.097	0.003	0.105
EMS	122.10	108.10	119.47	107.24	8.91	0.311	0.945	0.263

OM = organic matter, CP = crude protein, NDFap = neutral detergent fiber corrected for ash and protein, DOM = digestible organic matter (g/kg DM), NMIC = intestinal flow of microbial nitrogen compounds (g/d), NMIC/NI = NMIC in relation to nitrogen intake (g/g ingested N), EMS = efficiency of microbial protein synthesis (g microbial CP synthesis/ kg DOM intake)

[a]/ CONT = contrast between supplemented and non-supplemented; L and Q = linear and quadratic effects regarding to CP content in the supplements: 80, 200 and 320 g/kg

475 **Table 6** Least squares means, standard error of the mean (SEM) and significance
476 indicative for performance

Item	Crude protein content (g/kg DM)				P value[a]		
	Control	80	200	320	SEM	CONT	L	Q
BW3	492.80	490.65	489.58	496.48	5.63	0.931	0.471	0.568
BW6	469.29	476.49	477.06	476.04	5.88	0.295	0.958	0.913
BCS3	4.63	4.83	4.94	4.90	0.17	0.197	0.771	0.706
BCS6	4.25	4.57	4.51	4.60	0.17	0.145	0.934	0.716

477 BW3, BW6 = body weight in kg at 30 and 60 days post-calving; BCS3, BCS6 = body condition score
478 at 30 and 60 days post-calving
479 [a]/ CONT = contrast between supplemented and non-supplemented; L and Q = linear and quadratic
480 effects regarding to CP content in the supplements: 80, 200 and 320 g/kg
481
482

Table 7 Least squares means, standard error of the mean (SEM) and significance indicative for metabolites

Item	Crude protein content (g/kg DM)				P value[a]		
	Control	80	200	320	SEM	CONT	L	Q
NEFA3	0.57	0.61	0.55	0.57	0.08	0.929	0.730	0.651
NEFA6	0.44	0.52	0.43	0.51	0.06	0.526	0.963	0.264
SUN3	8.49	8.28	8.73	10.04	0.51	0.383	0.021	0.500
SUN6	13.58	12.47	12.58	15.25	1.33	0.920	0.182	0.451
P4/3	0.30	0.33	0.31	0.32	0.04	0.613	0.905	0.762
P4/6	0.25	0.24	0.32	0.25	0.05	0.915	0.595	0.128

NEFA = Non esterified fatty acids (mmol/L), SUN = serum urea nitrogen (mg/dL), P4 = Progesterone (ng/mL) 30 and 60 days post-calving

[a]/ CONT = contrast between supplemented and non-supplemented; L and Q = linear and quadratic effects regarding to CP content in the supplements: 80, 200 and 320 g/kg

Evaluation of supplementation plans for suckling beef calves managed on tropical pasture

Abstract This study assessed the effects of different amounts of supplement on the productive performance and behavioral characteristics of suckling beef calves, as well as on the productive performance of their dams on tropical pastures. Forty-four male Nellore beef calves with an average age of 120 days and an initial average body weight (BW) of 145 ± 3.7 kg and their respective dams, with an average BW of 449 ± 6.9 kg, were used. The amounts of supplement evaluated were as follows: 0 = calves received only mineral mixture ad libitum; 3, 6, and 9 = calves received 3, 6, or 9 g/kg BW of supplement, respectively, containing 250 g CP/kg. The experimental design was completely randomized. A linear effect ($P < 0.01$) was observed in the intakes of dry matter and organic matter, and a cubic effect ($P < 0.07$) was observed for the intake of neutral detergent fiber corrected for ash and protein. There was difference ($P < 0.06$) in total digestibility only for neutral detergent fiber. A cubic effect ($P < 0.01$) was observed for the calves' performance. However, the calves' supplementation did not affect the milk yield and performance ($P \geq 0.21$) of their dams. The supplementation decreased grazing time ($P < 0.01$) but did not influence suckling time ($P \geq 0.59$). It is recommended supplying of supplement containing 250 g CP/kg in amount 6 g/kg BW to suckling beef calves managed in tropical pasture. Supplementation of suckling beef calves increases the intake of dry matter, decreases grazing time and forage intake. However, it does not affect the suckling time and productive performance of their dams.

Keywords: calves, behavior, creep feeding, grazing, Nellore, supplements

Introduction

In the production of grazing young beef cattle in the tropics, weaning weight is one of the most important indicators related to production system efficiency.

During the first few weeks of life, milk is the main source of energy and nutrient for young beef calves. Starting at about three months of age, however, milk is no longer sufficient for supplying their requirements (Henriques et al., 2011). Thus, supplementation of suckling calves in creep feeding can therefore enable the exploration of optimum of each individual, resulting in better performance and higher body weight (BW) at weaning (Paulino et al., 2012).

Studies in the tropics on creep feeding have consistently shown an increase in the weaning weight of calves (Porto et al., 2009; Valente et al., 2013; Lopes et al., 2014). Studies have also suggested that calves' supplementation in creep feeding could influence the behavior of calves and reduce their suckling time and frequency. Thus, cows' milk may be reduced due to a reduction in suckling stimulation (Fordyce et al., 1996). As a result, this could have an influence on cow performance. Few studies have evaluated the behavior of beef calves in creep-feeding system, however, and questions remain about the relations among the amounts of supplements that are used, which may influence the biological response of beef calves managed in tropical pasture that are subjected to different supplementation plans.

The objective of this study was thus to evaluate the effect of different amounts of supplement on the productive performance and behavioral characteristics of suckling beef calves, as well as on the productive performance of their dams on tropical pastures.

Material and methods

All procedures involving animals were approved by the Institutional Committee of Universidade Federal de Viçosa for animal care and use experimentation, process UFV number 43/2014.

Animals, experimental design and diets

The experiment was conducted in the beef cattle sector at the Universidade Federal de Viçosa, Viçosa-MG, Brazil (20° 45′ S 42° 52′ W), in a 40-ha area with four paddock for grazing with continuous stocking. This study was carried out during the transition phase between rainy and dry seasons from February to June 2013. Over days of measurements, the average minimum and maximum temperatures were 18.3°C and 29.5°C in February, 19.1°C and 27.9°C in March, 16.5°C and 26.3°C in April, 13.7°C and 24.7°C in May and 13.8°C and 24.3ºC in June. The amounts of rainfall were 111 mm in February, 222 mm in March, 120 mm in April, 62 mm in May, and 25 mm in June.

Forty-four Nellore beef calves males in the suckling phase averaging age of 120 days and 145 ± 3.7 kg of initial body weight (BW), and their respective dams averaging 5 years-old and 449 ± 6.9 kg of BW were used. The experimental design was completely randomized with four treatments and eleven replicates. The treatments for the calves were: 0 = calves received only mineral mixture; 3, 6, and 9 = calves received 3, 6, or 9 g/kg BW of supplement. The supplement was formulated to contain 250 g CP/kg, with a mixture of corn (260 g/kg), sorghum (260 g/kg), soybean meal (450 g/kg), and molasses (30 g/kg), plus mineral mixture (40

g/animal/d) which was weighed and mixed with supplement to be supplied to the group of calves daily.

Each group was managed on a 10-ha paddock with Brachiaria decumbens pastures. There were private feeders for each group of calves (0.5 m per calf); where the cows had no access. Calves were fed daily at 11 a.m. Cows received a mineral mixture ad libitum. The mineral mixture consisted of dicalcium phosphate, (500.0 g/kg), sodium chloride (471.9 g/kg), zinc sulfate (15.0 g/kg), copper sulfate (7.0 g/kg), cobalt sulfate (0.5 g/kg), potassium iodide (0.5 g/kg), sodium selenite (0.1 g/kg) and manganese sulfate (5.0 g/kg).

In order to minimize the possible effects of paddock on the experimental treatments, animals were rotated among the paddocks every 7 days, so that each group stayed on each paddock for the same period.

Experimental procedures and sampling

Animals were submitted to 10 days of adaptation to the diet and experimental area, and 140 days of evaluation (five periods of 28 days). For performance evaluation, the animals were weighed at the beginning and end of the experiment after 14 hours of fasting. At the beginning of the experiment, and every 28 days thereafter, the calves were weighed without fasting (and always in the morning at 6h00) in order to adjust the amount of supplement to be provided to each group. The BCS of the cows was evaluated by the same four trained evaluators, using a scale of 1 to 9, as recommended by the NRC (1996).

The diurnal behavior of calves was monitored by human observation. It took 8 days of observations nonconsecutive, being that each group was observed in each

97 paddock twice. Using the continuous focal animal recording method, the observers
98 stayed at a distance of approximately 50 m from the animals, from 6 a.m. to 6 p.m.
99 similarly to the description by Valente et al. (2013). The behavior observed in this
100 study was the time the animals spent grazing, idling, suckling, and feeding.

101 Forage samples were collected on the 14th day of each experimental period to
102 evaluate forage availability. In each paddock, four samples of forage were randomly
103 selected using a metallic square (0.5 × 0.5 m) and cut approximately 1 cm above the
104 ground. Sampling for the qualitative assessment of forage consumed by the animals
105 was obtained every fourteen days by the hand-plucking method. All the samples
106 were oven-dried (60^{0}C /72 hours) and ground in a Wiley mill (model 3, Arthur H.
107 Thomas, Philadelphia, PA) to pass through a 2-mm screen. After that, half of each
108 ground sample was ground again to pass through a 1-mm screen.

109 To estimate milk yield, cows were milked at days 35, and 90 of the
110 experiment. Cows were separated from their calves at 18h00. At 6h00 of the
111 following day, cows were injected with 2-mL oxytocin (10 IU/mL; Ocitovet®,
112 Brazil) in the mammary artery and immediately milked. The exact time when each
113 cow was milked was recorded, and the milk produced was proportionally converted
114 into a 24-h based production. The milk produced was corrected to 4 % of fat (FCM)
115 according to NRC (2001):

116 $$FCM = 0.4 \times \text{milk yield (kg/d)} + 15 \times \text{fat yield (kg/d)} \quad (1)$$

117 Seventy days after the beginning of the experiment, a nine-day assay to
118 evaluate voluntary intake and digestibility of the calves was carried out. Chromium
119 oxide (Cr_2O_3), used to estimate fecal excretion, was packaged in paper cartridges in
120 the amount of 10 g per calf/d and was introduced (11h00) into the esophagus via a

rubber tube; while titanium dioxide (TiO_2), used to estimate individual supplement intake, was mixed with the supplement distributed to the calves in an amount equal to 10 g per animal/d. The first six days were used to stabilize the flow of markers in gastrointestinal tract of the animals, while the last three days were used for feces collection at 16h00 on day 7, at 11h00 on day 8, and at 6h00 on day 9. The fecal samples were oven-dried (60°C /72 hours) and proportionally pooled per animal, then ground in a Wiley mill (model 3, Arthur H. Thomas, Philadelphia, PA) to pass through a 2-mm screen. After that, half of each ground sample was ground again to pass through a 1-mm screen. In the fifth day of the digestibility assay, a sample of forage in each paddock was obtained by the hand-plucking method, to estimate voluntary intake and digestibility.

To evaluate the microbial protein production of calves, spot urine samples (10 mL) were collected from spontaneous micturition 4 hours after supply of supplement in the 80^{th} experimental day. Urine samples were diluted in 40 mL of H_2SO_4 (0.036 N) and frozen (-20°C). After urine collection, blood samples were collected by jugular vein puncture, using vacuum tubes with separator gel (BD Vacutainer ® SST II Advance), and centrifuged at 3000 × g for 15 minutes and the serum was then frozen (-20°C).

Chemical analysis

Samples of forage, feces, and supplement processed to pass through 1-mm screen sieve were analyzed according to the standard analytical procedures of the Brazilian National Institute of Science and Technology in Animal Science (INCT-CA; Detmann et al., 2012) for dry matter (DM; INCT-CA method G-003/1), ash

(INCT-CA method M-001/1), crude protein (CP; INCT-CA method N-001/1), ether extract (EE; INCT-CA method G-004/1), neutral detergent fiber (NDFap; INCT-CA method F-002/1), using alpha thermostable amylase without addition of sodium sulfite and corrected for ash and protein (NDIP; INCT-CA method N-004/1). Indigestible neutral detergent fiber (iNDF; INCT-CA method F-009/1) was quantified by in situ incubation procedures with F57 bags (Ankom®) for 288 hours in samples processed at 2-mm. In addition, fecal samples were evaluated for the contents of chromium (INCT-CA method M-005/1) and titanium (INCT-CA method M-007/1). Milk samples were analyzed with regards as protein, fat, lactose, and total solids content using infrared spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark). Milk samples were analyzed with regards as protein, fat, lactose, and total solids content using infrared spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark).

The percentage of potentially digestible DM (DMpd) in the forage samples obtained for evaluation of forage availability was estimated according to Paulino et al. (2008):

$$DMpd = 0.98 X (100 - NDF) + (NDF - iNDF) \quad (2)$$

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

Fecal excretion was estimated by rationing the quantity of chromic oxide offered and the concentration in feces.

Individual DM of the supplement (DMS) intake was estimated by relation of excretion of TiO_2 in feces and marker concentration in the supplement.

169 The voluntary intake of DM from forage (DMF) was estimated using an
170 internal iNDF according to Detmann et al. (2001), using the following equation:

171 $$DMF = \left[(FE \times iNDFfeces) - DMSi \times iNDF\ sup\right] \div iNDFforage \quad (3)$$

172 where FE = fecal excretion (kg/d), iNDFfeces = concentration of iNDF in the feces
173 (kg/kg), DMSi = dry matter supplement intake (kg/day), iNDFSup = concentration of
174 iNDF in the supplement (kg/kg), and iNDFforage = concentration of iNDF in the
175 forage (kg/kg).

176 The total DM intake was calculated by the sum of DMF intake, DMS intake,
177 and DM milk intake (DMM).

178 In the samples of urine, analyzes were carried out for creatinine, uric acid and
179 urea by methods colorimetric kinetic, enzymatic colorimetric and kinetic fixed time,
180 respectively, using automatic biochemical analyzer (Mindray, BS200E model).

181 Daily urinary volume was calculated using the relationship between the daily
182 creatinine excretion (CE), taking as reference the equation proposed by Costa e Silva
183 et al. (2012), and its concentration in the spot samples:

184 $$CE(g/d) = 0.0345 x SBW^{0.9491} \quad (4)$$

185 where: SBW = shrunk body weight

186 Allantoin was analyzed by the colorimetric method as described by Chen &
187 Gomes (1992). Total excretion of purine derivatives was calculated by the sum of the
188 amounts of allantoin, uric acid excreted in urine.

189 The purines absorbed were calculated from the excretion of purine derivatives
190 according to Barbosa et al. (2011):

191 $$AP = \frac{(PD - 0.301 x BW^{0.75})}{0.80} \quad (5)$$

where: AP = absorbed purines (mmol/d); PD = excretion of purine derivative (mmol/day); 0.301 = endogenous excretion of purine derivative in the urine (mmol) per unit of metabolic weight (BW0.75); and 0.80 = recovery of purine absorbed as purine derivative in the urine (mmol/mmol).

Ruminal synthesis of microbial nitrogen compounds was calculated as a function of AP using the equation described by Barbosa et al. (2011).

$$NMIC = \frac{(70 x AP)}{(0.93 x R x 1000)} \quad (6)$$

where: NMIC = intestinal flow of microbial nitrogen compounds (g/d); 70 = N content in the purines (mg N/mol); 0.93 = digestibility of microbial purines and R = 0.134 = N purine: total N in the bacteria according to Valadares et al. (1999).

Statistical analysis

The experiment was carried out according to a completely randomized design, including the fixed effect of treatments and using the initial BW as a covariate. The comparisons among treatments were performed out by orthogonal decomposition of the sum of squares of the treatments in linear, quadratic, and cubic effects related to the effects of supplement amounts. All statistical procedures were performed adopting 0.10 as the critical level of probability for the type I error and the MIXED procedure of the Statistical Analysis System 9.4 (SAS Institute, Inc.).

Results

The average availability of DM and DMpd during the experiment was 4.4 and 2.5 t/ha, respectively, which corresponded to the momentary mean availability of

256.2 and 148.3 g/kg BW. The forage sampled by hand-plucking had an average CP content of 86 g/kg DM (Table 1).

The observed supplement intake occurred as planned in the supplementation plans, and orts were not observed. The average supplement intakes throughout the study, were 0.6, 1.2, and 1.8 kg/animal/d for amounts of supplements 3, 6, or 9 g/kg BW, respectively.

The average milk yield and composition were not affected ($P \geq 0.18$) by the calves' supplementation (Table 2).

The intake of DM, OM, CP, and digestible organic matter (DOM) increased linearly ($P < 0.01$) with an increase in the amount of supplement supplied to the calves. DMF intake, however, decreased linearly ($P < 0.01$) (Table 3). In addition, a cubic effect ($P < 0.07$) was observed for NDF and iNDF intake (Table 3).

There were no differences ($P \geq 0.25$) in total digestibility of OM, CP, and EE; on the other hand, a quadratic effect ($P < 0.06$) was observed for the digestibility of NDFap (Table 4).

A quadratic effect ($P < 0.08$) was observed in the amount of supplement on the intestinal flow of microbial nitrogen compounds (NMIC); no differences were found ($P \geq 0.65$), however, in the efficiency of microbial protein synthesis (EMS). In addition, the NMIC in relation to nitrogen intake (NMIC/NI) decreased linearly ($P < 0.02$) with increases in the amount of supplement provided to calves. A positive linear effect of amount of supplement ($P < 0.01$) was observed in the urea nitrogen excretion in the urine (UUN) and the concentration of serum urea nitrogen (SUN) (Table 4).

The calves' supplementation did not affect ($P > 0.21$) the FBW and FBCS of their dams. A cubic effect ($P < 0.01$), however, was observed in the calves' performance with increases in amount of supplement (Table 5). Supplement use efficiency (the additional weight gain of the supplemented calves/supplement intake) was 0.15, 0.17, and 0.09 for amounts of supplements 3, 6, or 9 g/kg BW, respectively.

The different amounts of supplement did not affect ($P \geq 0.59$) the suckling behavior of calves. On the other hand, grazing time decreased linearly ($P < 0.01$) with increased amount of supplement, and a cubic effect ($P < 0.01$) was observed for feeding time (Table 6).

Discussion

Forage mass did not appear to limit forage intake in this study. The average mass of DMpd (148.3 g/kg BW) was higher than the 40-50 g/kg BW recommended by Paulino et al. (2004) for satisfactory animal performance in a grazing system. Nonetheless, the forage was considered to be of moderate quality (Table 1). According to Detmann et al. (2014a), positive effect on the utilization of energy substrates from forage has been observed with increasing dietary CP content up to 100 g/kg DM. In addition, voluntary intake forage have been stimulated with concentrations of up to 145 g CP/kg DM (Detmann et al., 2014b). Thus, supplementation may increase dietary CP content and could optimize the utilization and intake of forage and, as a result, animal performance.

In fact, in our study there was a linear increase in the intake of DM and OM, resulting in increased animal performance. However, there was a substitutive effect

of the intake of the supplement that was offered on forage intake. Some authors have associated the dietary protein to energy (P:E) ratio, represented by the ratio CP: DOM, with variations in the voluntary intake of forage (Costa et al. 2011; Detmann et al., 2014b). In a recent approach, Detmann et al. (2014b) inferred that the maximum voluntary intake of tropical forages is observed with the ratio of 288 g CP/kg DOM. The CP:DOM ratio observed in our study was 217.4, 248.0, 276.4, and 326.9 for the amounts of supplement 0, 3, 6, or 9 g /kg BW, respectively. It would therefore appear that a reduction in the forage intake observed may have been caused in parts by an excess of dietary protein, which supports the occurrence of negative metabolic effects on voluntary forage intake.

In situations, with excessive protein intake leads excess ammonia being directed to liver, and an increased liver synthesis of urea, which in turn increases caloric increments (Detmann et al., 2010). Thus, the excessive catabolism of nitrogenous compounds may have caused the reduction in voluntary intake (Detmann et al., 2014b) in an attempt to approach a situation of thermal comfort. This situation can be confirmed by the higher concentrations of SUN and the higher excretion of UUN (Table 4), given the positive correlation between these variables and protein intake. In addition, these arguments could justify the reduction in performance that was observed by providing supplements in the amount of 9 g/kg BW.

On the other hand, by analyzing the intake of each treatment it was observed that the provision of supplements in the amount of 6 g/kg BW allowed a ratio of 276.4 CP / kg DOM (close to the ratio suggested by the authors cited above); this reflected greater intake (Table 3), and, consequently, greater animal performance**.** One possible explanation for this superior performance is that this amount presented

an appropriate balance of protein and energy, where there occurred a supply of protein deficit dietary with appropriate energy offer, allowing an efficient utilization of protein (Souza et al., 2010) and in turn a greater availability of metabolizable energy and protein for animal metabolism to tissue synthesis.

The amounts of supplement assessed did not affect the calves' suckling behavior (Table 6). As a consequence, the calves' DM intake of milk (Table 3) did not differ between treatments; therefore, this was not a factor that would interfere with the difference in performance among calves. On the other hand, there was an increase in the intake of CP, DOM, and NFC, which resulted from increases in the supplement supply; this reflected in higher performance. This fact shows that during the suckling period, the nutrients available from milk and tropical forage alone were not sufficient for supplying the nutritional requirements for optimized weight gain. This result is similar to other studies (Valente et al., 2013; Lopes et al., 2014 and Barros et al., 2014) that have also found increases in performance due to creep feeding. According to Paulino et al. (2010), beef calves with a BW of 200 kg and an ADG of 1 kg/d require a daily intake of 3.0 of total digestible nutrients (TDN) and 0.708 kg of CP, respectively. Thus, in our study it was observed that the provision of 6 g/kg BW allowed an intake of energy and protein that was close to that suggested by the authors cited above.

According to Paulino et al. (2014), in order to enable the production of young beef cattle on tropical pastures, a feeding management system should be established that will allow weight gains from birth until slaughter at 16 months of about 900 g/animal/d. In our study, this performance was obtained by providing supplement in the amount of 6 g/kg BW, which provided an additional gain of 200 g/d in relation to

310 the nutritional plan 0 g/kg BW. This observation corroborates statements made by
311 Poppi & McLennan (1995) and Paulino et al. (2008) that stated that it is possible to
312 obtain an additional gain of 200-300 g/d with the use of fodder supplements when
313 the forage grazed has good nutritional quality.

314 Overall, protein supplementation increases the degradation rates of potentially
315 digestible NDF in the rumen (Detmann et al., 2011). In the suckling phase, however,
316 the rumens of calves are still in the development stage, which makes the calves more
317 susceptible to digestive disorders; as such they are less able to utilize rough feed.
318 This fact, together with the low participation of NDFap in the diet, may have
319 contributed to a reduction in NDFap digestibility observed in our study; in addition,
320 increased amounts of the supply of supplements have been reported to have
321 deleterious effects on the digestibility of NDFap (Paula, 2012). Others studies
322 conducted on the suckling phase have also shown the absence of effect or reduction
323 in NDF digestibility (Marquez et al., 2014; Lopes et al., 2014).

324 According to Vargas Jr et al. (2011), milk yield may be increased as the calf
325 stimulates the mammary gland. A high frequency of suckling could make the cow
326 increase milk yield in response to higher stimulation; as result, this may reduce her
327 body condition. On the other hand, as the calf grows, its ability to eat solid feed
328 increases while its milk intake decreases; the adoption of supplementation can thus
329 reduce dependence of calves in relation to cows' productivity levels, resulting in
330 lower variations in body condition. In our study, however, the milk yield was not
331 affected by calves' treatments (Table 2). Consequently, did not observe any effect on
332 cows' performance (Table 5) due to the calves' treatments. Similarly, other studies

(Valente et al., 2013; Barros et al, 2014) also did not find differences in milk yield and productive performance in beef cows due to the calves' supplementation.

This likely occurred because calves prefer milk, supplement and forage, in that order; it thus seems unlikely that calves would replace milk by supplements. Calves will therefore usually increase supplement and forage intake only after the maximization of milk intake. In our study, were not observed differences in suckling times for different amounts of supplement (Table 6), which is similar to results found by Valente et al. (2013). With increased amount of supplements, however, calves were less dependent upon pasture to supply their nutritional demands, and they spent less time grazing (Table 6), which indicates the replacement of forage intake by supplements (Table 3). Similarly, other studies (Lopes et al., 2014; Cardenas et al., 2015) have also observed that there were no changes in dry matter milk intake; although forage intake was lower for supplemented calves.

Conclusions

From the results obtained of this study, it is recommended the provision of supplement containing 250 g CP/kg in the amount of 6 g/kg BW to suckling beef calves managed in tropical pasture. Supplementation of suckling beef calves increases intake of dry matter, and decreases grazing time and forage intake. However, it does not affect suckling time and productive performance of their dams.

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Tables

Table 1 Chemical composition of the supplements and forage

Item	Supplement	Brachiaria decumbens	
		Forage[d]	Forage[e]
DM[a]	945.0	295.2±0.4	280.3±0.6
OM[b]	963.7	901.5±0.3	903.5±0.4
CP[b]	275.6	86.1±0.2	84.5±0.3
EE[b]	230.0	10.4±0.1	7.8±0.1
NDFap[b]	177.4	630.4±0.9	658.2±1.0
iNDF[b]	8.7	257.7±0.8	254.9±0.4
NFC[b]	487.7	170.0±0.6	152.9±1.2
NDIN[c]	161.7	209.8.±1.1	193.9.±0.8

DM = dry matter; OM = organic matter; CP = crude protein; EE = ether extract; NDFap = neutral detergent fiber correct for ash and protein; iNDF = indigestible neutral detergent fiber; NFC = Non-fiber carbohydrate; NIDN = insoluble neutral detergent nitrogen

[a]/ g/kg as fed

[b]/ g/kg DM

[c]/ g/kg total nitrogen

[d]/ Mean ± standard error of the mean (hand-plucked samples collected throughout study)

[e]/ Mean ± standard error of the mean (hand-plucked samples collected during digestibility trial)

491

492 **Table 2** Least squares means, standard error of the mean (SEM) and significance
493 indicative for milk production and composition

Item	Supplement amount (g/kg BW)					P value[a]		
	0	3	6	9	SEM	L	Q	C
	kg/d							
Milk	7.01	7.39	7.46	7.03	0.41	0.555	0.149	0.747
FCM[b]	7.48	7.87	7.97	7.11	0.45	0.559	0.124	0.622
	g/kg							
Fat	46.63	44.21	45.75	46.39	0.14	0.899	0.282	0.442
Protein	34.29	34.32	35.00	34.10	0.04	0.888	0.179	0.182
Lactose	43.80	44.63	43.79	44.88	0.04	0.182	0.743	0.917
Total solids	135.97	134.47	135.95	136.31	0.16	0.730	0.570	0.570

494 [a]/ L, Q and C = linear, quadratic and cubic effects of the supplement amount

495 [b]/ FCM = 4 % fat-corrected milk yield

496

497 **Table 3** Least squares means, standard error of the mean (SEM) and significance
498 indicative for voluntary intake of calves

Item	Supplement amount (g/kg BW)					P value[a]		
	0	3	6	9	SEM	L	Q	C
	Kg/d							
DM	3.15	3.52	4.20	3.98	0.21	0.001	0.181	0.214
DMF	2.29	1.99	2.06	1.70	0.11	0.004	0.813	0.139
DMM	0.9	1.0	1.0	0.9	0.08	0.786	0.630	0.846
OM	2.85	3.24	3.84	3.69	0.18	0.001	0.174	0.272
CP	0.416	0.563	0.716	0.827	0.05	<0.001	0.734	0.830
EE	0.299	0.345	0.374	0.337	0.03	0.251	0.135	0.699
NDFap	1.50	1.47	1.69	1.20	0.08	0.083	0.012	0.015
NFC	0.35	0.51	0.70	1.09	0.06	<0.001	0.079	0.528
iNDF	0.56	0.51	0.56	0.45	0.03	0.046	0.376	0.068
DOM	1.91	2.27	2.59	2.53	0.15	0.003	0.185	0.633
	g/kg BW							
DM	15.94	17.71	20.03	19.65	0.71	0.002	0.138	0.311
DMF	11.65	10.00	9.87	8.49	0.51	0.002	0.805	0.250
OM	14.43	16.32	18.39	18.22	0.65	<0.001	0.126	0.416
NDFap	7.64	7.40	8.13	6.00	0.36	0.014	0.013	0.023

499 DM = dry matter, DMF = forage dry matter, DMM = milk dry matter, OM = organic matter, CP =
500 crude protein, EE = ether extract, NDFap = neutral detergent fiber corrected for ash and protein, NFC
501 = Non-fiber carbohydrate, iNDF = indigestible neutral detergent fiber, DOM = digestible organic
502 matter, DNDF = digestible neutral detergent fiber
503 [a]/ L, Q and C = linear, quadratic and cubic effects of the supplement amount
504

505 **Table 4** Least squares means, standard error of the mean (SEM) and significance
506 indicative for digestibility and synthesis of microbial nitrogen compounds

Item	Supplement amount (g/kg BW)					P value [a]		
	0	3	6	9	SEM	L	Q	C
OM	66.80	69.54	67.15	68.47	1.21	0.625	0.553	0.105
CP	63.77	67.34	64.67	68.42	2.15	0.251	0.964	0.200
EE	82.51	86.66	84.17	84.87	1.75	0.569	0.339	0.226
NDFap	61.53	59.72	57.51	49.87	1.51	<0.001	0.056	0.451
NFC	50.51	67.19	65.46	76.14	2.71	<0.001	0.283	0.017
DOM	605	641	616	635	11.2	0.194	0.435	0.042
NMIC	23.97	35.63	33.24	34.74	2.61	0.017	0.069	0.152
NMIC/NI	0.41	0.40	0.28	0.28	0.05	0.018	0.933	0.295
EMS	80.72	92.94	82.20	89.21	7.41	0.652	0.725	0.229
SUN	11.35	13.17	17.41	18.95	0.91	<0.001	0.882	0.220
UNUE	24.44	31.85	43.72	43.42	2.21	<0.001	0.109	0.111

507 OM = organic matter, CP = crude protein, EE = ether extract, NDFap = neutral detergent fiber
508 corrected for ash and protein, NFC = Non-fiber carbohydrate, DOM = digestible organic matter (g/kg
509 DM), NMIC = intestinal flow of microbial nitrogen compounds (g/d), NMIC/NI = NMIC in relation
510 to nitrogen intake (g/g ingested N), EMS = efficiency of microbial protein synthesis (g microbial CP
511 synthesis/ kg DOM intake), SUN = serum urea nitrogen (mg/dL), UNUE = ureic nitrogen urinary
512 excretion (g/d)
513 [a]/ L, Q and C = linear, quadratic and cubic effects of the supplement amount
514

Table 5 Least squares means, standard error of the mean (SEM) and significance indicative for performance of calves and cows

Item	Supplement amount (g/kg BW)					P value [a]		
	0	3	6	9	SEM	L	Q	C
	Calves							
FBW, kg	246.4	258.3	275.4	269.6	3.70	<0.001	0.021	0.095
ADG	0.72	0.80	0.92	0.89	0.03	<0.001	0.021	0.095
	Cows							
FBW	465.5	462.5	465.9	473.6	4.65	0.212	0.253	0.924
FBCS	4.53	4.61	4.54	4.73	0.08	0.139	0.539	0.271

FBW = final body weight in kg, ADG = average daily gain in grams, FBCS = final body condition score

[a]/ L, Q and C = linear, quadratic and cubic effects of the supplement amount

Table 6 Least squares means, standard error of the mean (SEM) and significance indicative for diurnal behavior of calves

Item	Supplement amount (g/kg BW)					P value[a]		
	0	3	6	9	SEM	L	Q	C
(%)	Calves							
Grazing	33.39	33.07	32.54	27.76	1.28	0.003	0.503	0.707
Idle	61.28	59.49	56.41	61.72	1.30	0.761	0.009	0.105
Suckling	3.09	3.00	3.11	2.80	0.31	0.597	0.728	0.663
Feeding	2.22	3.80	7.93	7.71	0.55	<0.001	0.111	0.008

[a]/ L, Q and C = linear, quadratic and cubic effects of the supplement amount

Does beef calves' supplementation in creep feeding system influence body condition of their dams?

Abstract The aim of this study was to evaluate the effects of beef calves' supplementation in creep feeding system on milk yield, BW, and BCS of their dams on tropical pastures using a meta-analytical approach. The database was obtained from 11 experiments conducted between 2009 and 2014 in Brazil, totaling 485 observations (cows). The database was composed by 273 Nellore and 212 crossbred ($^{7}/_{8}$ Nellore × $^{1}/_{8}$ Holstein) cows. All experiments were carried out in the suckling phase (from 3 to 8 mo of age of calves) during transition phase between rainy and dry seasons from February to June of each year. The data were analyzed by a meta-analytical approach using mixed models and taking into account for the random variation among experiments. All statistical evaluations were performed using 0.05 as the critical level for the occurrence of type I error. Calves' supplementation ($P \geq 0.59$) and the calf' sex ($P \geq 0.48$) did not affect milk yield of cows. Average fat corrected milk (FCM) yield was 6.71 and 6.83 kg/d for cows that had their calves supplemented and not supplemented, respectively. Differences were observed ($P < 0.0001$) for milk yield due to the genetic group where crossbred cows presented greater FCM yield (7.37 kg/d) compared to Nellore cows (6.17 kg/d). There was no effect of the calves' supplementation on BW change ($P \geq 0.11$) and BCS change ($P \geq 0.23$) of the cows. Therefore, it is concluded that supplementation of beef calves using creep-feeding systems in tropical pastures does not affect milk yield, performance, or body condition of their dams.

Keywords: beef calves, beef cows, Nellore, supplements

Introduction

Continuous weight gain from suckling period until slaughter is a critical feature to the success of beef cattle production in the tropics. However, milk nutrients cannot be enough to supply the calves' nutritional requirements for optimized weight gains after three months of age (Henriques et al., 2011). Therefore, the supplementation of suckling calves using a creep feeding systems assumes great importance to assure an improved weaning weight (Paulino et al., 2012).

In fact, studies in the tropics on creep-feeding have consistently shown increases in the BW of calves at weaning (Porto et al., 2009; Valente et al., 2013; Barros et al., 2014; Lopes et al., 2014). However, few studies have evaluated the possible effects of creep feeding on cow performance. In addition, most of these studies had a small number of observations and contradictory results have been obtained, which can be, at least partially, confounded with the variation between years of evaluation. Different studies have shown that creep feeding supplementation of calves can either increase (Nogueira et al., 2006; Souza et al., 2007), decrease (Sampaio et al., 2010) or produce no affect (Porto et al., 2009; Valente et al., 2013) cow’s performance. Considering these inconsistencies concerning inferences taken from individual studies, it is believed that increasing the number of observations can improve the inferences and a meta-analytical approach can be useful for this.

Therefore, the objective of this study was to evaluate the effects of beef calves’ supplementation in creep-feeding system on milk yield, BW and BCS of their dams on tropical pastures using a meta-analytical approach.

Material and methods

Data acquisition and experimental procedures

The dataset used to evaluate milk yield, BW and BCS of cows was obtained from 11 experiments conducted between 2009 and 2014 in Brazil, totaling 485 observations (cows). The experiments were carried out at the Animal Science Department of the Universidade Federal de Viçosa, Brazil (20° 45′ S 42° 52′ W), in a 90-ha area consisting of thirteen paddocks, covered with Brachiaria decumbens, and grazing under continuous stocking rate. There were water dispensers and shaded feeders in each paddock. In addition, there were shaded feeders with access restricted to calves (creep feeders).

Experiments were performed during the suckling phase to evaluate the effects of supplementation on calves' performance (Table 1). The database were composed by 273 Nellore and 212 crossbred ($^{7}/_{8}$ Nellore × $^{1}/_{8}$ Holstein) cows averaging 5 yr old.

All experiments were conducted during transition phase between rainy and dry seasons from February to June of each year according to completely randomized designs. The treatments were applied to calves and each study included a control treatment without supplementation (calves receiving only mineral mixture) and three to four treatments where calves had access to creep feeding supplements. In all experiments calves were fed daily at 1100h from three months of age until weaning with approximately eight months old. Overall, the CP content in the supplements ranged from 80 to 550 g/kg as-fed, and the amount of supplements provided to calves ranged from 450 to 1600 g/d (Table 1). Cows received a mineral mixture ad libitum

and 100 g of ground corn per day in feeders located close to creep feeders to allow calves to spend more time in the feeder for intake of the supplement. The mineral mixture consisted of dicalcium phosphate, (500.0 g/kg), sodium chloride (471.9 g/kg), zinc sulfate (15.0 g/kg), copper sulfate (7.0 g/kg), cobalt sulfate (0.5 g/kg), potassium iodide (0.5 g/kg), sodium selenite (0.1 g/kg) and manganese sulfate (5.0 g/kg).

In all experiments, BCS of the cows was evaluated using a scale from 1 to 9, as recommended by the NRC (1996). Variation of the BCS of cows was determined by the difference between scores recorded at the end and beginning of the supplementation period. For BW evaluations, the cows were weighed at the beginning and end of the supplementation period after 14-h fasting. To minimize possible interference due to differences among cows used in the different experiments, the variation in BW and BCS was scaled according to the following equations:

$$VBWr = \frac{(FBW - IBW)}{IBW} \quad [1]$$

$$VBCSr = \frac{(FBCS - IBCS)}{IBCS} \quad [2]$$

where VBWr is the variation of BW scaled to initial BW, FBW is the final BW (kg), IBW is the initial BW (kg), VBCSr is the variation of BCS scaled to initial BCS, FBCS is the final BCS, and IBCS is the initial BCS.

To estimate the average milk yield, two to three milkings were performed in each experiment during the supplementation. Cows were separated from their calves at 18h00. At 6h00 of the following day, cows were injected with 2-mL of oxytocin (10 IU/mL; Ocitovet®, Brazil) in the mammary artery and immediately milked. The

exact time when each cow was milked was recorded, and the milk produced was proportionally converted into a 24-h based production. Milk samples were analyzed with regard fat content using infrared spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark). The milk produced was corrected to 4 % of fat (FCM) according to NRC (2001):

$$FCM = 0.4 \times \text{milk yield (kg/d)} + 15 \times \text{fat yield (kg/d)} \quad [3]$$

A summary of the dataset on calves' and cows' performance is shown in Tables 1 and 2, respectively.

Statistical analysis

The data was analyzed according to meta-analysis techniques (St-Pierre, 2001; Van Houwelingen et al., 2002) using the MIXED procedures of SAS 9.4. The basic model included the fixed effects of calves' supplementation (with or without supplementation), genetic group of the cow (Nellore or crossbred), sex of the calves (male or female), and their interactions. The random effect of the different experiments was considered in the parameters estimation. The best (co)variance structures were evaluated using the corrected Akaike's information criterion and the degrees of freedom were estimated according to the Kenward-Roger method. All variance components were estimated using the restricted maximum likelihood method and the statistical evaluations were performed using 0.05 as the critical level for the probability of type I error.

Results

Calves' supplementation ($P \geq 0.59$) and sex ($P \geq 0.48$) did not affect the FCM yield of the cows (Table 3). Average FCM yield was 6.71 and 6.83 kg/d for the cows that had their calves supplemented and no supplemented, respectively. However, there was an effect ($P < 0.0001$) of genetic group of cows on milk yield. Average FCM yield was 7.37 and 6.17 kg/d for crossbred and Nellore cows, respectively (Table 4).

There was no effect ($P \geq 0.11$) of calves' supplementation on VBWr of the cows (Table 3). Average VBWr was 0.047 and 0.035 for the cows that had their calves supplemented and no supplemented, respectively (Table 4). Similarly, there was no effect of the genetic group ($P \geq 0.36$) of the cows and calf' sex ($P \geq 0.13$) on VBWr (Table 3).

There was no effect ($P \geq 0.23$) of calves' supplementation, genetic group of the cows and calf' sex on VBCSr of the cows (Table 3). Average VBCSr was 0.023 and 0.021 for cows that had their calves supplemented and no supplemented, respectively (Table 4).

No interaction was detected ($P \geq 0.09$) in this study (Table 3).

Discussion

According to Vargas Junior et al. (2011), milk yield would be increased as the calf searching for milk would stimulate the mammary gland. On the other hand, solid intake by the calf is negatively correlated with milk yield (Henriques et al., 2011). Thus, supplementation of calves would, in theory, decrease milk yield of cows due to a decreased suckling stimulation (Fordyce et al., 1996).

However, according to the present study, calves' supplementation did not affect milk yield of the cows. Similarly, other studies (Gelvin et al., 2004; Barros et al, 2014) also did not find differences for milk yield in beef cows due to calves' supplementation. Thus, it seems that calves' supplementation does not change the suckling behavior of calves, and consequently would not affect milk yield.

In fact, several authors in the tropics have not found a decrease in milk intake due to the creep feeding. Valente et al. (2013) evaluated the behavior of beef calves supplemented with different protein-to-carbohydrate ratios during suckling phase and did not observe differences in time and frequency of suckling between supplemented and not supplemented calves. Consequently, these authors did not observe difference in milk yield of cows. Similarly, Lopes (2015), evaluating the effect of increasing the amount of supplement (0, 3, 6, or 9 g/kg BW) for beef calves from 3 to 8 months of age, also did not observe any differences in suckling time and milk yield. The lack of effect of creep feeding on the milk yield probably occurred because calves prefer milk, supplement, and forage, in that order. Therefore, it seems unlikely that calves replace milk by supplement. Thus, calves will usually increase supplement and forage intake only after maximizing milk intake.

In fact, in both studies cited above, a decrease in grazing time was observed with supplementation, which means there was the replacement of the forage by supplement, supporting the lack of alteration on milk intake. Other authors (Barros et al., 2014; Lopes et al., 2014; Cardenas et al., 2015) evaluated the effects of supplementation of beef calves in tropical pastures on the intake, and observed that there was no change in milk intake, although, forage intake was lower for supplemented calves.

Differences on milk yield were observed with regards the genetic group of the cows, where crossbred animals were more productive. Similarly, Oliveira et al. (2007) and Valente et al. (2012) observed higher milk yield in crossbred cows compared with Nellore cows. Generally, Nellore cows have been considered to present lower potential for milk yield compared to European or crossbred animals. Greater milk yield of crossbred cows may be explained by their greater genetic potential for milk yield related to the effect of heterosis provided by crossbreeding that uses European animals with higher potential for milk yield and lactation persistence.

The influence of calf‘s sex on milk yield of beef cows has been reported in some studies. However, the results seem to be contradictory sometimes. Cruz et al. (1997) found higher milk yield for cows that were breastfeeding male calves. The authors attributed this behavior to the higher suckling by the male calf due to its greater demand for nutrients, thus stimulating the milk yield of their dams. On the other hand, Spasadin et al. (2001) evaluated milk yield and suckling behavior in five beef cattle production systems and did not find any effect of calf’s sex on the suckling time and milk yield. A similar behavior was also reported by Fagundes et al. (2004). Similarly, in this study, no influence was observed for calf’s sex on milk yield of their dams.

Suckling frequency influences milk yield and body condition of dams (Kress et al., 1990). Thus, a higher amount of suckling could increase milk yield in response to stimulation and, consequently, it could lead to higher mobilization of reserves and decrease BCS of the cow. On the other hand, as calf grows up, it increases its ability to eat solid feed while milk intake decreases, so that supplementation could reduce

its dependence in relation to cow production, resulting in lower variation in BCS of the cow.

However, differences in VBWr were not observed as a consequence of the calves' supplementation. It was observed for all experiments that there was an increase on body weight of calves at weaning with adoption of creep feeding. However, there were no favorable results on cows when adopting the supplementation of their offspring.

The absence of effect of calves' supplementation on cow BW and BCS variations may be associated with the lack of difference in milk yield due to the calves' supplementation as discussed previously. It must be emphasized that environmental conditions were the same for all animals and differences should be focused only on the supplementation of calves. Other authors (Duarte, 2007; Valente et al., 2012) evaluated the effects of supplementation of beef calves in tropical pastures from 3 to 8 months of age on BW and BCS of their dams, and also reported none difference caused by calves' supplementation.

However, Oliveira et al. (2006) highlighted that the evaluation of BCS would be more efficient than BW, because it would be more able to indicate variation the accumulation of body reserves then a direct measurement of BW variation. Variations in BW could still occur due to variations in rumen fill, physiological condition associated to pregnancy, calving, and tissue hydration, instead of to represent consistent changes in the body as fat and protein contents.

Similarly to VBWr, there was no effect of calves' supplementation on VBCSr. Overall, results of the present study shown that other factors such as the availability and quality of forage, or even supplementation of cows, seem to be more

related to possible changes in BW and BCS than adoption of supplementation strategy of the offspring.

Conclusions

Supplementation of beef calves using creep feeding systems in tropical pastures does not affect milk yield, performance and body condition of their dams.

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336

337 **Tables**

338

339 **Table 1** Summary of the data of experiments used in this study

Study[a]	Experimental period (d)	Calf sex	ASI[c](g)	ADG[d]	
				NS	SUP
Paula et al. (2012)	112	Male	583	662	728
Valente et al. (2013)	112	Male	530	608	804
Barros et al. (2014)	112	Female	500	687	769
Lopes et al. (2014)	140	Male	900	727	880
Cardenas et al. (2015)	140	Female	500	619	677
Barros et al. (2015)	140	Male	850	731	843
Marquez et al. (2014)	150	Female	450	628	677
Lopes et al. (2014)	140	Male	1200	720	873
Lima (2014)	112	Male/Female	700	511	631
Almeida (2015)[d]	140	Female	800	642	732
Martins (2015)[d]	140	Male	1600	500	900

340 [a/]Processed data; to access individualized data consult the references.
341 [b/]ASI = average supplement intake in supplemented animals (as-fed).
342 [c]/ADG = average daily gain (g) , NS = calves received only mineral mixture; or SUP = Calves
343 received multiple supplements in creep feeding system
344 [d]/ Unpublished data.
345

346

Table 2 Descriptive statistics of database utilized to access the effects of beef calves' supplementation on beef cows performance

Item[a]	Mean	Minimum	Maximum	SD[b]	n[c]
		Overall dataset			
IBCS	4.44	2.40	6.80	0.82	468
VBCS	0.11	-1.80	1.70	0.56	468
VBCSr	0.037	-0.300	0.538	0.136	468
IBW, kg	444	311	620	58.53	485
VBW, kg	18.82	-77.00	110.00	20.60	485
VBWr	0.044	-0.161	0.277	0.047	485
Milk yield, kg/d	6.18	1.10	11.80	1.90	477
FCM, kg/d	6.78	0.70	13.70	2.09	477
		Without creep feeding			
IBCS	4.50	2.60	6.50	0.87	105
VBCS	0.06	-1.80	1.60	0.63	105
VBCSr	0.028	-0.300	0.524	0.151	105
IBW, kg	439	312	595	62.70	109
VBW, kg	14.10	-77.00	63.00	21.74	109
VBWr	0.034	-0.161	0.159	0.050	109
Milk yield, kg/d	6.04	1.90	11.80	1.82	104
FCM, kg/d	6.73	1.70	11.20	1.95	104
		With creep feeding			
IBCS	4.41	2.40	6.80	0.80	363
VBCS	0.13	-1.50	1.70	0.53	363
VBCSr	0.040	-0.222	0.531	0.129	363
IBW, kg	445	311	620	57.26	376
VBW, kg	20.19	-60.00	110.00	57.32	376
VBWr	0.047	-0.107	0.278	0.047	376
Milk yield, kg/d	6.23	1.10	11.60	1.93	373
FCM, kg/d	6.80	0.70	13.70	2.13	373

[a]/IBCS = initial BCS; VBCS = variation of BCS during experimental período; VBCSr = variation of BCS scaled to initial BCS (see Equation 2); IBW = initial BW; VBW = variation of BW during experimental period; VBWr = variation of BW scaled to initial BW (see Equation 1); FCM = 4 % fat-corrected milk yield

[b]/Standard deviation

[c]/number of observations

Table 3 Descriptive level of probability for type I error for the fixed effects of the calves' supplementation (SUP), cows' genetic group (GG), calf' sex (SEXC), and their interactions on cows' performance

	Variable[a]		
Effect	VBCSr	VBWr	FCM, kg/d
SUP	0.89	0.11	0.59
GG	0.42	0.36	<0.0001
SEXC	0.23	0.13	0.48
SUPxGG	0.19	0.40	0.72
SUPxSEXC	0.09	0.93	0.46
GGxSEXC	0.92	0.46	0.87
SUPxGGxSEXC	0.76	0.33	0.31

[a]/VBCSr = variation of BCS scaled to initial BCS (see Equation 2); VBWr = variation of BW scaled to initial BW (see Equation 1); FCM = FCM = 4 % fat-corrected milk

Table 4 Least square means for the variables associated with cows' performance according to the effects of the calves' supplementation (SUP), cows' genetic group (GG), calves' sex (SEXC), and their interactions on cows' performance

			Variable[a]		
SUP[b]	GG[c]	SEXC[d]	VBCSr	VBWr	FCM
-	C	F	0.058±0.052	0.048±0.014	7.88±0.64
+	C	F	0.056±0.048	0.061±0.011	7.33±0.55
-	C	M	-0.039±0.043	0.021±0.011	7.06±0.54
+	C	M	-0.004±0.040	0.042±0.009	7.23±0.45
-	N	F	0.084±0.050	0.040±0.012	6.45±0.60
+	N	F	0.046±0.046	0.054±0.010	6.42±0.53
-	N	M	-0.017±0.042	0.028±0.011	5.94±0.51
+	N	M	-0.006±0.041	0.032±0.009	5.87±0.45

[a]/VBCSr = variation of BCS scaled to initial BCS (see Equation 2); VBWr = variation of BW scaled to initial BW (see Equation 1); FCM = 4 % fat-corrected milk yield

[b]/ "-" SUP = not supplemented calves, and "+" = supplemented calves

[c]/C = crossbred cows; and Nellore cows

[d] / F = female and M = male