

FERNANDA HELENA MARTINS CHIZZOTTI

**NÍVEIS DE NITROGÊNIO NÃO-PROTÉICO E SILAGENS DE DIFERENTES
HÍBRIDOS DE MILHO NA DIETA DE BOVINOS DE CORTE**

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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APROVADA: 28 de setembro de 2007.

Prof^a. Maria Ignez Leão

Prof. Sebastião de Campos Valadares
Filho (Co-orientador)

Prof^a. Karina Guimarães Ribeiro

Prof. Luis Orlindo Tedeschi

Prof. Odilon Gomes Pereira
(Orientador)

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BIOGRAFIA

Fernanda Helena Martins Chizzotti, filha de José Maria Martins e Osvanda das Graças Martins, nasceu em Ituiutaba, Minas Gerais, em 05 de setembro de 1977.

Em Setembro de 2002, graduou-se em Zootecnia pela Universidade Federal de Viçosa.

Em Setembro de 2002, iniciou o curso de mestrado em Zootecnia, na Universidade Federal de Viçosa, concentrando seus estudos na área de Forragicultura e Pastagens, submetendo-se à defesa de tese em 16 de fevereiro de 2004.

Em Março de 2004, iniciou o curso de doutorado em Zootecnia, na Universidade Federal de Viçosa, realizando um estágio na Texas A & M University no período de janeiro de 2006 a maio de 2007, submetendo-se à defesa de tese em 28 de setembro de 2007.

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RESUMO

CHIZZOTTI, Fernanda Helena Martins, D.Sc., Universidade Federal de Viçosa, setembro de 2007. **Níveis de nitrogênio não-protéico e silagens de diferentes híbridos de milho na dieta de bovinos de corte.** Orientador: Odilon Gomes Pereira. Co-Orientadores: Sebastião de Campos Valadares Filho e Rasmão Garcia

O presente trabalho foi desenvolvido a partir de quatro experimentos, os quais foram realizados na Central de Experimentação, Pesquisa e Extensão do Triângulo Mineiro (CEPET) – UFV, de abril a julho de 2004. Os experimentos 1 e 2 foram conduzidos com os objetivos de avaliar níveis de nitrogênio não-protéico (NNP) sobre o desempenho, consumo e digestibilidade de nutrientes, pH e amônia ruminais, balanço de nitrogênio e eficiência de síntese de proteína microbiana em novilhos cruzados (Holandês x Zebu). No experimento 1, foram utilizados 24 novilhos, com peso inicial médio de 350 ± 20 kg, distribuídos em seis blocos casualizados para avaliação do consumo e digestibilidade dos nutrientes, bem como do desempenho dos mesmos. As dietas consistiram de 70% de silagem de milho e 30% de concentrado e foram formuladas para conter 12,5% de PB na base da matéria seca. Os tratamentos consistiram de 0; 15,5; 31 e 46,5% de NNP em relação ao N dietético. Não houve efeito dos níveis de NNP ($P > 0,05$) sobre os consumos de matéria seca (MS), matéria orgânica (MO), proteína bruta (PB), carboidratos não-fibrosos (CNF) e nutrientes digestíveis totais (NDT), enquanto que o consumo de fibra em detergente neutro (FDN) decresceu linearmente ($P = 0,02$) com o aumento dos níveis de NNP na dieta. Também não foram observados efeitos dos níveis de NNP ($P > 0,05$) sobre as digestibilidades totais aparentes de MS, MO, FDN e CNF. A digestibilidade total aparente da PB incrementou linearmente ($P = 0,01$) em decorrência do aumento dos níveis de NNP enquanto que o ganho médio diário (GMD) não foi influenciado ($P = 0,96$) pelos tratamentos, registrando-se valor médio de 1,14 kg/d. No experimento 2, foram utilizados quatro novilhos com peso médio inicial de 300 ± 55 kg, fistulados no rúmen e abomaso, distribuídos em um quadrado latino 4 x 4, para avaliação de níveis de NNP sobre o consumo e digestibilidade dos nutrientes, características ruminais, balanço de nitrogênio e eficiência microbiana. As dietas e os tratamentos utilizados foram os mesmos descritos no experimento 1. Os consumos de MS, MO, PB, FDN e NDT não foram influenciados pelos níveis de NNP ($P > 0,05$). Entretanto, houve aumento linear no

consumo de CNF ($P = 0,02$) e efeito quadrático positivo ($P = 0,01$) no consumo de extrato etéreo (EE) com o aumento dos níveis de NNP. A digestibilidade ruminal da PB aumentou linearmente ($P = 0,01$) com o aumento dos níveis de NNP na dieta. Não houve efeito dos níveis de NNP sobre os valores de pH ruminal ($P > 0,05$). Por outro lado, a concentração de N-amônia ruminal foi influenciada de forma quadrática ($P = 0,013$), pela interação entre tempo de coleta e níveis de NNP registrando-se valores máximos de 14,6; 19,5; 18,1 e 25,4 mM às 3,3; 3,5; 3,1 e 3,8 h após a alimentação dos animais com dietas que continham 0; 15,5; 31 e 46,5% de NNP, respectivamente. Houve incremento linear ($P = 0,01$) na excreção urinária de nitrogênio e de uréia com o aumento dos níveis dietéticos de NNP. O balanço de nitrogênio, expresso em % do N ingerido e em % do N digerido, também aumentou linearmente ($P < 0,05$) com o incremento de NNP na dieta. Não houve efeito dos níveis de NNP ($P > 0,05$) sobre a eficiência de síntese protéica microbiana. A inclusão de NNP em até 46,5% do N dietético pode ser utilizada para novilhos cruzados (H x Z) com ganhos de peso próximos de 1,0 kg/d, recebendo dietas à base de silagem de milho sem afetar o desempenho e a eficiência de síntese microbiana. Entretanto, a eficiência de utilização do nitrogênio diminuiu significativamente. Os experimentos 3 e 4 foram realizados num esquema fatorial 2 x 2 para avaliar silagens de dois híbridos de milho: Agromen (AGN35-A42) e Bayer (A3663) e dois níveis de concentrado (25 e 50%) sobre o desempenho animal, o consumo e a digestibilidade dos nutrientes, características ruminais, balanço de nitrogênio e eficiência microbiana em novilhos cruzados (H x Z). O experimento 3 foi conduzido com 24 novilhos, com peso inicial médio de 335 ± 30 kg, distribuídos em seis blocos casualizados, para avaliar o consumo e digestibilidade de nutrientes, bem como o desempenho animal. Os tratamentos consistiram em 75% de silagem de milho A + 25% de concentrado (A25); 50 % de silagem de milho A + 50% de concentrado (A50); 75% de silagem de milho B + 25% de concentrado (B25); 50 % de silagem de milho B + 50% de concentrado (B50); na base da MS e foram formuladas para serem isonitrogenadas, contendo 13% de PB na base da MS. Não houve efeito dos tratamentos ($P > 0,05$) sobre os consumos de MS, MO e PB. Entretanto, houve efeito de níveis de concentrado sobre o consumo de FDN, o qual foi menor para novilhos alimentados com 50% de concentrado em relação aos alimentados com 25% ($P < 0,0001$). Adicionalmente, houve efeito de níveis de concentrado sobre os consumos de CNF e NDT ($P < 0,01$), com maiores consumos

observados em animais recebendo dietas com maior proporção de concentrado. O consumo de EE foi influenciado pelo tipo de silagem ($P = 0,02$) sendo observado maiores valores para a silagem do híbrido Agromen. Não foram observados efeitos dos tratamentos ($P > 0,05$) sobre a digestibilidade total aparente de PB, EE, FDN e CNF. Por outro lado, houve efeito de níveis de concentrado sobre a digestibilidade total aparente da MS ($P = 0,02$) e MO ($P = 0,01$) sendo observado maiores valores em animais recebendo dietas com 50% de concentrado. Entretanto, não foi observado efeito dos tratamentos ($P > 0,05$) sobre o GMD e conversão alimentar, registrando-se valores médios de 1,10 kg/d e 8,13, respectivamente. No experimento 4, foram utilizados quatro novilhos, com peso médio inicial de 512 ± 25 kg, fistulados no rúmen e abomaso, distribuídos num quadrado latino 4×4 , alimentados com as mesmas dietas do experimento 3. Foram avaliados os consumos e as digestibilidades dos nutrientes, as características ruminais, o balanço de nitrogênio e a eficiência de síntese microbiana. Não houve diferenças ($P > 0,05$) nos consumos médios de MS, MO, PB e EE. Entretanto, os consumos de FDN, CNF e NDT foram afetados pelos níveis de concentrado ($P < 0,05$), sendo reportados maiores consumos de CNF e NDT e menor consumo de FDN em animais alimentados com dietas contendo 50% em relação aos alimentados com 25%. A digestibilidade aparente total da MS, MO e o teor de NDT das dietas foram afetadas positivamente pelos níveis de concentrado ($P < 0,05$), apresentando maiores valores para dietas contendo 50% de concentrado. As digestibilidades ruminais e intestinais de MS, MO, PB, EE, FDN e CNF não foram influenciadas ($P > 0,05$) pelos tratamentos. Da mesma forma, não houve efeito dos tratamentos sobre os valores de pH ruminal e concentração de N amônia ruminal ($P > 0,05$). Adicionalmente, o balanço de N, bem como a eficiência de síntese microbiana também não foram influenciados pelos tratamentos ($P > 0,05$). O uso de silagem de milho de ambos híbridos avaliados, associada a 25% de concentrado é uma boa opção para alimentação de novilhos cruzados (H x Z) com potencial de ganho diário de peso em torno de 1,0 kg, propiciando desta forma, menor gasto com ração concentrada. A associação de silagem de milho de ambos híbridos avaliados com 25 ou 50% de concentrado não altera o pH e N amônia ruminais, o balanço de nitrogênio, bem como a eficiência de síntese microbiana.

ABSTRACT

CHIZZOTTI, Fernanda Helena Martins, D.Sc., Universidade Federal de Viçosa, September of 2007. **Non protein nitrogen levels and different corn silage hybrids in beef cattle diets.** Adviser: Odilon Gomes Pereira. Co-Advisers: Sebastião de Campos Valadares Filho and Rasmô Garcia

The present work was developed based on four experiments that were conducted at the experimental, research, and extension center (CEPET) of the Federal University of Viçosa, Brazil, during April to July of 2004. Experiments 1 and 2 were conducted aiming to evaluate the effects of dietary non protein nitrogen (NPN) levels on animal performance, intake and digestibility of nutrients, ruminal characteristics, N balance and microbial protein synthesis in crossbred steers (Holstein x Zebu). Exp. 1 was conducted with 24 Holstein x Zebu crossbred steers (350 ± 20 kg of BW) distributed in 6 randomized blocks to evaluate intake and digestibility of nutrients and performance. The diets consisted of 70% corn silage and 30% concentrate, and were formulated to be 12.5% CP (DM basis). Treatments consisted of 0, 15.5, 31, and 46.5% of NPN of dietary N. There were no treatment differences in the daily intakes of DM ($P = 0.47$), OM ($P = 0.60$), CP ($P = 0.24$), non-fiber carbohydrates (NFC) ($P = 0.74$), and TDN ($P = 0.63$); however, NDF intake decreased linearly as NPN increased ($P = 0.02$). Additionally, no effects of NPN were observed on apparent total tract digestibility of DM ($P = 0.50$), OM ($P = 0.53$), NDF ($P = 0.63$), and NFC ($P = 0.44$). The CP apparent digestibility increased linearly ($P = 0.01$) but ADG (1.14 kg/d) was not influenced ($P = 0.96$) as NPN increased. In Exp. 2, 4 ruminally and abomasally cannulated steers (300 ± 55 kg of BW) were used in a 4×4 latin square design and fed with the same diet used in exp. 1 to evaluate the effects of NPN levels on intake and digestibility of nutrients, ruminal characteristics, N balance, and microbial efficiency. There were no differences in the daily intakes of DM ($P = 0.22$), OM ($P = 0.17$), CP ($P = 0.31$), NDF ($P = 0.29$) and TDN ($P = 0.49$). However, NFC intake increased linearly ($P = 0.02$) and there was a quadratic effect ($P = 0.01$) on EE intake as NPN increased. Ruminal digestibility of CP increased linearly ($P = 0.01$) with the increase of dietary NPN. There was no NPN effect on ruminal pH values ($P > 0.05$). On the other hand, ammonia-N concentration was affected quadratically ($P = 0.013$) by sampling times by NPN levels

interactions with maximum ammonia N of 14.6, 19.5, 18.1, and 25.4 mM at 3.3, 3.5, 3.1 and 3.8 h after feeding to diets with 0, 15.5, 31, and 46.5% of NPN, respectively. As dietary NPN increased, urinary N and urea increased linearly ($P = 0.01$). The N balance as % of ingested, and, as % of digested also had linear increase as dietary NPN levels increased. There were no differences ($P \geq 0.28$) on microbial protein synthesis and microbial efficiency among the treatments. Dietary NPN levels up to 46.5% of total N can be fed to crossbreds steers (Holstein x Zebu) with ADG close to 1.0 kg, receiving corn silage-based diets without affecting performance and ruminal protein synthesis. However, efficiency of N utilization may decrease significantly. The third and fourth experiments were realized involving a 2 x 2 factorial arrangement of treatments to evaluate the effects of corn silages hybrids Agromen (AGN35-A42) and Bayer (A3663) and concentrate levels (25 and 50%) on animal performance, digestibility, ruminal characteristics, N balance and microbial efficiency on crossbred steers. Exp. 3 was conducted with 24 Holstein x Zebu crossbred steers, averaging 335 ± 30 kg of BW, distributed in six randomized blocks to evaluate intake and digestibility of nutrients and performance. Treatments consisted of 75% of corn silage A + 25% of concentrate (A25), 50 % of corn silage A + 50% of concentrate (A50), 75% of corn silage B + 25% of concentrate (B25), 50 % of corn silage B + 50% (B50), on DM basis, and were formulated to be isonitrogenous (13% CP, DM basis). There were no treatment differences ($P > 0.05$) in the daily intakes of DM, OM, and CP. However, there was a concentrate effect on NDF intake which was lower to steers fed 50% of concentrate than those fed 25% ($P < 0.0001$). Additionally, there was a concentrate effect on NFC and TDN intakes ($P < 0.01$), with higher intakes to steers fed diets with more concentrate. The ether extract (EE) intake was affected by silage ($P = 0.02$) and was higher to corn silage hybrid A than corn silage hybrid B. No treatments effects ($P > 0.05$) were observed on apparent total digestibility of CP, EE, NDF, and NFC. On the other hand, there was a concentrate effect on total apparent digestibility of DM ($P = 0.02$) and OM ($P = 0.01$), which were greater to steers fed diets with 50% of concentrate. However, ADG (1.10 kg/d) and feed efficiency (8.13) were not influenced ($P > 0.05$) by treatments. In Exp. 4, four ruminal and abomasal cannulated steers (512 ± 25 kg of BW), were used in a 4 x 4 latin square design and fed with the same diet used in the exp. 3. The intake and digestibility of nutrients, ruminal characteristics, N balance, and microbial efficiency were evaluated. There were no differences ($P > 0.05$) in the daily intakes of DM, OM,

CP, and EE. However, the intakes of NDF, NFC, and TDN were affected ($P < 0.05$) by concentrate levels, with greater intakes of NFC and TDN and lower intake of NDF to steers fed diets with 50% of concentrate than those fed 25%. The total digestibility of DM, OM and the content of TDN of diets were affected by concentrate levels ($P < 0.05$), which were greater to diets with 50% of concentrate than those with 25%. Ruminal and intestinal tract digestibility of DM, OM, CP, EE, NDF and NFC were not affected ($P > 0.05$) by treatments. In the same way, there were no effects of treatments on ruminal pH and ruminal ammonia-N concentration ($P > 0.05$). In addition, N balance and microbial efficiency were not affected by treatments ($P > 0.05$). The utilization of either corn silage hybrids evaluated in association with 25% of concentrate is a good option to feed crossbreeds steers (Holstein x Zebu) with ADG close to 1.0 kg, resulting in a reduction of diet cost. The association of either corn silage hybrids evaluated with 25 or 50% of concentrate not affects ruminal pH and ruminal ammonia-N concentration, N balance, and microbial efficiency.

INTRODUÇÃO

Atualmente, a constante busca pelo aumento da eficiência de produção de carne no país, devido à crescente competitividade, se tornou condição básica para a sobrevivência da atividade pecuária.

A alimentação animal compreende a maior parcela dos custos de produção da carne bovina, e dentre os nutrientes, a proteína constitui a fração das rações que possui custo relativo mais elevado (Velloso, 1984). Dessa forma, a alimentação torna-se o principal componente do sistema produtivo a ser criteriosamente analisado com o intuito de se aliar a qualidade do alimento a sua economicidade.

A substituição das tradicionais fontes de proteína verdadeira, como os farelos de soja e algodão por fontes de nitrogênio não protéico (NNP), como a uréia, tem se constituído em boa alternativa de redução dos custos da alimentação de bovinos confinados. Devido ao seu baixo custo por unidade de nitrogênio, facilidade de utilização e disponibilidade no mercado, a uréia tem sido a fonte de NNP mais utilizada (Santos et al., 2001).

De acordo com Salman et al. (1997), o uso da uréia é questionado por alguns pesquisadores devido à sua aceitabilidade pelos animais, toxicidade e pela baixa quantidade de proteína não degradada no rúmen, que, juntamente com a proteína microbiana, podem não ser suficientes para atender às necessidades de animais jovens com elevada taxa de ganho de peso. A recomendação tradicionalmente adotada pela maioria dos pesquisadores é que o NNP pode substituir até 33% do nitrogênio protéico da dieta dos ruminantes, ou que a quantidade de uréia seja limitada em até 1% na matéria seca total da dieta (Reid, 1953; Chalupa, 1968).

Entretanto, estudos realizados com níveis de uréia acima dos recomendados não registraram efeitos prejudiciais aos animais (Thomas et al., 1984; Hussein & Berger, 1995; Shain et al., 1998; Knaus et al., 2001; Souza et al., 2002; Magalhães et al. 2005; Rennó et al., 2005). Portanto, torna-se necessária uma avaliação mais detalhada sobre níveis elevados de uréia na dieta de bovinos, uma vez que ainda não foi evidenciado qual o nível máximo de inclusão, a partir do qual o desempenho animal poderia ser negativamente afetado.

Valadares Filho et al. (2004), em trabalho de revisão, concluíram que a uréia pode substituir totalmente os farelos protéicos em dietas para bovinos alimentados com níveis moderados de concentrados e com potencial para aproximadamente 1

kg de ganho por dia, propiciando considerável redução no custo da alimentação dos animais.

Entretanto, na literatura, poucos são os estudos que avaliaram o impacto de se utilizar altos níveis de uréia (NNP) sobre a excreção de compostos nitrogenados. Tal avaliação, juntamente com a análise de N-uréico no plasma, irá permitir um melhor entendimento da utilização da uréia nas rações, uma vez que altos níveis de N na urina, assim como altos níveis de uréia na urina e no plasma sanguíneo, demonstram uma ineficiente utilização do N dietético pelos animais.

No rúmen, a uréia é rapidamente hidrolisada em amônia devido a urease produzida por bactérias ruminais (Bloomfield et al., 1960). A amônia é utilizada para incorporação no nitrogênio microbiano, sendo a disponibilidade de energia o principal fator determinante de sua assimilação. De acordo com Nolan (1993), a produção de amônia no rúmen muitas vezes excede a sua utilização, o que resulta em acúmulo e, conseqüentemente, em sua posterior remoção do ambiente ruminal, principalmente por difusão através da parede ruminal. Essa amônia é transportada para o fígado, onde é convertida em uréia a qual é liberada no sangue, podendo ser excretada na urina ou reciclada para o rúmen, retornando via saliva ou através da difusão pelo epitélio ruminal (Coelho da Silva & Leão, 1979).

Assim, a concentração plasmática de uréia é diretamente relacionada à ingestão de compostos nitrogenados. Portanto, quando o animal é submetido a uma alimentação que proporcione uma elevada concentração de N ruminal em relação aos carboidratos, esse sistema resulta em grandes concentrações de uréia sanguínea e grandes perdas de N urinário. Tal situação não é interessante economicamente, pois além do gasto energético para a formação da uréia, em aproximadamente 12 kcal/g de nitrogênio (Van Soest, 1994), há perdas de compostos nitrogenados, um componente oneroso na alimentação animal, conforme já mencionado anteriormente.

Outro problema causado pelo excesso de N na dieta ou assincronia entre a liberação de N e energia no rúmen é a questão da poluição ambiental. Segundo Broderick (2003), a uréia constitui a principal forma de N urinário, e uma vez excretada na urina é rapidamente convertida em amônia, a qual pode ser extensivamente volatilizada, contribuindo assim para a poluição do ambiente. De acordo com Van Horn et al. (1994), em solução aquosa, a amônia (NH₃) reage com H⁺ formando o íon não gasoso amônio (NH₄⁺). Assim, em um ambiente ácido esta

reação ocorre de forma rápida, o que diminuiu significativamente a volatilização de amônia para a atmosfera. Entretanto, a maioria dos dejetos de animais possui pH básico (maior que 7.0), o que é insuficiente para converter amônia em amônio e, conseqüentemente, as perdas por volatilização podem ser elevadas. De acordo com alguns autores (Pain et al., 1988; Tamminga, 1996; Van Horn et al., 1996), o excesso de amônia na atmosfera contribui para a ocorrência de chuva ácida.

Por outro lado, quando as perdas por volatilização são pequenas, ou seja, quando a conversão de amônia em amônio não for favorecida por condições ambientais, o amônio será nitrificado em nitrato (NO_3^-). Devido a sua alta solubilidade e baixa energia de ligação com os colóides do solo, o excesso de nitrato no solo pode contaminar águas superficiais e lençóis freáticos.

Além disso, durante os processos microbiológicos no solo de nitrificação (amônio em nitrato) e denitrificação (nitrato em gás dinitrogênio), o óxido nitroso (N_2O), que é um dos gases intermediários destes processos, pode ser liberado na atmosfera. Segundo Saggari et al. (2004), o óxido nitroso contribui para destruição da camada de ozônio, possui tempo de permanência na atmosfera de 150 anos, além de ser considerado um gás estufa significativamente mais efetivo que o CO_2 , contribuindo assim para o aquecimento global.

Neste contexto, torna-se extremamente importante que a utilização do nitrogênio na dieta dos animais seja feita de forma mais eficiente, evitando perdas e, conseqüentemente, o desequilíbrio no ciclo geoquímico deste nutriente causado pelo excesso do mesmo no meio ambiente.

Embora a questão ambiental, no que diz respeito a excreção de nitrogênio, ainda não seja uma preocupação no Brasil, devido principalmente ao predominante sistema de produção extensivo, em outros países essa questão é de suma importância, uma vez que sistemas intensivos, como grandes confinamentos, contribuem de forma significativa para poluição do ambiente. Dessa forma, o problema ambiental (notadamente o aquecimento global e a poluição dos lençóis freáticos) tem merecido destaque mundial e sido foco de inúmeras pesquisas, o que, futuramente, poderá influenciar na tomada de decisão quanto ao uso da uréia na alimentação dos ruminantes.

Hoje em dia, a tomada de decisão quanto à utilização de NNP, como a uréia, na dieta dos animais, se restringe à relação custo/benefício que seu uso irá gerar. Porém, a eficiência com que esse nitrogênio será aproveitado também deve ser

levada em consideração, evitando dessa forma, que o animal gaste energia para excretar o excesso de nitrogênio, um ingrediente oneroso da dieta.

Portanto, a recomendação do nível de NNP (uréia) na dieta irá depender do critério utilizado para definir uma ótima eficiência de uso do nitrogênio, uma vez que os níveis de uréia requeridos para reduzir custos da dieta, e, ou, maximizar a performance e a eficiência de síntese microbiana, podem não ser os mesmos níveis necessários para minimizar a poluição ambiental.

Outra questão importante quanto à formulação de dietas para ruminantes é o uso da quantidade de ração concentrada. Como a alimentação é responsável por grande parte dos custos de produção nos sistemas de confinamento, a condução criteriosa dos programas de alimentação exige o respaldo de estudos para o conhecimento, com maior precisão, das interações e dos impactos produzidos pelo emprego do concentrado na alimentação de bovinos. Além disso, a escolha da proporção de forragem conservada:concentrado na dieta deve ser bioeconomicamente viável para se obter sucesso no sistema de produção.

Existem na literatura, tanto nacional quanto internacional, vários estudos onde níveis de concentrado foram avaliados na dieta de bovinos de corte. Entretanto, os resultados experimentais são bastante variáveis, o que pode ser decorrência de variações entre estudos, como tipo de animal, tipo de volumoso utilizado, idade dos animais, questões ambientais, etc. Embora muitos resultados de pesquisas mostrem que o ganho de peso diário é maior quando se utilizam rações com maior porcentagem de concentrado, a resposta animal à adição de concentrado tende a ser quadrática (Gesualdi Júnior et al., 2000). Portanto, o estudo de diferentes proporções de concentrado nas rações ainda é necessário, pois permite determinar seu nível ótimo, para que se obtenha o melhor desempenho animal aliado à melhor resposta econômica. Neste contexto, a qualidade do volumoso torna-se de fundamental importância para que se obtenha bons resultados econômicos, uma vez que, o uso de volumosos de boa qualidade implica em menores gastos com ração concentrada.

No Brasil, o número de animais que são alimentados com volumosos conservados durante pelo menos um período de sua vida produtiva vem aumentando a cada dia. Neste contexto, a conservação de espécies forrageiras, principalmente na forma de silagem, tem obtido destaque entre os pecuaristas, uma vez que é uma prática indispensável para manter uma oferta constante de produto

animal durante todo o ano. Segundo McDonald et al. (1991), o interesse pela silagem é grande principalmente devido a sua menor dependência das condições climáticas, quando comparada à confecção do feno.

Entre as opções de forrageiras para ensilagem, o milho, considerado padrão, se destaca por apresentar produtividade de matéria seca elevada, bom valor nutritivo e boa digestibilidade (Gomes, 2002). O grande potencial do milho para ensilagem deve-se, também, ao seu elevado teor de carboidratos solúveis e baixa capacidade tampão, garantindo, assim, adequada fermentação no interior do silo.

Existem inúmeros relatos na literatura nacional que confirmam a qualidade das silagens de milho por meio de pesquisas envolvendo animais (Pilar et al., 1994; Ferreira et al., 1995; Silva et al., 2001; Souza et al., 2006).

Com base em resultados obtidos nos trabalhos descritos na literatura, pode-se observar que dietas à base de silagens de milho ou sorgo, sobressaem-se sobre os demais tipos de volumosos conservados, o que pode ser atribuído ao maior valor energético dessas silagens (principalmente a de milho), necessitando assim, de um menor aporte de concentrado na dieta. Tal fato contribui para a redução do custo de produção de arrobas em confinamento, uma vez que a ração concentrada constitui uma fração onerosa da dieta, conforme já relatado anteriormente.

Recentemente, foi realizado por Pereira et al. (2007) um estudo de simulação bioeconômica de diferentes fontes de forragens conservadas para bovinos de corte em confinamento. Os autores concluíram que, independentemente do nível de ganho de peso dos animais, as dietas à base de silagens de milho e sorgo foram as opções mais atrativas economicamente, tanto sob aspectos do desempenho animal, quanto por área. Além disso, foram as únicas que apresentaram saldos positivos com a alimentação, o que explica a maior preferência destes volumosos (principalmente a silagem de milho) pela maioria dos pecuaristas.

Uma vez que o milho é considerado uma forrageira padrão para confecção de silagem, a busca por melhores híbridos é extremamente importante. Entretanto, como apenas 10% das sementes comercializadas anualmente são utilizadas para a produção de silagem, não há grande interesse das empresas em desenvolver novos híbridos com boas características para este fim. Esta constatação leva a necessidade de estudar os híbridos novos que são lançados no mercado para a produção de grãos e avaliar o seu potencial para a produção de silagem (Neumann et al., 2003). Entretanto, devido aos grandes avanços genéticos, a rotatividade de

novos híbridos no mercado é muito rápida, dificultando a avaliação adequada de cada um destes em ensaios com animais.

Dentre os fatores que interferem na quantidade e no valor nutricional da forragem produzida, destaca-se a cultivar ou híbrido de milho utilizado. Tem sido observada em diversos estudos a existência de ampla variabilidade entre híbridos, tanto para produtividade de matéria seca como para qualidade (Melo et al., 1999; Oliveira et al., 2003). Segundo Gomes et al. (2000), a escolha da cultivar de milho para produção de silagem deve incluir tanto aspectos agronômicos da cultura, como características bromatológicas e nutricionais.

Entre os inúmeros estudos encontrados na literatura, com relação à caracterização agronômica produtiva e qualitativa de diferentes híbridos de milho, há um consenso, entre os autores, da necessidade do estudo das respostas dos diferentes materiais genéticos referentes ao consumo de alimentos e desempenho animal.

Portanto, é de suma importância que além da avaliação agronômica dos diferentes híbridos de milho existentes, estes sejam também avaliados em ensaios com animais, propiciando, desta forma, maior segurança aos produtores na tomada de decisão, no que se refere à alimentação de seus rebanhos.

Os híbridos de milho AGN35-A42 (Agromen) e A3663 (Bayer) são encontrados facilmente no mercado sendo indicados para produção de grãos e para silagem, embora pesquisas envolvendo a avaliação destes na alimentação de animais em confinamento, sob mesma condição, sejam muito escassas.

O AGN35-A42 é um híbrido duplo, de ciclo superprecoce (820 graus dias), possui textura do grão semiduro, requer nível tecnológico baixo a médio, e é indicado para plantio em todo o Brasil. Por outro lado, o A3663 é um híbrido triplo, de ciclo precoce (925 graus dias), com textura do grão semiduro, requer nível tecnológico médio e é indicado para plantio nas regiões sudeste, nordeste, centro oeste e sul (Cruz e Pereira Filho, 2006).

Conforme discutido anteriormente, é importante que as características agronômicas sejam levadas em consideração na escolha da cultivar a ser adotada, uma vez que pequenas diferenças entre as cultivares podem ter um impacto significativo dentro de um sistema de produção. Segundo Cruz e Pereira Filho (2006), os híbridos triplos são potencialmente mais produtivos que os duplos, apresentando maior uniformidade de plantas e espigas, embora apresentem

sementes mais caras. Tais informações podem ser tornar essenciais na escolha da cultivar.

Outra característica que pode ser importante na escolha da cultivar é o tipo de ciclo fenológico da mesma. O ciclo de uma cultivar leva em consideração as unidades de calor necessárias para atingir o florescimento. As cultivares normais apresentam exigências térmicas maior do que 890 graus-dias (GD), as precoces, de 830 a 890 GD, e as superprecoces, menor do que 830 GD (Cruz e Pereira Filho, 2006). Assim, em situações onde se deseja efetuar sucessão de culturas ou plantios escalonados, a escolha do ciclo adequado da cultivar pode torna-se imprescindível.

Portanto, a escolha da cultivar mais adequada deve levar em consideração as informações disponíveis nas empresas produtoras de sementes, na assistência técnica e na pesquisa, de forma a ajustar a semente escolhida ao sistema de produção desejado.

Diante do exposto, conduziu-se o presente trabalho com os objetivos de:

- Avaliar o consumo e a digestibilidade aparente total de nutrientes e o desempenho animal, em novilhos Holandês x Zebu submetidos a dietas com níveis crescentes de nitrogênio não-protéico;
- Avaliar o consumo e a digestibilidade aparente total e parcial de nutrientes, determinar o pH e a concentração de amônia ruminais, estimar a produção de proteína microbiana, a concentração de uréia plasmática e a excreção de compostos urinários em novilhos Holandês x Zebu, submetidos a dietas com níveis crescentes de nitrogênio não-protéico;
- Avaliar o consumo e a digestibilidade aparente total de nutrientes e o desempenho animal, em novilhos Holandês x Zebu alimentados com dietas à base de silagens de dois híbridos de milho e dois níveis de concentrado;
- Avaliar o consumo e a digestibilidade aparente total e parcial de nutrientes, determinar o pH e a concentração de amônia ruminais, a produção de proteína microbiana e a excreção de compostos urinários em novilhos Holandês x Zebu alimentados com dietas à base de silagens de dois híbridos de milho e dois níveis de concentrado;

Essa tese foi redigida no formato de artigos científicos. O artigo científico 1 foi redigido de acordo com as normas do Journal of Animal Science e o artigo científico 2 de acordo com as normas do periódico Animal Feed Science and Technology.

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**Effects of dietary NPN on performance, digestibility, ruminal characteristics,
and nitrogen metabolism in crossbred steers ¹**

F. H. M. Chizzotti^{*,†}, O. G. Pereira^{*}, L.O. Tedeschi^{†,2}, S. C. Valadares Filho^{*}, M. L. Chizzotti^{*,†}, M. I. Leão^{*}, D. H. Pereira^{*}

^{*} Universidade Federal de Viçosa, Viçosa, MG 36571, Brazil

[†] Texas A&M University, College Station, TX 77843, USA

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² Correspondence: Phone: (979) 845-5065, Fax (979) 845-5292; email: luis.tedeschi@tamu.edu

ABSTRACT: Two trials were conducted to evaluate the effects of dietary NPN levels on animal performance, digestibility, ruminal characteristics, and N metabolism. Exp. 1 was conducted with 24 Holstein x Nellore crossbred steers (350 ± 20 kg of BW) distributed in 6 randomized blocks to evaluate intake and digestibility of nutrients and performance. The diets consisted of 70% corn silage and 30% concentrate, and were formulated to be 12.5% CP (DM basis). Treatments consisted of 0, 15.5, 31, and 46.5% of NPN of dietary N. There were no treatment differences in the daily intakes of DM ($P = 0.47$), OM ($P = 0.60$), CP ($P = 0.24$), non-fiber carbohydrates (**NFC**) ($P = 0.74$), and TDN ($P = 0.63$); however, NDF intake decreased linearly as NPN increased ($P = 0.02$). Additionally, no effects of NPN were observed on apparent

total tract digestibility of DM ($P = 0.50$), OM ($P = 0.53$), NDF ($P = 0.63$), and NFC ($P = 0.44$). The CP apparent digestibility increased linearly ($P = 0.01$) but ADG (1.14 kg/d) was not influenced ($P = 0.96$) as NPN increased. In Exp. 2, 4 ruminally and abomasally cannulated steers (300 ± 55 kg of BW) were fed the same diet used in Exp. 1 to evaluate the effects of NPN levels on intake and digestibility of nutrients, ruminal characteristics, N balance, and microbial efficiency. There were no differences in the daily intakes of DM ($P = 0.22$), OM ($P = 0.17$), CP ($P = 0.31$), NDF ($P = 0.29$) and TDN ($P = 0.49$). However, NFC intake increased linearly ($P = 0.02$) and there was a quadratic effect ($P = 0.01$) on EE intake as NPN increased. Ruminal digestibility of CP increased linearly ($P = 0.01$) with the increase of dietary NPN. As dietary NPN increased, urinary N and urea increased linearly ($P = 0.01$). There were no differences ($P \geq 0.28$) on microbial protein synthesis and microbial efficiency among the treatments. The results of these trials suggested that dietary NPN levels (up to 46.5% of total N) can be fed to crossbreds steers receiving corn silage-based diets without affecting ruminal protein synthesis, but efficiency of N utilization may decrease significantly.

Keywords: feedlot, nitrogen balance, non-protein nitrogen, protein supplementation

INTRODUCTION

True protein supplements are expensive ingredients in diets for dairy and beef cattle. Therefore, partial or total substitution of a true protein supplement with a NPN source can significantly reduce feeding cost. Urea is the most commonly used NPN source in cattle diets due to availability and low cost. Urea dissolves quickly in water and is rapidly hydrolyzed to ammonia because of rumen microbial urease activity

(Bloomfield et al., 1960). Consequently, asynchrony of N and digestible energy occurs because of rapid N release from urea and excess ammonia will not be used.

According to early studies, urea can be effectively utilized when dietary inclusion is limited to one third of supplemental N or 1% of dietary DM (Reid, 1953; Chalupa, 1968). In contrast, other studies (Souza, 2004; Rennó et al., 2005) have demonstrated that performance was not affected either when high urea levels (1.5 to 1.95% of dietary DM, approx. 30 and 45.8% of total N as NPN, from urea/ammonium sulfate, respectively) were added in the diet or when supplemental true protein was replaced with urea. However, few experiments have been designed to identify the amount of dietary NPN needed for maximum animal performance, ruminal fermentation, and metabolizable protein supply in cattle consuming corn silage-based diets while evaluating N excretion. Therefore, determining the correct levels of dietary NPN required for optimum N use by ruminal microbes would allow adequate performance, thereby improving feed efficiency, reducing feed costs, and N losses to the environment.

The objectives of this study were to evaluate effects of dietary NPN levels in steers consuming corn silage-based diets on intake, growth, nutrient digestion, microbial protein synthesis, ruminal characteristics, and efficiency of N utilization.

MATERIALS AND METHODS

Location and Diets

The experiments were conducted at the Federal University of Viçosa, Brazil, during April to July of 2004. Diets were the same for Exp. 1 and Exp. 2, and were formulated to provide increasing levels of dietary NPN. Treatments consisted of 0, 15.5, 31, and 46.5% of dietary N as NPN (Table 1). The 2 sources of added NPN

were urea and ammonium sulfate. In treatment diets, urea, ammonium sulfate, and ground corn replaced cottonseed meal of the control diet. Diets consisted of 70% corn silage and 30% concentrate, formulated to be 12.5% CP (DM basis). Animal care and handling procedures followed Federal University of Viçosa guidelines.

Exp. 1

Animals and Sampling Protocol. Twenty-four Nellore x Holstein crossbred steers (350 ± 20 kg BW) were distributed in 6 randomized blocks to evaluate intake and digestibility of nutrients and their performance in the feedlot. Steers were blocked into 6 groups based on initial BW and allotted randomly to 1 of 4 treatments (6 steers per treatment). The animals were treated for internal and external parasites at the beginning of the experiment and kept in individual pens of approximately 10 m², with protected feeders and water. The experiment was conducted for 99 d (15 d for diet adaptation and 3 periods of 28 d for data collection).

Steers were individually fed ad libitum twice daily at 0700 and 1500. Diets were fed as a total mixed ration in which corn silage and concentrate (previously mixed) were weighed and mixed before feeding. Orts were collected and weighed once daily and diets were adjusted daily to yield Orts of about 5 to 10% of offered. Animals had free access to water. Feed ingredients and Orts were sampled daily and composited by weight and within a period.

For each animal, DMI was measured daily and grab samples of feces (around 200 g) were collected between d 14 and 16 of the second period with collection intervals of 28 h. Indigestible ADF (**iADF**) was used as an internal marker to estimate apparent nutrient digestibility and fecal output. After drying at 60°C for 72 h, feed, Ort, and fecal samples were ground to pass a 1-mm screen (Willey mill) and period

composites per steer were prepared. Composite samples were used to determine DM (method 934.01; AOAC, 1990); OM determined by ash (method 924.05; AOAC, 1990); CP obtained by total N determination using the micro Kjeldahl technique (method 920.87; AOAC, 1990) and a fixed conversion factor (6.25); EE determined gravimetrically after extraction using petroleum ether in a Soxhlet instrument (method 920.85; AOAC, 1990); NDF (Van Soest et al., 1991); ADF (method 973.18; AOAC, 1990), and sulfuric acid lignin (Robertson and Van Soest, 1981). The NDF and ADF were not corrected for ash or protein. The iADF (ADF remaining after a 144 h in situ incubation in a rumen-cannulated cow) was determined according to Cochran et al. (1986) and the digestibility of nutrients was calculated as:

$$\text{Digestibility, \%} = 100 \times [100 - (\text{dietary iADF, \%} / \text{fecal iADF, \%}) \times (\text{fecal nutrient, \%} / \text{dietary nutrient, \%})]$$

Non-fiber carbohydrate (NFC; %) was calculated by difference as:

$$\text{NFC} = 100 - (\text{CP, \%} + \text{NDF, \%} + \text{EE, \%} + \text{ash, \%}),$$

except for diets containing urea and ammonium sulfate where NFC was calculated as (Hall, 2000):

$$\text{NFC} = 100 - [(\% \text{CP} - \% \text{CP from urea} + \% \text{ of urea}) + \% \text{NDF} + \% \text{EE} + \% \text{ash}].$$

Apparent TDN (%) was calculated as (NRC, 2001):

$$\text{Apparent TDN} = (\text{digestible CP} + \text{digestible NDF} + \text{digestible NFC} + (2.25 \times \text{digestible EE})) / \text{DMI}.$$

The NPN percentage of feed samples was determined through precipitation of true protein by trichloroacetic acid, and subsequent filtration and determination of the

insoluble N in the residue (Licitra et al., 1996). The NPN was calculated as the difference between total N and precipitated true protein N.

Animal Performance. The ADG was calculated as the difference between the final and initial shrunk BW (16 h of fasting, only water was provided) divided by the number of days of feeding (84 d). Gain efficiency was calculated as ADG divided by DMI.

Exp. 2

Animals and Sampling Protocol. Four Holstein steers (300 kg \pm 55 kg of BW) fitted with abomasal and ruminal cannulas were used in a 4 \times 4 Latin square design to evaluate intake and apparent total tract and partial digestibilities of nutrients, ruminal pH and ammonia concentration, rumen microbial protein synthesis, plasma urea, and urinary excretion of nitrogenous compounds. Each experimental period was 20 d: 10 d for adaptation to the diet, 6 d to collect fecal and abomasal samples, 1 d for ruminal pH measurements and collection of ruminal fluid samples, 1 d for urine collection, 1 d for blood collection, and 1 d to collect ruminal contents to isolate bacteria. The experiment was conducted for 80 d (4 periods of 20 d).

Steers were assigned randomly to 4 dietary treatment sequences and fed individually ad libitum twice daily (0700 and 1500). Diets were fed as total mixed rations as described previously. Orts were collected and weighed once daily and feed offered was adjusted daily to yield Orts of about 5 to 10% of total offered. Animals had free access to water. Feed ingredients and Orts were sampled daily and composited by weight for each steer within each period.

Feces and abomasal digesta samples (approximately 200 g and 500 mL, respectively) were collected between d 11 and 16 of each period with intervals of 26 h between the samplings. Abomasal fluid subsamples were preserved with 1 mL of 50% (vol/vol) of H₂SO₄, and stored at -20°C for analysis of NH₃. Abomasal fluid NH₃ was analyzed by distilling with 2 N KOH in a micro-Kjeldahl system, after previous centrifugation at 1,000 g for 15 min, according to the original procedures of Fenner (1965) and adaptations of Vieira (1980).

Fecal and abomasal samples were dried in a forced draft oven (60°C for 72 h), and then ground to pass a 1-mm screen. Fecal and abomasal samples were composited on a DM basis to obtain one representative composite sample for each steer within each period. Composite samples of feeds, orts, feces, and abomasal digesta were analyzed for total N, DM, ash, OM, EE, NDF, and iADF as described above.

Ruminal contents (100 mL) were obtained at 0, 1, 2, 4, 6 and 8 h after the morning feeding on d 17 of each period and subsequently strained through 2 layers of cheesecloth. The pH was measured immediately. The ruminal fluid was preserved by addition of 1 mL of 50% (vol/vol) of H₂SO₄, and stored at -20°C for analyses of NH₃. Ruminal fluid NH₃ was analyzed according to the original procedures of Fenner (1965) and adaptations of Vieira (1980), as described above.

Estimation of Urinary N excretion. Urine was collected for 24 h on d 18 of each experimental period, with a rubber funnel system attached to the ventral portion of the abdomen to collect urine into a plastic carboy. The 1 d urine collection was adopted based on a previous work that reported no differences on the daily urine output and urinary purine derivatives and creatinine excretions across collection periods of 24, 48 and 72 h (Valadares et al., 1997).

A solution of 500 mL of H₂SO₄, 20% (vol/vol) was previously added in the plastic carboys to reduce pH of urine (pH < 2) to prevent ammonia loss. At the end of the collection, one aliquot of urine (50 mL) was obtained for each animal and stored at -20°C. Frozen urine samples were thawed at room temperature and filtered (Whatman #1) to remove contaminants. Filtrates were analyzed for total N (micro Kjeldahl technique, method 920.87; AOAC, 1990) and for urea with the enzymatic-colorimetric technique using a commercial kit (Uréia CE, Labtest Diagnóstica S.A., Lagoa Santa, Brazil) which is based on urease reaction according to Bergmeyer (1985).

On d 19 of the experimental period and 4 h after feeding, blood was collected from the coccygeal artery or vein of each steer into blood collection tubes containing heparin. All samples were placed on ice, centrifuged at 1,500 g for 15 min to obtain plasma, and frozen at -20°C until urea analysis by the colorimetric assay used for urine.

Determination of Microbial Protein Synthesis. On d 20, the ruminal contents were obtained 4 h post-feeding and squeezed through 2 layers of cheesecloth to yield about 1,500 mL of strained fluid. Particles retained on the cheesecloth were mixed with 500 mL of 9 g of NaCl/L, blended for 1 min, refiltered through cheesecloth, and added to the 1.5 L ruminal fluid sample. Bacteria were isolated by differential centrifugation (500 g and 27,000 g) according to procedures of Cecava et al. (1990). The resulting bacterial pellets were dried at 60°C for 48 h and ground in a ball mill (TE350, Tecnal, Piracicaba, Brazil). The ground bacterial samples were analyzed for DM, ash, and total N according to procedures described earlier, and total purines were determined (Ushida et al., 1985). To quantify microbial protein and subsequently determine microbial efficiency, approximately 400 mg of

dry abomasal digesta were used, which also were analyzed for purines according to Ushida et al. (1985).

Statistical Analyses

Exp. 1. Intake, digestibility, and performance were analyzed as a randomized complete block design using the MIXED procedure of SAS (SAS Inst. Inc.). The model included the fixed effects of treatment and random effects of blocks. Linear, quadratic, and cubic effects of dietary NPN were tested using orthogonal contrasts. The statistical model is described below.

$$Y_{ijk} = \mu + b_i + t_j + e_{ijk}$$

Where Y_{ijk} was the measured variable, μ was the overall mean, b_i was the random effect of the i^{th} block, assuming identical, independent, and normal distribution $(0, \sigma_b^2)$, t_j was the fixed effect of the j^{th} treatment, and e_{ijk} was the residual error. Treatment differences were considered to be significant when $P \leq 0.05$.

Exp. 2. The data of digestibility, intake, N metabolism, and microbial protein efficiencies were analyzed as a 4×4 Latin square design (Kuehl, 2000) using the MIXED procedure of SAS (SAS Inst. Inc.) and the statistical model was:

$$Y_{ijkl} = \mu + \alpha_i + b_j + c_k + e_{ijkl}$$

Where Y_{ijkl} was the variable measured, μ was the overall mean, α_i was the fixed effect of the i^{th} treatment; b_j was the random effect of the j^{th} animal assuming identical, independent, and normal distribution $(0, \sigma_b^2)$; c_k was the random effect of the k^{th} period assuming identical, independent, and normal distribution $(0, \sigma_c^2)$; and e_{ijkl} was the residual error. Linear, quadratic, and cubic effects of NPN levels were tested using orthogonal contrasts. Treatment differences were considered to be significant when $P \leq 0.05$ and were considered to indicate a trend at $0.05 < P \leq 0.10$.

The ruminal characteristics data collected over time were analyzed as repeated measures (Kuehl, 2000) using the MIXED procedure of SAS (SAS Inst. Inc.). Model effects in the whole plot were animal, period, and treatment, whereas subplot effects were sampling time and treatment × sampling time interactions as shown below.

$$Y_{ijklm} = \mu + \alpha_i + b_j + p_k + \varepsilon_{ijk} + z_l + z\alpha_{li} + \omega_{ijklm},$$

Where Y_{ijklm} was the dependent variable, μ was the overall mean, α_i was the fixed effect of the i^{th} treatment; b_j was the random effect of the j^{th} animal; p_k was the random effect of the k^{th} period; ε_{ijk} was whole plot error, z_l was the effect of time; $z\alpha_{li}$ was interaction between time and treatment, and ω_{ijklm} was the subplot error. When treatment interacted ($P < 0.05$) with sampling time, variables were analyzed within time periods. The variance-covariance structure AR(1) was used for estimating covariances and the subject of the repeated measurements were defined as animal (period × treatment). Differences were considered to be significant when $P \leq 0.05$.

RESULTS AND DISCUSSION

Diet Composition

Diets (Table 1) provided similar amounts of DM, OM, CP, EE, and NDF to all steers. Calculated ruminally degraded protein (**RDP**) and NPN percentages increased as urea and ammonium sulfate were added to the diets, creating a variation in the amount of available N in the rumen that was received by steers as the protein supplement. Additionally, as cottonseed meal was replaced by urea and ammonium sulfate, ground corn was also added to the diets increasing dietary NFC.

Feed Intake and Performance

There were no differences ($P \geq 0.24$) in the daily intakes of DM, OM, CP, NFC, and TDN among treatments in the Exp. 1 (Table 2). The NDF intake decreased linearly ($P = 0.01$) as dietary NPN increased due to lesser dietary NDF with more NPN (Table 1) and numerical decrease in DMI. In Exp. 2, there were differences ($P \leq 0.02$) among the treatments only in NFC and EE intakes (Table 3). The NFC intake increased linearly ($P = 0.02$) with NPN due to a higher NFC percentage in those diets with more NPN, which contained greater amounts of ground corn (Table 1) and there was a quadratic effect ($P = 0.05$) on DMI (% of BW) as NPN increased. There are many studies with contrasting results about urea levels (NPN) in cattle diets. Rennó et al. (2005) observed no effect of dietary urea (0 to 1.95%) on intake of nutrients in steers of 4 different genetic groups fed diets with 50% of bermudagrass hay. Similarly, Souza (2004) evaluated the effects of dietary urea (0, 0.5, 1.0, or 1.5%) on intake and performance in steers fed 70% sorghum silage-based diets and reported no differences on intake of nutrients. On the other hand, Milton et al. (1997) observed a cubic effect on DMI, which was smaller for steers consuming 0.5 and 1.5% urea than for those fed 0 and 1.0% urea in steers fed 90% of concentrate diets. However, these authors reported that DMI responded quadratically to dietary urea level (0, 0.35, 0.70, 1.05, and 1.40%, DM basis); the maximum DMI was observed in steers consuming 1.05% urea and a decrease of the 0.7 kg of DMI/d was observed with 1.40% urea.

There were no effects ($P \geq 0.46$) of dietary NPN on ADG and gain efficiency (Table 4). These results are consistent with nutrients intake data because no differences were found between intakes of the majority of dietary nutrients. Results similar to ours were reported by Souza (2004), who found no differences in ADG among steers fed 70% sorghum silage-based diets containing urea up to 1.5% of

DM. Similarly, Shain et al. (1998) evaluated the effect of dietary urea (0, 0.88, 1.34, or 1.96%, DM) in steers fed 90% concentrate diets, and reported no differences in DMI, ADG, and gain efficiency. In contrast, Milton et al. (1997) observed that the optimal dietary urea level was 0.5% for ADG and feed efficiency when fed a 90% concentrate diet. Zinn et al. (2003) evaluated the influence of urea supplementation level on growth performance of steers fed 90% concentrate diets and observed that ADG was optimized by dietary inclusion of 0.8% urea. Gleghorn et al. (2004) evaluated effects of CP concentration (11.5, 13, and 14% of dietary CP) and degradability (100% urea and 0% cottonseed meal; 50% urea and 50% cottonseed meal; and 100% cottonseed meal) on performance of beef steers fed 90% concentrate diets, and no differences in ADG were observed among CP sources.

In our experiment, the ratio of NFC:RDP was similar among the diets (4.8, 4.9, 4.7, and 4.7 for diets with 0, 15.5, 31 and 46.5% NPN). This was due to the replacement of cottonseed meal (little starch) with urea and ground corn (lots of starch), and consequently all diets had energy to use the supplemental N. Furthermore, the lack of differences on majority of nutrients intake and animal performance possibly was due to this constant ratio of NFC:RDP.

Diet Digestibility

In Exp. 1, no effects ($P \geq 0.24$) of NPN levels were observed on apparent total tract digestibility of DM, OM, NDF, NFC and on dietary TDN (Table 5), which were on average, 70.1, 71.3, 54.0, 86.8, and 70.9% respectively. The linear increase in the CP apparent total tract digestibility with increasing levels of dietary NPN is likely due to greater absorption of NPN as ammonia than digestibility and absorption of cottonseed meal N.

Similarly, in the Exp. 2, NPN levels had no effect ($P \geq 0.31$) on apparent total tract digestibility of DM, OM, EE and NDF, and consequently, the TDN percentage of the diets also was not influenced by treatments (Table 6). The apparent digestibility of CP showed a positive linear tendency ($P = 0.09$) and apparent digestibility of NFC had a quadratic behavior ($P = 0.04$) as NPN levels increased. Rennó et al. (2005) reported no differences among levels of dietary urea (up to 45.9% NPN of total N) on total tract digestibility of several nutrients in steers fed bermudagrass hay. Accordingly, there were no differences ($P \geq 0.11$) on apparent ruminal and intestinal tract digestibility of DM, OM, EE, NDF, and NFC among the treatments (Table 7). As expected, urea level linearly increased ($P = 0.01$) ruminal disappearance of CP likely because of absorption of NPN as ammonia prior to the abomasum. Similar result was reported by Milton et al. (1997) who observed linear increases in apparent ruminal N digestion evaluating the effects of dietary urea levels up to 1.5% on site and extent of digestion in steers. Furthermore, these authors observed that all diets had negative coefficients for apparent ruminal N digestion, indicating a large amount of N being recycled, especially when steers were fed the basal diet (no urea). In our trial this decrease in ruminal N digestion did not occur maybe due to the excess of total dietary NPN, which would lead to a surplus of N or a possible asynchrony between energy and N available. Although the diets had a similar ratio NFC:RDP, the percentage of NPN was different, what likely resulted in faster release of ammonia than energy in those diets with more NPN. Therefore, these results suggested an imbalance over time between available energy and N and consequently a positive apparent CP ruminal digestibility due to the ruminal absorption of the N excess.

Ruminal Characteristics and Nitrogen Utilization

There were no effects of the NPN levels ($P = 0.14$) and treatment by sampling time interactions ($P = 0.58$) on ruminal pH. Overall, the mean ruminal pH value observed was 6.03, which is greater than the 5.0 to 5.5 range that was suggested by Hoover (1986) in which ruminal digestibility of fiber is negatively affected. In fact, the apparent ruminal tract digestibility of NDF was not affected by the variation of NPN levels in the diets (Table 7).

As expected, the ruminal NH_3 concentration was affected by the interaction of sampling times by treatment ($P = 0.013$), by treatment ($P < 0.0001$), and by time after feeding ($P < 0.0001$). The sampling times by treatment interactions (Figure 1) showed a quadratic behavior with maximum ammonia of 14.6, 19.5, 18.1, and 25.4 mM at 3.3, 3.5, 3.1 and 3.8 h after feeding to diets with 0, 15.5, 31, and 46.5% of NPN, respectively. During all time sampling the ruminal NH_3 concentrations were well above levels recommended to optimize ruminal digestion (Satter and Slyter, 1974), although the concentration of ruminal NH_3 necessary for optimal ruminal digestion on various diets is not well defined yet.

Nevertheless, these results suggested that the increase of supplementation with NPN resulted in an accumulation of ruminal NH_3 , indicating that microbial requirements for NH_3 were exceeded, or ruminal microbes were not able to utilize the N either because energy was first limiting or microbial growth was slower than the solubilization of N. Milton et al. (1997) and Shain et al. (1998) also observed that by increasing urea levels in 90% concentrate diets, ruminal NH_3 concentration increased linearly.

Total N flow to the abomasum and non-ammonia N flow decreased linearly with increasing dietary NPN ($P \leq 0.05$), whereas $\text{NH}_3\text{-N}$ increased linearly ($P =$

0.003). The increase of NH₃-N flow probably occurred because of NPN excess in the rumen; therefore, contributing to a greater concentration in the abomasum. On the other hand, the total N and NAN flow to the abomasum decreased due to the greater soluble N and greater RDP in diets with increasing levels of NPN.

Fecal N excretion was not affected ($P \geq 0.14$) by dietary NPN. The inclusion of urea and ammonium sulfate in the diet resulted in a linear increase ($P = 0.01$) of urinary N. Consequently, urea-N excretion increased linearly ($P = 0.01$) as NPN increased. Therefore, the main route for N excretion was via urine. According to Broderick (2003), urea is the main form of urinary N that can be rapidly converted to ammonia by microbial urease and volatilized, contributing to environmental pollution (Van Horn et al., 1994).

Consistently, the plasma urea concentrations increased linearly ($P = 0.01$) with addition of NPN in the diets. During periods of excessive N availability in the rumen, NH₃ is absorbed and appears as urea in the plasma urea pool (Cocimano and Leng, 1967). This N may then be recycled back to the rumen during subsequent periods of N shortage or excreted in the urine. Our results of the ruminal ammonia, urinary N and urea excretion, and plasma urea N presumably reflect the less-efficient total N utilization that could be a result of an excessive supply of NPN. This excessive supply of NPN would lead to a surplus of ammonia in the rumen that occurred in this trial, increasing urea synthesis. These results are in agreement with previous observations by Rennó (2003) and Magalhães et al. (2005), who used diets similar to ours and reported that plasma urea was increased when NPN (urea) was added to the diets. Cecava and Hancock (1994) also determined that urinary N and plasma urea were greater for steers fed 60% corn silage-based diets with urea than steers receiving combinations of soybean meal and feather meal.

The N balance (g/d) was not influenced by treatments, showing only a tendency ($P = 0.07$), but decreased linearly when expressed as a percentage of N intake ($P = 0.04$) and N digested ($P = 0.01$). The lack of difference in the N balance (g/d) can be occurred due to low numbers of animals per treatments ($n = 4$) and the limitations of 1-d urine collection technique, which might have contributed to increase the urinary N variations and consequently, the N balance data. The linear reduction of efficiency of N retention observed with addition of NPN in the diets could be explained by the increased rate of urinary N excretion which was consequence of excess ruminal ammonia as discussed above. Rennó (2003) observed a linear decrease in the N balance, % of N intake, when steers were fed with 50% Bermudagrass hay with increasing levels of dietary urea (up to 45.8 % NPN, total N). Accordinally, Cecava and Hancock (1994) reported a lower N retention in steers fed diets with 1.72% urea, DM than those fed diets with soybean meal and feather meal. However, Knaus et al. (2001) observed no difference on the N balance (g/d and % of N intake) among Holstein steers fed diets with urea (1.8%, DM), those fed soybean meal and those fed combinations of urea, meat and bone meal, fish meal, hydrolyzed feather meal, and blood meal.

Ruminal Fermentation and Microbial Efficiency

Dietary NPN levels had a quadratic effect ($P = 0.03$) on rumen-degraded OM while rumen-degraded carbohydrate only showed a tendency ($P = 0.06$). Bacterial growth is largely dependent on the amount of ammonia and fermentable OM available in the rumen (Bryant and Robinson, 1962). Readily fermentable carbohydrate, such as starch or sugars, is more effective than other carbohydrate sources, such as cellulose, in promoting microbial growth (Stern and Hoover, 1979).

Microbes that degrade structural carbohydrate (cellulolytic) have low maintenance requirements, grow slowly, and use ammonia as their main N source, whereas microorganisms that degrade nonstructural CHO (amylolytic) have high maintenance requirements, grow rapidly, and use ammonia, peptides, and amino acids as N sources (Russell et al., 1992).

Although rumen-degraded OM changed quadratically as dietary levels of NPN increased and ruminal NH_3 was not limiting, microbial CP production (Table 9) was not affected by treatment. Moreover, microbial efficiency was not influenced by dietary NPN ($P \geq 0.11$) when expressed on a rumen-degraded OM, rumen-degraded carbohydrate, or TDN basis. As discussed previously, diets had a similar NFC:RDP ratio; however, the percentage of NPN increased, which likely resulted in faster release of ammonia than energy in those diets with more NPN. This could have limited microbial protein synthesis. Our results suggested that due to the accumulation of ruminal NH_3 microbial requirements for NH_3 might have been exceeded, or ruminal microbes were not able to utilize the N either because energy was first limiting or microbial growth was slower than the solubilization of N. Either or both might have limited microbial production and (or) efficiency.

Zinn et al. (2003) evaluated the influence of urea supplementation (0, 0.4, 0.8, and 1.2%, DM) on digestive function of cattle and observed no effect on flow of rumen microbial N to the small intestine. Indeed, microbial efficiency (g of microbial N/kg of truly fermented OM) decreased linearly with increasing urea level. Milton et al. (1997) observed that flows of total and microbial N to the duodenum and microbial efficiency were not affected by dietary urea.

IMPLICATIONS

These findings suggested that dietary NPN (up to 46.5% of total N) can be fed to crossbreds steers receiving corn silage-based diets without affecting ruminal protein synthesis. However, efficiency of N utilization may decrease significantly because of feeding an excess of NPN, will not improve animal performance and may cause excessive nitrogen excretion and ammonia volatilization into the environment.

Nevertheless, the recommended level of dietary NPN depends on the criteria used to define optimum N utilization because NPN levels required to reduce diet costs, to maximize performance and microbial efficiency may not match those required to minimize environmental N load.

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Table 1. Ration composition and analyzed content of diets fed in Exp. 1 and 2

Item	Dietary NPN, % of total N			
	0	15.5	31	46.5
Corn silage, %	70.00	70.00	70.00	70.00
Ground corn, %	11.63	16.79	21.82	26.99
Cottonseed meal, %	17.60	11.73	5.88	-
Urea, %	-	0.65	1.30	1.95
Ammonium sulfate, %	-	0.06	0.13	0.19
Sodium chloride, %	0.25	0.25	0.25	0.25
Dicalcium phosphate, %	-	-	0.10	0.20
Calcitic limestone, %	0.50	0.50	0.50	0.40
Mineral premix ¹ , %	0.02	0.02	0.02	0.02
	Analyzed content of diets ²			
DM, %	49.8	49.9	50.1	50.3
OM, % of DM	95.0	94.5	93.8	95.2
CP, % of DM	12.7	12.6	12.5	12.4
RDP ³ , % of CP	64.5	67.6	73.9	81.8
NPN ⁴ , % of N	26.8	39.7	52.6	66.3
NDF, % of DM	40.9	39.7	38.4	37.2
ADF, % of DM	22.4	21.3	20.3	19.3
iADF, % of DM	10.0	9.25	8.49	7.73
EE, % of DM	1.60	1.58	1.57	1.55
NFC, % of DM	39.7	41.8	43.7	47.6
Lignin, % of DM	3.30	3.13	2.97	2.81
Ash, % of DM	5.0	5.5	6.2	4.8

¹Composition (%): cooper sulfate (22.50), cobalt sulfate (1.40), zinc sulfate (75.40), potassium iodate (0.50), and sodium selenite (0.20).

²OM = organic matter, RDP = ruminally degraded protein, NPN = non-protein N, ADF = acid detergent fiber, iADF = indigestible ADF, EE = ether extract, and NFC = non-fiber carbohydrates.

³Calculated using ingredients kd values of NRC (2001) and $K_p = 4.2\%/h$ which was estimated in Exp. 2.

⁴Proportion of total N soluble in 10% (wt/vol) trichloroacetic acid (Licitra et al., 1996).

Table 2. Effect of dietary NPN on of nutrient intake in Exp. 1^a

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^b		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
Intake, kg/d								
DM	10.35	9.80	9.85	9.37	0.42	0.14	0.94	0.56
OM	9.66	9.32	9.39	8.90	0.38	0.22	0.86	0.59
CP	1.29	1.23	1.23	1.11	0.05	0.06	0.68	0.47
EE	0.26	0.27	0.26	0.23	0.009	0.07	0.04	0.62
NDF	4.05	3.79	3.62	3.44	0.16	0.01	0.78	0.90
NFC	4.12	4.09	4.34	4.18	0.17	0.58	0.72	0.38
TDN	7.18	6.91	7.20	6.63	0.35	0.40	0.68	0.37
Intake, % of BW								
DM	2.26	2.17	2.20	2.13	0.07	0.30	0.83	0.50
NDF	0.89	0.84	0.81	0.78	0.03	0.02	0.83	0.94

^aOM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bProbability of a linear, quadratic or cubic effect of NPN level of the diet.

Table 3. Effect of dietary NPN on nutrient intake in Exp. 2^a

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^b		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
Intake, kg/d								
DM	9.20	9.90	10.06	9.59	0.82	0.31	0.07	0.93
OM	8.70	9.39	9.62	9.15	0.79	0.22	0.06	0.84
CP	1.21	1.28	1.29	1.21	0.09	0.96	0.08	0.82
EE	0.18	0.22	0.21	0.18	0.02	0.95	0.01	0.59
NDF	3.51	3.78	3.67	3.47	0.34	0.68	0.09	0.60
NFC	3.80	4.12	4.43	4.29	0.33	0.02	0.11	0.47
TDN	6.28	6.75	6.71	6.72	0.54	0.27	0.37	0.61
Intake, % of BW								
DM	2.20	2.32	2.39	2.24	0.08	0.46	0.05	0.53
NDF	0.84	0.88	0.87	0.81	0.03	0.38	0.11	0.92

^aOM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

Table 4. Effect of dietary NPN on ADG, and gain efficiency in Exp. 1

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^a		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
ADG, kg/d	1.14	1.13	1.10	1.18	0.10	0.83	0.66	0.76
Gain efficiency, g/kg	110.2	114.6	111.0	126.4	7.8	0.20	0.53	0.46

^aProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

Table 5. Effect of dietary NPN on apparent total tract digestibility of nutrients and dietary TDN in Exp. 1^a

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^b		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
DM	68.9	69.4	71.7	70.2	1.4	0.32	0.48	0.36
OM	70.4	70.5	72.9	71.3	1.4	0.40	0.51	0.32
CP	65.6	65.1	70.9	69.4	1.5	0.01	0.73	0.04
EE	82.9	86.9	85.1	82.1	2.4	0.51	0.03	0.50
NDF	53.5	52.3	54.8	55.3	1.8	0.35	0.65	0.46
NFC	86.9	88.1	87.3	84.8	1.4	0.29	0.21	0.99
TDN	69.4	70.4	73.0	70.9	1.4	0.24	0.24	0.30

^aValues are expressed as %. OM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

Table 6. Effect of dietary NPN on apparent total tract digestibility of nutrients and dietary TDN in Exp. 2^a

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^b		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
DM	69.0	67.7	67.1	69.6	1.9	0.86	0.28	0.75
OM	70.0	69.5	68.3	71.2	1.8	0.73	0.33	0.52
CP	64.4	65.8	67.0	69.1	2.8	0.09	0.86	0.87
EE	78.6	82.6	75.1	76.8	4.7	0.52	0.79	0.32
NDF	53.1	54.8	53.6	56.2	2.6	0.50	0.88	0.59
NFC	86.7	83.5	80.4	83.6	1.5	0.07	0.04	0.31
TDN	68.2	68.3	67.2	69.9	1.7	0.61	0.47	0.54

^aValues are expressed as %. OM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

Table 7. Effect of dietary NPN on apparent ruminal and intestinal tract digestibility of nutrients in Exp. 2^a

Item	Dietary NPN, % of total N				SEM	<i>P</i> -value ^b		
	0	15.5	31	46.5		Linear	Quadratic	Cubic
Ruminal digestibility								
DM ^c	74.7	77.9	75.4	72.5	2.9	0.51	0.34	0.72
OM ^{ac}	78.7	80.4	78.3	74.8	2.5	0.27	0.35	0.85
CP ^c	35.4	47.5	51.2	58.2	5.4	0.01	0.61	0.60
EE ^{ac}	-5.5	4.3	-7.4	-8.1	15.3	0.30	0.22	0.11
NDF ^c	89.1	93.5	90.4	90.1	3.0	0.99	0.45	0.46
NFC ^{ac}	86.8	84.4	81.6	73.3	4.3	0.06	0.52	0.80
Intestinal digestibility								
DM ^c	25.3	22.1	24.6	27.5	2.9	0.51	0.34	0.72
OM ^{ac}	21.3	19.6	21.7	25.2	2.5	0.27	0.35	0.85
CP ^c	64.6	52.5	48.8	41.8	5.4	0.01	0.61	0.60
EE ^{ca}	105.5	95.7	107.4	108.1	15.3	0.30	0.22	0.11
NDF ^c	10.9	6.5	9.6	9.9	2.9	0.99	0.45	0.46
NFC ^{ac}	13.2	15.6	18.4	26.7	4.3	0.06	0.52	0.80

^aOM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

^cDigestibility calculated as % of total digestion

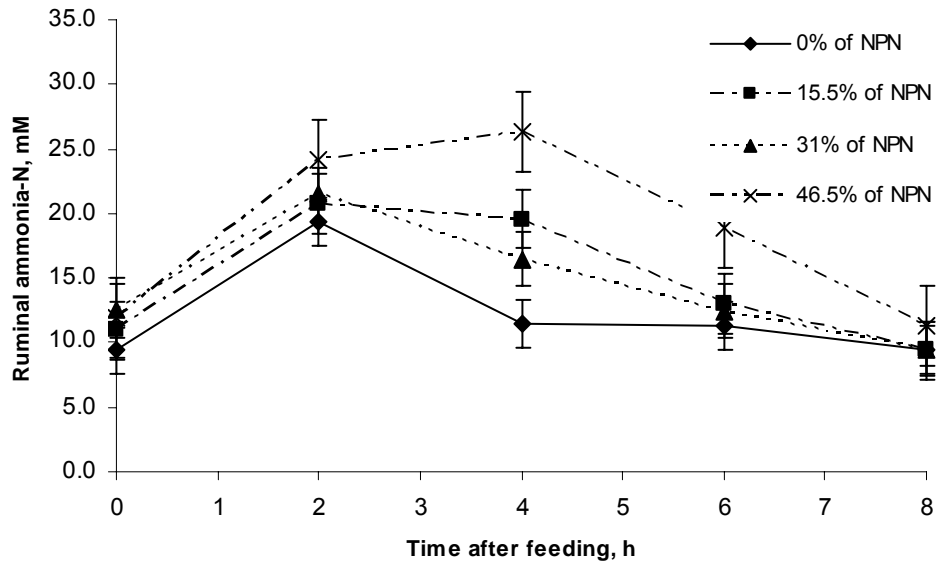


Figure 1. Ruminal ammonia-N concentrations (means + SE) after feeding.

◆ $\text{NH}_3\text{-N} = -0.2786x^2 + 1.896x + 11.47$; $R^2 = 0.37$; $\text{SE} = 4.3$;

■ $\text{NH}_3\text{-N} = -0.5775x^2 + 4.079x + 12.28$; $R^2 = 0.82$; $\text{SE} = 4.9$;

▲ $\text{NH}_3\text{-N} = -0.4091x^2 + 2.518x + 14.22$; $R^2 = 0.70$; $\text{SE} = 5.0$;

× $\text{NH}_3\text{-N} = -0.8819x^2 + 6.734x + 12.72$; $R^2 = 0.94$; $\text{SE} = 7.4$.

Table 8. Effects of dietary NPN on balance of nitrogen in the Exp. 2

Item	Dietary NPN, % of total N					P-value ^a		
	0	15.5	31	46.5	SEM	Linear	Quadratic	Cubic
N intake, g/d	193.7	204.2	206.1	193.5	15.5	0.96	0.08	0.82
Plasma urea, mM	7.7	10.7	10.9	15.0	1.4	0.01	0.69	0.34
	Abomasal flow, g/d							
Total N	150.0	142.0	137.7	115.1	16.7	0.05	0.50	0.64
NH ₃ -N	1.58	2.86	3.09	3.40	0.74	0.003	0.11	0.36
Non-NH ₃ -N ^b	148.5	139.1	134.6	111.7	16.2	0.05	0.53	0.62
Microbial N	118.9	117.7	117.5	106.7	13.8	0.28	0.52	0.73
	N excretion							
Fecal N, g/d	68.6	69.1	68.7	59.0	6.7	0.14	0.23	0.63
Urinary N, g/d	46.7	57.9	68.2	79.9	11.1	0.01	0.97	0.94
Urinary Urea-N, g/d	20.3	31.7	32.3	48.0	5.9	0.01	0.65	0.26
	N balance							
N balance, g/d	78.6	77.3	69.3	54.6	11.7	0.07	0.43	0.99
N balance, % of N intake	40.1	37.0	34.3	28.9	5.0	0.04	0.71	0.82
N balance, % of N digested	61.7	55.7	50.9	42.3	6.5	0.01	0.75	0.79

^aProbability of a linear, quadratic, or cubic effect of NPN level of the diet.

^bNon-NH₃-N = Total N abomasal flow - NH₃-N abomasal flow

Table 9. Effect of dietary NPN on microbial N efficiency in the Exp. 2

Item	Dietary NPN, % of total N				SEM	Linear	P-value ^d	
	0	15.5	31	46.5			Quadratic	Cubic
Rumen degradability, kg/d								
OM	4.77	5.24	5.08	4.86	0.36	0.84	0.03	0.35
CHO ^a	4.44	4.80	4.59	4.35	0.32	0.42	0.06	0.39
Microbial N								
g/kg of RDOM ^b	25.0	22.2	22.8	21.8	1.9	0.19	0.55	0.46
g/kg of RDCHO ^c	27.1	24.3	25.3	24.7	2.2	0.48	0.57	0.53
g/kg of TDN	120.0	116.5	107.5	102.8	8.3	0.08	0.93	0.74

^aCHO = 100 - (%CP + %EE + %Ash), Sniffen et al. (1992);

^bRuminally degraded organic matter.

^cRuminally degraded carbohydrates.

^dProbability of a linear, quadratic or cubic effect of urea NPN of the diet

Performance, digestibility, ruminal characteristics, and nitrogen balance in crossbred steers fed diets based on two corn silage hybrids and concentrate levels

F. H. M. Chizzotti^{1,2,#}, O. G. Pereira¹, S. C. Valadares Filho¹, L.O. Tedeschi², M. I. Leão¹, K. G. Ribeiro³, D. H. Pereira¹

¹ Universidade Federal de Viçosa, Viçosa, MG 36571, Brazil

² Texas A&M University, College Station, TX 77843, USA

³ Universidade Federal dos Vales do Jequitinhonha e Mucuri, Diamantina, MG 39100, Brazil

Correspondence: Phone: (55) (31) 3892-5283, Fax (979) 845-5292; email: fernandachizzotti@yahoo.com.br

ABSTRACT: Two trials involving a 2 x 2 factorial arrangement of treatments were conducted to evaluate the effects of corn silage hybrids (Agromen, AGN35-A42) and (Bayer, A3663) and concentrate levels (25 and 50%) on animal performance, digestibility, ruminal characteristics, N balance and microbial efficiency on crossbred steers. Exp. 1 was conducted with 24 Holstein x Zebu crossbred steers, averaging 335 ± 30 kg of BW, distributed in six randomized blocks to evaluate intake and digestibility of nutrients and performance. Treatments consisted of 75% of corn silage A + 25% of concentrate (A25), 50 % of corn silage A + 50% of concentrate (A50), 75% of corn silage B + 25% of concentrate (B25), 50 % of corn silage B + 50% (B50), on DM basis, and were formulated to be isonitrogenous (13% CP, DM basis). There were no treatment differences ($P > 0.05$) in the daily intakes of DM, OM, and

CP. However, there was a concentrate effect on NDF intake which was lower to steers fed 50% of concentrate than those fed 25% ($P < 0.0001$). Additionally, there was a concentrate effect on NFC and TDN intakes ($P < 0.01$), with higher intakes to steers fed diets with more concentrate. The ether extract (EE) intake was affected by silage ($P = 0.02$) and was higher to corn silage hybrid A than corn silage hybrid B. No treatments effects ($P > 0.05$) were observed on apparent total digestibility of CP, EE, NDF, and NFC. On the other hand, there was a concentrate effect on total apparent digestibility of DM ($P = 0.02$) and OM ($P = 0.01$), which were greater to steers fed diets with 50% of concentrate. However, ADG (1.10 kg/d) and feed efficiency (8.13) were not influenced ($P > 0.05$) by treatments. In the Exp. 2, four ruminal and abomasal cannulated steers (512 ± 25 kg of BW), were used in a 4×4 latin square design and fed with the same diet used in the Exp. 1 to evaluate the intake and digestibility of nutrients, ruminal characteristics, N balance, and microbial efficiency. There were no differences ($P > 0.05$) in the daily intakes of DM, OM, CP, and EE. However, the intakes of NDF, NFC, and TDN were affected ($P < 0.05$) by concentrate levels, with greater intakes of NFC and TDN and lower intake of NDF to steers fed diets with 50% of concentrate than those fed 25%. The total digestibility of DM, OM and the percentage of TDN of diets were affected by concentrate levels ($P < 0.05$), which were greater to diets with more concentrate. Ruminal and intestinal tract digestibility of DM, OM, CP, EE, NDF and NFC were not affected ($P > 0.05$) by treatments. In the same way, there were no effects of treatments on ruminal pH values and ruminal ammonia-N concentration ($P > 0.05$). In addition, N balance and microbial efficiency also were not affected by treatments ($P > 0.05$). The utilization of either corn silage hybrids evaluated in association with 25% of concentrate is a good option to feed crossbreds steers (Holstein x Zebu) with ADG close to 1.0 kg,

resulting in a reduction of diet cost. The association of both corn silage hybrids evaluated with 25 or 50% of concentrate not affects the ruminal pH and ruminal ammonia-N concentration, N balance, and microbial efficiency.

Keywords: feedlot, microbial protein synthesis, roughage, total digestible nutrients

INTRODUCTION

The feeding is the most expensive component of feedlot systems, mainly the concentrate ration cost. Thus, interactions and impacts of use of different forage and concentrate ratios are extremely important to analyze the optimal relationship between cost and animal performance. Many studies have showed contradictory results about concentrate levels in beef cattle diets, so researches regarding concentrate ratio is yet necessary. In addition, the quality of silage used also is fundamental to get good economic results, because of using good quality silage, the utilization of concentrate can be decreased and, consequently, the diet cost will be lower.

Whole-plant corn silage is a popular forage source for ruminants due to its high yielding properties, energy content, relatively high palatability, and incorporating easily into total mixed ration. Furthermore, the plant corns have high water soluble carbohydrate content, adequate lactic acid production and, consequently, results in good quality silage.

However, hybrid, maturity, and moisture content are some of the factors that can alter the nutritive value of corn silage (Johnson et al., 2002a). Several studies

have shown differences between hybrids in nutrient composition of whole plant corn and yield of DM (Johnson et al., 1985; Hunt et al., 1993; Qiu et al., 2003; Xu et al., 1995; Melo et al., 1999; Oliveira et al., 2003). Commercial corn hybrids that are to be harvested for silage have been selected on the basis of agronomic traits such as grain yield and disease resistance (Bal et al., 2000; Clark et al., 2002) and differences in the nutritive value of whole plant corn silages related to corn genetics have been ignored (Bal et al., 2000). Therefore, the choice of hybrid for silage should include agronomic traits, evaluation of nutritive value as well as the intake and performance of animals fed those silages.

The commercial corn hybrids AGN35-A42 (Agromen) and A3663 (Bayer) are indicated for both corn grain and whole-plant corn silage although feeding trials to evaluate animal performance differences involving these corn silage hybrids at the same environment condition are limited.

The objective of this study was to determine the effects of two corn silage hybrids and two concentrate levels on intake, digestibility, animal performance, ruminal characteristics, N balance, and microbial protein synthesis in crossbred steers.

2. Materials and Methods

2.1 Location and climatic conditions

The study was conducted at the experimental, research, and extension center (CEPET) of Federal University of Viçosa, Brazil, during April to July of 2004. The CEPET is located at latitude 18° 41' S, and longitude 49° 34' W, with an average altitude of 620.2m. According to Köppen (1948), the climate is classified as type Aw, hot and humid, with coldest month temperatures above 18°C; annual average

precipitation between 1400 and 1600 mm, rain season in the summer and dry season in the winter.

2.2 Corn hybrids

Two corn hybrids, (Agromen, AGN35-A42, and Bayer, A3663) were used in this trial.

Agromen (AGN35-A42) is a double cross hybrid and has a ultra-short-season life cycle while Bayer (A 3663) is three-way cross hybrid with a short-season life cycle. Both corn hybrids are indicated to production of grains and whole plant silage.

2.3 Experimental Diets

Diets were the same for Exp. 1 and Exp. 2, and were formulated to be isonitrogenous (13% CP, DM basis). Treatments were factorialized in a 2 x 2 arrangement and included main effects of corn silage hybrids (Agromen, AGN35-A42 and Bayer, A3663) and concentrate levels (25 and 50%).

Treatments consisted of 75% of corn silage A + 25% of concentrate (A25), 50 % of corn silage A + 50% of concentrate (A50), 75% of corn silage B + 25% of concentrate (B25), 50 % of corn silage B + 50% (B50), on DM basis (Table 2).

2.4 Ensiling process

Both corn hybrids, Agromen (AGN35-A42) and Bayer (A 3663), were sowed on November 2003 and approximately 52 kg N ha⁻¹; 183 kg P₂O₅ ha⁻¹ and 105 kg K₂O ha⁻¹ were applied as start up fertilizer. Thirty days later, 112 kg N ha⁻¹; 26 kg P₂O₅ ha⁻¹ and 112 kg K₂O ha⁻¹ were applied. Corn hybrids were harvested at 100 days after the sowing, when the milkline was between 1/3 and 2/3 of the grain, and

ensiled without additives in a horizontal-type silo, with approximately 50 tons capacity.

2.5 Animals and Sampling Protocol

Exp. 1. Twenty-four Holstein x Zebu crossbred steers, averaging 335 ± 30 kg BW at the beginning of the study, were distributed in six randomized blocks to evaluate intake and digestibility of nutrients and their performance in the feedlot. Steers were blocked into six groups based on initial BW and allotted randomly to one of four treatments (six steers per treatment). The animals were treated for internal and external parasites at the beginning of the experiment and kept in individual pens of approximately 10 m^2 , with protected feeders and waterier. The Trial was conducted for 99 d (15 d for diet adaptation and 3 periods of 28 d for data collection).

Steers were individually fed ad libitum twice daily at 0700 and 1500. Diets were fed as total mixed ration in which corn silage and concentrate (previously mixed) were weighted and mixed during the feeding. Orts were collected and weighed once daily and diets were adjusted daily to yield orts of about 5 to 10% of offered. Animals had free access to water at all times. Feed ingredients and orts were sampled daily and composited by weight and period.

For each animal, the DMI was measured daily and grab samples of feces (around 200 g) were collected between d 14 and 16 of the second period with collecting intervals of 28 h. Indigestible ADF (iADF) was used as an internal marker to estimate apparent nutrient digestibility and fecal output.

The ADG was calculated as the difference between the final and initial shrunk BW (SBW) (16 h of fasting) divided by the number of days of feeding (84 d). Gain efficiency was calculated as ADG divided by DMI.

Exp. 2. Four Holstein x Zebu crossbred steers, averaging 512 kg \pm 25 kg of BW, and fitted with abomasal and ruminal cannulas were used in a 4 \times 4 Latin Square design to evaluate intake and apparent total tract and partial digestibility of nutrients, ruminal pH and ammonia concentration, ruminal microbial protein synthesis, and urinary excretion of nitrogenous compounds. Each experimental period were 19 d: 10 d for adaptation to the diet, 6 d to collect fecal and abomasal samples, 1 d for ruminal pH measurements and collection of ruminal fluid samples, 1 d to collect spot urine, and 1 d to collect ruminal contents to isolate bacteria. The experiment was conducted for 76 d (4 periods of 19 d). Steers were surgically fitted with ruminal and abomasal cannulae in agreement with techniques described by Leão and Coelho da Silva (1980). Ruminal and abomasal cannulae, and surrounding areas were cleaned routinely during the trial.

Steers were randomly assigned to four dietary treatment sequences and fed individually *ad libitum* twice daily (0700 and 1500). Diets were fed as total mixed ration, being corn silage and concentrate (previously mixed) weighted and mixed at the feeding time. Orts were collected and weighed once daily and the feed offered was adjusted daily to yield ors of about 5 to 10% of total offered. Animals had free access to water at all times. Feed ingredients and ors were sampled daily and composed by weight for each steer within each period.

Feces and abomasal digesta samples (approx. 200g and 500 mL, respectively) were collected between the d 11 and 16 of each period with intervals of 26 h between the samplings. Indigestible ADF (iADF) was used as an internal marker to estimate apparent nutrient digestibility and fecal and abomasal output. After drying at 60°C for 72 h, feed, ors, and fecal and abomasal samples were

ground to pass a 1-mm screen (Willey mill) and period composites per steer were prepared.

Abomasal digesta subsamples (approx. 50 mL) were preserved with 1 mL of 50% (vol/vol) of H₂SO₄, and stored at -20°C for analysis of NH₃-N concentration.

Ruminal contents (100 mL) were obtained at 0, 1, 2, 4, 6 and 8 h after the morning feeding on d 17 of each period and subsequently strained through 2 layers of cheesecloth. The pH was measured immediately. The ruminal fluid was preserved by addition of 1 mL of 50% (vol/vol) of H₂SO₄, and stored at -20°C for analyses of NH₃-N concentration.

Determination of Urinary N excretion

Spot urine samples were collected from all animals at 11:00 h (4 h after feeding) of 18th d of the each experimental period to assess creatinine concentrations to estimate the daily urinary excretion. According to Valadares et al. (1999) creatinine concentration in the spot urine sample is a good indicator of the daily urine production. In addition, creatinine excretion (CE) is little affected by dietary factors such as protein (Kertz et al., 1968), non-fiber carbohydrates (Rennó et al., 2000), or non-protein nitrogen content in the diet (Susmel et al., 1995; Oliveira et al., 2001). Thus, once we can estimate the daily creatinine excretion from animal's body weight (BW), the daily urine volume can be estimated from the creatinine concentration in a urine sample collected during the day (spot sample) and, from the estimated urinary volume, the daily excretion of N and urea could be estimated.

At the end of the collection, one aliquot of urine (50 ml) was obtained for each animal and urine samples were stored at -20°C. Frozen urine samples were thawed

at room temperature and filtered through Whatman #1 filter papers to avoid contamination of steer's hairs.

Determination of Microbial Protein Synthesis

On d 19, the rumen contents were obtained 4 h post-feeding and squeezed through two layers of cheesecloth to yield about 1,500 ml of strained fluid. Particles retained on the cheesecloth were mixed with 500 ml of 9 g of NaCl/L, blended for 1 min, refiltered through cheesecloth, and added to the 1.5 L ruminal fluid sample. Bacteria were isolated by differential centrifugation (500 *g* and 27,000 *g*) according to procedures of Cecava et al. (1990). The resulting bacterial pellets were dried at 60°C for 48 h and ground in a ball mill (TE350, Tecnal, Piracicaba, Brazil). The dried bacterial samples were ground and analyzed for DM, ash, and total N according to procedures described earlier, and total purines were determined as proposed by Ushida et al. (1985).

2.6 Chemical analyses

The composite sample for each material (silage, concentrate, orts, abomasal digesta, and feces) was used to determine the DM (method #934.01; AOAC, 1990); OM determined by ash (method #924.05; AOAC, 1990); CP obtained by total N determination using the micro Kjeldahl technique (method #920.87; AOAC, 1990) and a fixed conversion factor (6.25); EE determined gravimetrically after extraction using petroleum ether in a Soxhlet instrument (method #920.85; AOAC, 1990); NDF (Van Soest et al., 1991); ADF (method #973.18; AOAC, 1990), and sulfuric acid lignin (Van Soest et al., 1991).

The NDF and NFC were not corrected for ash or protein. The iADF (ADF remaining after a 144 h in situ incubation in a rumen-cannulated cow) was determined according to Cochran et al. (1986) and the digestibility of nutrients was calculated as shown in Eq. 1.

$$\text{Nutrient digestibility} = 100 \times (100 - (\% \text{ of iADF of the feed} / \% \text{ of iADF of the feces}) \times (\% \text{ of nutrient of the feces} / \% \text{ of nutrient of the feed})) \quad [1]$$

Non-fiber carbohydrates (NFC) were calculated by difference as shown in Eq. 2

$$\text{NFC} = 100 - [\% \text{CP} + \% \text{NDF} + \% \text{EE} + \% \text{ash}] \quad [2]$$

Apparent TDN was calculated as shown in Eq. 3 (NRC, 2001).

$$\text{App TDN} = (\text{digestible CP} + \text{digestible NDF} + \text{digestible NFC} + (2.25 \times \text{digestible EE})) / \text{DMI}. \quad [3]$$

The determination of the contents of $\text{NH}_3\text{-N}$ of ruminal and abomasal fluid samples were done by the micro-Kjeldahl system with distillation with potassium hydroxide (2N), after previous centrifugation of the sample to 1,000 g, for 15 min, without acid digestion according to the original procedures of Fenner (1965) and adaptations of Vieira (1980).

Creatinine from the urine samples was determined by the colorimetric system with end point reaction, using picrate and acidifier, using commercial kits (555-A Sigma Chemical Co., St. Louis, MO). To estimate the daily urinary volume was used the mean daily creatinine excretion of 27.99 mg/Kg BW obtained by Chizzotti (2004) and the creatinine concentration (mg/L) in the *spot* urine sample, according to the equation:

$$\text{Urine volume (L)} = \frac{\text{BW (kg)} \times \text{creatinine excretion (mg/kg BW)}}{\text{Creatinine concentration (mg/L)}}$$

Urine samples also were analyzed for total N (micro Kjeldahl technique, method #920.87; AOAC, 1990) and for urea with the enzymatic-colorimetric technique using a commercial kit (Uréia CE, Labtest Diagnóstica S.A., Lagoa Santa, Brazil) which was based on Bergmeyer (1985). The estimate urinary volume was used to calculate the daily excretion of total N and urea of each animal.

To quantify microbial protein and subsequently determine microbial efficiency, approximately 400 mg of dry abomasal digesta samples were used, which also were analyzed for purines according to Ushida et al. (1985).

2.7 Statistical Analysis

Exp. 1. All data were analyzed as a randomized complete block design with a 2 x 2 factorial arrangement of treatments using the GLM procedure of SAS (SAS Inst. Inc., Cary, NC). The model included the fixed effects of corn silage hybrid, concentrate levels, the corn silage hybrid x concentrate levels interaction, and random effects of blocks. The statistical model utilized was:

$$Y_{ijkl} = \mu + b_i + s_j + c_k + (sc)_{jk} + e_{ijkl}$$

Where Y_{ijkl} was the dependent variable, μ = overall mean, b_i was the random effect of the i^{th} block, s_j was the fixed effect of the j^{th} silage, c_k was the fixed effect of the k^{th} concentrate level, $(sc)_{jk}$ was the interaction of j^{th} silage with k^{th} concentrate level, and e_{ijkl} was residual error. Treatment differences were considered to be significant when $P \leq 0.05$.

Exp. 2. Data of intake, digestibility, N balance and microbial protein production were analyzed with the GLM procedure of SAS assuming a 4 x 4 Latin square

design with a 2 x 2 factorial arrangement of treatments. The statistical model is described in following equation:

$$Y_{ijklm} = \mu + s_i + c_j + (sc)_{ij} + a_k + p_l + e_{ijklm}$$

Where Y_{ijklm} was the dependent variable, μ = overall mean, s_i was the fixed effect of the i^{th} silage; c_j was the fixed effect of the j^{th} concentrate level, $(sc)_{ij}$ was the interaction of i^{th} silage with j^{th} concentrate level, a_k was the random effect of the k^{th} animal assuming identical, independent, and normal distribution $(0, \sigma_a^2)$; p_l was the random effect of the l^{th} period assuming identical, independent, and normal distribution $(0, \sigma_p^2)$ and e_{ijklm} was the residual error. Differences were considered to be significant when $P \leq 0.05$.

The ruminal characteristics data collected over time were analyzed as repeated measures (Kuehl, 2000) using the MIXED procedure of SAS (SAS Inst. Inc.). Model effects in the whole plot were animal, period, and treatment, whereas subplot effects were sampling time and treatment \times sampling time interactions as shown below.

$$Y_{ijklmno} = \mu + s_i + c_j + (sc)_{ij} + a_k + p_l + e_{ijklm} + z_n + (zsc)_{ijn} + \omega_{ijklmno}$$

Where $Y_{ijklmno}$ was the dependent variable, μ = overall mean, s_i was the fixed effect of the i^{th} silage; c_j was the fixed effect of the j^{th} concentrate level, $(sc)_{ij}$ was the interaction of i^{th} silage with j^{th} concentrate level, a_k was the random effect of the k^{th} animal assuming identical, independent, and normal distribution $(0, \sigma_a^2)$; p_l was the random effect of the l^{th} period assuming identical, independent, and normal distribution $(0, \sigma_p^2)$, e_{ijklm} was whole plot error, z_n was the effect of time; $(zsc)_{ijn}$ was interaction between time and treatment, and $\omega_{ijklmno}$ was the subplot error. When treatment interacted ($P < 0.05$) with sampling time, variables were analyzed within time periods. The variance-covariance structure AR(1) was used for estimating

covariances and the subject of the repeated measurements were defined as animal (period × treatment). Differences were considered to be significant when $P \leq 0.05$.

3. Results

3.1. Silages and Diets

Chemical composition of silages is presented in Table 1. The both corn silage hybrids had similar values of nutrients, except for DM and NFC, which were numerically lower to corn silage hybrid A than those observed to corn silage hybrid B. In addition, the corn silage A had pH value of 3.6 and N-NH₃/Total N of 8.4% while corn silage B had pH of 3.5 and N-NH₃/Total N of 6.3%.

The nutrient composition of the diets is shown in Table 2. As expected, diets with 50% of concentrate provided higher amounts of DM, NFC and TDN than those diets with 25% of concentrate.

3.2. Feed Intake

Table 3 shows the mean intake of nutrients in Exp.1. The intake of DM, OM and CP were not affected by treatments ($P > 0.05$). On the other hand, concentrate levels had a positive effect ($P < 0.05$) on the intakes of NFC and TDN, which were higher to diets with 50% of concentrate than those with 25%. The intake of NDF also was influenced by concentrate levels ($P < 0.0001$) which were lower to steers fed diets with 50% of concentrate. There was an effect ($P < 0.001$) of silage on EE intake.

The intake of nutrients in Exp. 2 is shown in Table 4. There were no treatments effects ($P > 0.05$) on the intakes of DM, OM, CP, and EE. Similar to Exp.

1, the intakes of NDF, NFC and TDN were influenced ($P < 0.05$) by concentrate levels.

3.3. Animal performance

The average daily gain (ADG) and feed efficiency were not affected ($P > 0.05$) by treatments (Table 5).

3.4. Diet Digestibility

The digestibility of nutrients is shown in Table 6 and 7 for Exp. 1 and 2, respectively. Total tract apparent digestibility of DM and OM were affected by concentrate levels ($P < 0.05$), with higher values to diets with 50% of concentrate than those with 25%. The TDN percentage of the diets was also influenced by concentrate levels ($P < 0.05$), which, as expected, was higher to diets with 50% of concentrate than diets with 25%. There was a tendency ($P = 0.08$) of concentrate effect on NDF digestibility.

In the same way, the total tract apparent digestibility of DM and OM, as well as % of TDN were affected by concentrate levels ($P < 0.05$) in Exp. 2 (Table 7). There was a tendency ($P = 0.06$) of concentrate effect on NFC digestibility in Exp.2.

The ruminal tract digestibility and intestinal tract digestibility of nutrients were similar ($P > 0.05$) among all treatments (Table 8). There were no effects of corn silage hybrids on digestibility of all nutrients in both experiments.

3.5 Ruminal Characteristics

There were no treatments effects on ruminal pH values and ruminal ammonia-N concentrations ($P > 0.05$), and the means are shown in Table 9.

3.6. Nitrogen Metabolism

The N metabolism is shown in Table 10. There were no effects ($P > 0.05$) of treatments on N intake, and abomasal flows of total N, $\text{NH}_3\text{-N}$, Non- $\text{NH}_3\text{-N}$ and microbial N, which were similar among all treatments.

In addition, N excretion as fecal N, urinary N and urinary urea-N, and N balance, were also not affected ($P > 0.05$) by treatments.

3.7. Microbial Nitrogen Efficiency

Table 11 shows the data of microbial N efficiency. There was a concentrate effect on rumen-degraded organic matter (RDOM) ($P = 0.02$) and rumen-degraded carbohydrate (RDCHO) ($P = 0.04$). However, the efficiency of microbial synthesis, g of microbial N/kg RDOM, g of microbial N/kg RDCHO and g of microbial crude protein/kg TDN were not affected ($P > 0.05$) by treatments.

4. Discussion

4.1 Silages and Diets

Overall, both silages had similar contents of nutrients, except for % of DM, NDF and NFC (Table 1). Silage A had about 4% less DM, 2% more NDF and 3% less NFC than silage B. These characteristics probably are related with own characteristics of each corn hybrid, since both hybrids got the same fertilization and were harvested at the same occasion. However, these slight numeric differences did not affect the intake of nutrients and performance of animals. According to Muck and Pitt (1993), both silages had good quality due to their adequate percentage of $\text{N-NH}_3/\text{Total}$ and pH values.

The diets with 50% of concentrate in association with either corn silage hybrids provided higher amounts of DM, NFC and TDN and lower NDF than 25% concentrate diets, certainly due to higher percentage of corn ground in those diets.

4.2. Feed Intake

The effect of treatments on intake of nutrients was similar in both experiments, except the EE intake that was affected by silage hybrid in Exp.1.

Although some authors have found linear (Dias et al., 2000; Souza et al. 2002; Pereira et al., 2006a) or quadratic (Verás et al. 2000; Itavo et al., 2002; Silva et al., 2005; Costa et al. 2005) increases in DM intake with increase of concentrate levels in beef cattle diets, no effects of treatments were observed in this study. Moraes et al. (2002) also found no increases in DM intake with the concentrate levels. Probably these variations among studies occurred due to other variables related to kind of animal, age of animal, climatic conditions, days on feeding, etc, that can affect the DM intake.

The higher intake of NFC and TDN and lower intake of NDF in steers fed diets with 50% of concentrate likely occurred as a result of higher amounts of ground corn in these diets than those with 25%. Similar results were found by Costa et al. (2005) who evaluated the effects of levels of concentrate (5, 35, and 65%, DM) on intake of nutrients in Nellore steers.

The higher EE intake observed to diets with corn silage hybrid A in the Exp.1 probably was due to slightly higher amount of EE in this silage (Table 1) although that effect has no been found in Exp.2.

4.3. Animal performance

The lack of effect of treatments on ADG and feed efficiency was not expected. Increases in ADG are expected when animals are fed with high energy diets. In these 2 studies, 50% concentrate diets had more TDN content than those diets with 25%. It's possible that animal breed might have limited the gain. Also, when animals are fed with more energy diets, their maintenance energy requirements increase too (i.e. the internal organs increase their sizes), what can be limited the gain and feed efficiency. Pereira et al. (2006a) found no differences in ADG and feed efficiency when fed crossbred steers with sorghum silage and levels of concentrate (20, 35, 50 and 65%, DM). On the other hand, Costa et al. (2005) and Resende et al. (2001) observed increases in the ADG when concentrate levels were increased in the diets.

4.4. Diet Digestibility

The increases observed in DM and OM digestibilities in diets with 50% of concentrate likely occurred due to higher amount of non-fiber carbohydrates in those diets than in 25% concentrate diets. In general, the apparent digestibility of NFC is higher than digestibility of structural carbohydrates, so the replacement of corn silage by corn ground in 50% concentrate diets provided more NFC to those diets than in 25% concentrate diets. Similar results were observed by Cardoso et al. (2000) and Ítavo et al. (2002).

There was a tendency ($P = 0.08$, Table 6) of concentrate on NDF digestibility in exp. 1. The NDF digestibility was numerically lower to diets with more concentrate, what might have occurred because of a competition between cellulolytic and amylolytic microbes. Microbes that degrade structural carbohydrate (cellulolytic) have low maintenance requirements, grow slowly, and use ammonia as their main N source, whereas microorganisms that degrade nonstructural CHO (amylolytic) have

high maintenance requirements, grow rapidly, and use ammonia, peptides, and amino acids as N sources (Russell et al., 1992). So, diets with more concentrate provide more amylolytic microbes, what can affect negatively the fiber digestion. However, NDF apparent digestibility in Exp.2 was not affected by concentrate levels what can be explained by lack of differences in ruminal pH values. So, cellulolytic microbes were not affected by pH values and, consequently, the fiber digestion was not influenced. Pereira et al. (2006a) and Costa et al. (2005) observed linear decreases in NDF digestibility as concentrate levels in beef cattle diets increased.

In Exp.2, there was a tendency of concentrate levels ($P = 0.06$, Table 7) on NFC digestibility what might be occurred due to higher amount of NFC in those diets and because of higher digestibility of NFC than fiber carbohydrates.

The ruminal and intestinal digestibility of all nutrients were similar among all treatments.

4.5 Ruminal Characteristics

Ruminal pH values were not affected either by silage or by concentrate levels, what means that ruminal environment was adequate for microbial growth. Overall, the mean ruminal pH value observed was 6.02, which is greater than the 5.0 to 5.5 range that was suggested by Hoover (1986) in which ruminal digestibility of fiber is negatively affected. In fact, the apparent ruminal tract digestibility of NDF was not affected by treatments (Table 8).

In the same way, $\text{NH}_3\text{-N}$ concentration was also not affected by treatments and averaged 16.2 mg/100mL. During all time sampling the ruminal $\text{NH}_3\text{-N}$ concentrations were above levels (5 mg/100mL) recommended by Satter and Slyter

(1974) to optimize ruminal digestion. Pereira et al. (2006b) also found no effect of different concentrate levels on ruminal pH and ruminal NH₃-N concentrations.

4.6. Nitrogen Metabolism

No effects of treatments were observed on nitrogen metabolism (Table 10). There were no effects ($P > 0.05$) of treatments on N flows and N excretion. For that reason, the N balance, as g/d; % of N intake and as % of N digested, were not affected by treatments. As the N intake, CP total and ruminal digestibility, and ruminal NH₃-N concentrations were similar among treatments, probably the N excretion and N balance should not be affected too, as observed in this study. This behavior means that the efficiency of N utilization was similar among all diets.

4.7. Microbial Nitrogen Efficiency

Concentrate levels affected the rumen-degraded OM ($P = 0.02$) and rumen-degraded carbohydrate ($P = 0.04$) which were higher to steers fed 50% concentrate diets than those fed 25%. This increase of ruminal degradability observed certainly occurred due to higher amounts of ground corn in those diets with more concentrate, what provided more readily fermentable carbohydrates.

Bacterial growth is largely dependent on the amount of ammonia and fermentable organic matter available in the rumen (Bryant and Robinson, 1962). Readily fermentable carbohydrate, such as starch or sugars, is more effective than other carbohydrate sources, such as cellulose, in promoting microbial growth (Stern and Hoover, 1979). Although rumen-degraded OM and rumen-degraded CHO were high to 50% concentrate diets than those with 25%, microbial N production (Table 10) was not affected by treatments. Moreover, microbial efficiency was also not

influenced by treatments ($P > 0.05$) when expressed on a rumen-degraded OM, rumen-degraded carbohydrate, or on a TDN basis. Maybe, the lack of differences in microbial synthesis and efficiency occurred because of microbial requirements were supplied or microbial growth was slower than the solubilization of N. Either or both might have limited microbial production and (or) efficiency.

The mean microbial efficiency of 105.95 g microbial CP/ kg TDN is lower of that recommended by NRC (1996), which is 130 g microbial CP/ kg TDN. Although the analysis of variance did not show any diet effect, there was a numeric decrease in this variable (Table 11) with the increase of the concentrate levels, probably due to the larger TDN intake (Table 4) recorded for diets with higher concentrate levels.

5. Conclusions

The utilization of either corn silage hybrids evaluated in association with 25% of concentrate is a good option to feed crossbreeds steers (Holstein x Zebu) with ADG close to 1.0 kg, what might result in the reduction of diet cost.

The association of either corn silage hybrids evaluated with 25 or 50% of concentrate not affects the ruminal pH and ruminal ammonia-N concentration, N balance, and microbial efficiency.

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Table 1. Chemical composition of corn silages

Item	Corn Silages	
	A	B
DM, %	28.01	32.45
OM, % of DM	96.12	96.02
CP, % of DM	7.00	6.02
NDF, % of DM	53.00	51.14
iADF, % of DM	12.50	11.80
EE, % of DM	2.25	2.02
NFC, % of DM	33.87	36.86
Lignin, % of DM	3.62	3.45
pH	3.60	3.50
NH ₃ -N, % of total N	8.40	6.30

OM = organic matter; ADF = acid detergent fiber; iADF = indigestible ADF;

EE = ether extract; and NFC = non-fiber carbohydrates.

Table 2. Ration composition and analyzed content of diets fed in exp. 1 and 2

Item	Corn silage A		Corn silage B	
	25	50	25	50
Corn silage	75.0	50.0	75.0	50.0
Ground corn	14.86	41.0	14.86	41.0
Cottonseed meal	8.27	7.03	8.27	7.03
Urea	1.00	1.00	1.00	1.00
Ammonium sulfate	0.10	0.10	0.10	0.10
Sodium chloride	0.25	0.25	0.25	0.25
Calcite limestone	0.50	0.60	0.50	0.60
Mineral premix ¹	0.02	0.02	0.02	0.02
	Nutrient content of diets ²			
DM, %	43.53	59.38	46.91	61.60
OM, % of DM	94.28	94.44	94.20	94.39
CP, % of DM	13.06	13.13	12.32	12.64
NDF, % of DM	43.74	32.73	42.35	31.80
iADF, % of DM	10.58	7.45	10.06	7.10
EE, % of DM	2.14	2.15	1.95	2.02
NFC, % of DM	38.81	50.46	41.04	51.95
Lignin, % of DM	3.16	2.46	3.03	2.37

¹Composition (%): cooper sulfate (22.50), cobalt sulfate (1.40), zinc sulfate (75.40), potassium iodate (0.50), sodium selenite (0.20).

²OM = organic matter; ADF = acid detergent fiber; iADF = indigestible ADF; EE = ether extract; and NFC = non-fiber carbohydrates.

Table 3. Effect of corn silage hybrids and concentrate levels on nutrient intake in Exp. 1^a

Item	Corn silage A		Corn silage B		SEM	Silage	<i>P</i> -value ^b	
	25	50	25	50			Concentrate	S x C
	Intake, kg/d							
DM	8.84	9.00	8.57	9.01	0.29	0.67	0.33	0.65
OM	8.50	8.70	8.26	8.72	0.28	0.69	0.26	0.66
CP	1.18	1.15	1.11	1.10	0.04	0.16	0.61	0.83
EE	0.20	0.20	0.18	0.19	0.006	0.02	0.15	0.42
NDF	3.56	2.60	3.58	2.66	0.12	0.81	<0.0001	0.83
NFC	3.60	4.83	3.61	4.94	0.14	0.64	<0.0001	0.70
TDN	5.78	6.32	5.60	6.14	0.17	0.32	0.006	0.99
	Intake, % of BW							
DM	2.32	2.37	2.30	2.37	0.06	0.81	0.34	0.90
NDF	0.93	0.69	0.96	0.70	0.03	0.52	<0.0001	0.90

^aOM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

Table 4. Effect of corn silage hybrids and concentrate levels on of nutrient intake in Exp. 2^a

Item	Corn silage A		Corn silage B		SEM	Silage	<i>P</i> -value ^b	
	25	50	25	50			Concentrate	S x C
	Intake, kg/d							
DM	10.97	11.12	10.22	11.79	0.62	0.94	0.21	0.30
OM	10.49	10.71	9.78	11.44	0.59	0.99	0.16	0.27
CP	1.48	1.47	1.32	1.53	0.08	0.55	0.24	0.17
EE	0.24	0.22	0.21	0.23	0.02	0.64	0.83	0.33
NDF	4.61	3.23	4.17	3.29	0.35	0.61	0.02	0.49
NFC	4.46	6.07	4.17	6.44	0.23	0.87	<0.001	0.21
TDN	7.38	8.13	6.62	8.45	0.47	0.66	0.03	0.29
	Intake, % of BW							
DM	1.95	1.98	1.80	2.08	0.11	0.81	0.20	0.30
NDF	0.82	0.58	0.74	0.58	0.06	0.54	0.02	0.51

^aOM = organic matter, EE = ether extract, and NFC = non-fiber carbohydrates.

^bS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

Table 5. Effect of corn silage hybrids and concentrate levels on ADG, and feed efficiency in the Exp. 1

Item	Corn silage A		Corn silage B		SEM	<i>P</i> -value ^a		
	25	50	25	50		Silage	Concentrate	S x C
ADG, kg/d	1.07	1.13	1.06	1.14	0.07	0.96	0.39	0.92
Feed efficiency	8.31	8.09	8.19	7.92	0.42	0.73	0.57	0.95

^aS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

Table 6. Effect of corn silage hybrids and concentrate levels on apparent total tract digestibility of nutrients and % of TDN of the diets in Exp. 1^a

Item	Corn silage A		Corn silage B		SEM	Silage	<i>P</i> -value ^b	
	25	50	25	50			Concentrate	S x C
DM	65.26	69.43	63.25	66.42	1.38	0.09	0.02	0.72
OM	66.06	70.30	64.57	68.15	1.32	0.19	0.01	0.80
CP	65.22	64.93	63.04	65.73	0.87	0.43	0.18	0.11
EE	82.15	84.37	81.48	84.46	1.59	0.86	0.12	0.81
NDF	48.31	46.44	49.08	41.24	2.61	0.41	0.08	0.27
NFC	81.58	83.17	78.70	79.96	1.66	0.09	0.40	0.92
TDN	65.47	70.65	65.46	68.19	1.40	0.39	0.01	0.40

^aValues are expressed as %;

^bS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

Table 7. Effect of corn silage hybrids and concentrate levels on apparent total tract digestibility of nutrients and % of TDN of the diets in the Exp. 2^a

Item	Corn silage A		Corn silage B		SEM	Silage	<i>P</i> -value ^b	
	25	50	25	50			Concentrate	S x C
DM	66.22	71.41	64.05	71.55	2.60	0.71	0.05	0.67
OM	66.80	72.11	64.68	72.44	2.57	0.74	0.04	0.65
CP	66.77	66.88	65.87	66.81	2.51	0.85	0.84	0.87
EE	78.94	76.85	78.89	79.47	2.42	0.61	0.77	0.60
NDF	53.96	49.35	53.52	54.10	3.21	0.53	0.55	0.45
NFC	79.36	85.30	75.77	81.61	2.52	0.20	0.06	0.98
TDN	67.91	73.22	64.97	71.64	2.44	0.39	0.05	0.79

^aValues are expressed as %;

^bS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

Table 8. Effect of corn silage hybrids and concentrate levels on apparent ruminal and intestinal tract digestibility of nutrients in Exp. 2

Item	Corn silage A		Corn silage B		SEM	Silage	<i>P</i> -value ^a	
	25	50	25	50			Concentrate	S x C
	Ruminal tract digestibility							
DM ^b	83.2	82.1	84.1	82.4	1.88	0.74	0.48	0.88
OM ^b	84.3	82.7	84.9	83.2	1.73	0.73	0.39	0.98
CP ^b	55.60	58.06	53.35	60.92	3.36	0.93	0.17	0.48
EE ^b	-18.95	-28.38	-13.47	-21.67	19.82	0.77	0.67	0.98
NDF ^b	91.3	95.4	96.9	95.8	1.75	0.13	0.41	0.19
NFC ^b	89.9	87.6	86.8	86.9	2.53	0.50	0.67	0.67
	Intestinal tract digestibility							
DM ^b	16.8	17.9	15.9	17.6	1.88	0.74	0.48	0.88
OM ^b	15.7	17.3	15.1	16.8	1.73	0.73	0.39	0.98
CP ^b	44.40	41.94	46.65	39.08	3.36	0.93	0.17	0.48
EE ^b	118.95	128.38	113.47	121.67	19.82	0.77	0.67	0.98
NDF ^b	8.72	4.59	3.07	4.13	1.75	0.13	0.41	0.19
NFC ^b	10.1	12.4	13.2	13.1	2.53	0.50	0.67	0.67

^aS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

^bCalculated as % of digested.

Table 9. Means values of ruminal pH and ammonia-N concentration after feeding time (hours)

Time (hours)	Corn silage A		Corn silage B		Means
	25	50	25	50	
	pH				
0	6.38	6.18	6.20	6.04	6.20
2	6.29	6.09	6.02	5.85	6.05
4	5.88	5.90	6.02	6.01	5.95
6	5.92	5.92	5.90	5.78	5.88
8	6.23	5.97	6.01	5.95	6.03
	NH ₃ -N (mg/100mL)				
0	9.18	11.55	9.18	10.50	10.10
2	24.49	20.12	22.48	25.80	23.22
4	21.52	22.74	17.64	19.46	20.34
6	18.67	17.76	14.43	12.60	15.86
8	13.12	11.35	10.13	10.74	11.34

Table 10. Effects of corn silage hybrids and concentrate levels on balance of nitrogen in the Exp. 2

Item	Corn silage A		Corn silage B		SEM	<i>P</i> -value ^a		
	25	50	25	50		Silage	Concentrate	S x C
N intake, g/d	237.4	234.4	211.0	245.4	12.0	0.55	0.24	0.17
<i>Abomasal flow, g/d</i>								
Total N	149.6	143.4	137.2	145.4	11.38	0.66	0.93	0.55
NH ₃ -N	1.00	0.97	0.96	1.15	0.20	0.75	0.71	0.60
Non-NH ₃ -N ^b	148.6	142.4	136.2	144.3	11.41	0.66	0.94	0.56
Microbial N	129.7	126.9	127.1	129.3	12.40	0.99	0.98	0.85
N excretion								
Fecal N, g/d	79.06	76.34	70.40	81.07	5.69	0.74	0.51	0.28
Urinary N, g/d	95.37	97.61	82.38	98.53	4.70	0.25	0.10	0.19
Urinary Urea-N, g/d	48.37	49.97	40.90	50.16	8.27	0.67	0.54	0.66
N balance								
N balance, g/d	62.91	60.46	58.23	65.84	9.79	0.97	0.80	0.63
N balance, % of N intake	26.29	25.59	27.71	26.79	3.57	0.73	0.83	0.97
N balance, % of N digested	38.76	37.67	41.30	39.98	4.50	0.61	0.80	0.98

^aS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

^bNon-NH₃-N = Total N abomasal flow - NH₃-N abomasal flow

Table 11. Effect of corn silage hybrids and concentrate levels on microbial N efficiency in the Exp. 2

Item	Corn silage A		Corn silage B		SEM	<i>P</i> -value ^d		
	25	50	25	50		Silage	Concentrate	S x C
Rumen degradability, kg/d								
OM	5.85	6.39	5.36	6.90	0.33	0.98	0.02	0.18
CHO ^a	5.39	5.93	4.96	6.26	0.36	0.90	0.04	0.33
Microbial N efficiency								
g mic N/ kg RDOM ^b	22.29	20.09	23.71	18.80	1.86	0.97	0.11	0.50
g mic N/kg of RDCHO ^c	24.28	21.70	25.59	20.74	2.11	0.94	0.13	0.61
g mic CP / kg of TDN	110.3	98.2	120.2	95.1	10.04	0.75	0.11	0.54

^aCHO = 100 - (%CP + %EE + %Ash), Sniffen et al. (1992);

^bRDOM = ruminally degraded organic matter; RDOM (kg/d) = Intake of OM – abomasal OM flow

^cRDCHO = ruminally degraded carbohydrates; RDCHO (kg/d) = Intake of CHO – abomasal CHO flow

^dS = Silage source effect; C= Concentrate level effect; S × C = Silage and concentrate interaction effect.

CONCLUSÕES GERAIS

Níveis de NNP até 46,5% do N dietético podem ser utilizados para novilhos cruzados (H x Z) com ganhos de peso próximos de 1,0 kg/d, recebendo dietas a base de silagem de milho, sem afetar o desempenho bem como a eficiência de síntese microbiana, embora a eficiência de utilização do nitrogênio possa diminuir significativamente.

A recomendação do nível de NNP (uréia) dietética irá depender do critério utilizado para definir uma ótima eficiência de uso do nitrogênio, uma vez que níveis de uréia requeridos para reduzir custos da dieta, e/ou maximizar performance e eficiência de síntese microbiana, podem não ser os mesmos níveis necessários para minimizar a poluição ambiental.

O uso de silagem de milho de ambos híbridos avaliados associado a 25% de concentrado é uma boa opção para alimentação de novilhos cruzados (H x Z) com potencial de ganho diário de peso em torno de 1,0 kg, propiciando desta forma, menor gasto com ração concentrada.

A associação de silagem de milho de ambos híbridos avaliados com 25 ou 50% de concentrado não altera o pH e N-amônia ruminais, o balanço de nitrogênio bem como a eficiência de síntese microbiana.

Apêndice

Tabela 1. Blocos (BL), tratamento (TMT), ganho médio diário (GMD), consumo de matéria seca (MS), matéria orgânica (MO), proteína bruta (PB), fibra em detergente neutro (FDN), extrato etéreo (EE), carboidratos não-fibrosos (CNF) e nutrientes digestíveis totais (NDT), expressos em kg/dia e consumo de matéria seca (CMS) e fibra em detergente neutro (CFDN) expressos em % do peso vivo, (experimento 1).

BL	TMT	GMD	CMS	CMO	CPB	CFDN	CEE	CCNF	CNDT	CMS	CFDN
1	0	1,30	9,7	9,21	1,13	3,99	0,25	3,88	6,76	2,4	0,97
1	15,5	0,77	8,5	8,05	0,98	3,47	0,23	3,40	6,05	2,2	0,92
1	31	1,13	9,7	9,26	1,18	3,64	0,26	4,23	7,08	2,4	0,89
1	46,5	1,30	8,8	8,42	1,06	3,22	0,22	3,96	6,49	2,2	0,79
2	0	0,95	9,0	8,46	1,13	3,46	0,23	3,68	6,11	2,1	0,83
2	15,5	1,50	11,2	10,65	1,39	4,37	0,30	4,62	7,72	2,5	0,96
2	31	0,81	8,1	7,69	1,02	2,87	0,22	3,61	5,50	1,9	0,67
2	46,5	1,36	9,4	8,94	1,08	3,67	0,23	4,02	6,46	2,1	0,84
3	0	1,31	11,1	10,51	1,40	4,43	0,27	4,45	7,60	2,3	0,91
3	15,5	1,15	9,9	9,44	1,20	3,77	0,26	4,25	7,40	2,1	0,81
3	31	1,14	9,7	9,28	1,21	3,53	0,26	4,33	6,76	2,2	0,79
3	46,5	1,17	8,9	8,44	1,04	3,17	0,22	4,07	5,46	2,1	0,73
4	0	1,45	12,0	10,58	1,44	4,40	0,28	4,52	8,39	2,4	0,88
4	15,5	1,14	9,5	9,04	1,20	3,65	0,26	3,97	6,62	2,0	0,77
4	31	1,50	10,9	10,40	1,34	4,01	0,29	4,80	8,27	2,3	0,84
4	46,5	0,76	8,9	8,45	1,06	3,23	0,22	3,99	6,56	2,0	0,71
5	0	1,02	9,5	9,00	1,17	3,79	0,24	3,84	6,71	2,1	0,82
5	15,5	1,05	9,5	9,05	1,20	3,63	0,26	4,00	6,64	2,1	0,81
5	31	1,25	11,5	10,94	1,41	4,23	0,30	5,04	8,80	2,5	0,92
5	46,5	1,38	11,0	10,46	1,34	4,01	0,27	4,90	7,85	2,4	0,87
6	0	0,81	10,8	10,21	1,34	4,26	0,27	4,37	7,53	2,3	0,91
6	15,5	1,19	10,2	9,68	1,28	3,86	0,28	4,30	7,01	2,1	0,80
6	31	0,77	9,2	8,74	1,12	3,41	0,25	4,01	6,82	1,9	0,72
6	46,5	1,13	9,2	8,72	1,07	3,33	0,22	4,15	6,97	2,0	0,72

Tabela 2. Blocos (BL), tratamento (TMT), digestibilidade total da matéria seca (MS), matéria orgânica (MO), proteína bruta (PB), fibra em detergente neutro (FDN), extrato etéreo (EE), carboidratos não-fibrosos (CNF) e teor de nutrientes digestíveis totais (NDT). (experimento 1).

BL	TMT	DMS	DMO	DPB	DFDN	DEE	DCNF	NDT
1	0	68,73	70,08	63,57	59,60	82,69	82,16	69,40
1	15,5	70,47	71,50	65,24	49,69	88,30	90,94	71,43
1	31	71,68	72,78	72,71	53,70	90,74	87,68	73,01
1	46,5	72,92	73,51	73,63	55,50	91,84	86,24	73,43
2	0	67,14	68,70	64,29	48,64	89,16	87,41	68,24
2	15,5	67,77	68,87	58,66	49,68	90,13	88,65	68,79
2	31	66,64	67,88	70,27	48,73	88,20	81,85	68,28
2	46,5	68,41	69,26	67,60	52,09	81,07	85,39	68,77
3	0	67,97	69,23	65,54	52,17	79,92	86,82	68,36
3	15,5	73,92	75,01	69,08	59,70	80,49	90,85	74,48
3	31	68,57	69,74	64,22	53,47	73,74	84,85	69,50
3	46,5	60,63	62,05	62,13	45,27	69,85	77,09	61,57
4	0	69,98	70,96	66,10	54,31	77,65	88,44	69,89
4	15,5	68,08	69,60	65,49	49,65	84,58	89,71	69,44
4	31	75,38	76,39	72,06	57,94	83,02	92,54	75,97
4	46,5	74,00	74,69	68,78	59,63	74,83	88,09	73,96
5	0	70,13	71,16	68,86	54,17	85,51	87,57	70,48
5	15,5	68,58	69,54	66,04	52,05	89,62	85,30	69,59
5	31	75,01	76,54	73,37	61,92	87,23	88,73	76,77
5	46,5	70,22	71,82	68,00	59,35	86,26	82,67	71,49
6	0	69,62	72,10	65,47	52,59	82,78	89,17	69,73
6	15,5	67,40	68,84	65,78	52,98	88,23	82,97	68,69
6	31	73,06	74,05	73,32	53,56	87,81	88,71	74,48
6	46,5	74,99	76,40	76,33	59,77	88,68	89,30	76,10

Valores expressos em %

Tabela 3. Período (PER), animal (AN), tratamento (TMT), consumo de matéria seca (MS), matéria orgânica (MO), proteína bruta (PB), fibra em detergente neutro (FDN), extrato etéreo (EE), carboidratos não-fibrosos (CNF) e nutrientes digestíveis totais (NDT), expressos em kg/dia e consumo de matéria seca (CMSPV) e fibra em detergente neutro (CFDNPV) expressos em % do peso vivo, (experimento 2).

PER	AN	TMT	CMS	CMO	CPB	CFDN	CEE	CCNF	CNDT	CMS	CFDN
1	21	0	8.96	8.47	1.17	3.54	0.14	3.63	6.48	2.19	0.86
2	8	0	8.88	8.38	1.26	3.12	0.21	3.81	5.86	1.92	0.67
3	18	0	10.64	10.07	1.36	4.11	0.21	4.39	7.37	2.47	0.95
4	5	0	8.34	7.88	1.06	3.29	0.16	3.38	5.43	2.23	0.88
1	5	15.5	7.01	6.65	0.92	2.59	0.12	3.01	4.87	2.28	0.84
2	21	15.5	11.15	10.58	1.48	4.22	0.26	4.63	7.69	2.47	0.93
3	8	15.5	10.62	10.07	1.36	4.05	0.25	4.42	7.18	2.19	0.83
4	18	15.5	10.80	10.27	1.35	4.26	0.24	4.43	7.25	2.34	0.92
1	18	31	9.32	8.91	1.22	3.35	0.16	4.19	6.26	2.61	0.94
2	5	31	7.51	7.18	1.00	2.69	0.16	3.34	5.44	2.29	0.82
3	21	31	11.77	11.23	1.51	4.23	0.28	5.13	7.52	2.38	0.86
4	8	31	11.64	11.16	1.43	4.43	0.24	5.05	7.61	2.26	0.86
1	8	46.5	9.73	9.28	1.25	3.41	0.16	4.46	6.92	2.16	0.76
2	18	46.5	9.11	8.68	1.20	3.21	0.20	4.09	6.87	2.31	0.82
3	5	46.5	8.11	7.73	1.02	2.90	0.17	3.64	5.18	2.32	0.83
4	21	46.5	11.41	10.91	1.37	4.36	0.20	4.98	7.90	2.18	0.83

Tabela 4. Período (PER), animal (AN), tratamento (TMT), digestibilidade total da matéria seca (DMS), matéria orgânica (DMO), proteína bruta (DPB), fibra em detergente neutro (DFDN), extrato etéreo (DEE), carboidratos não-fibrosos (DCNF) e teor de nutrientes digestíveis totais (NDT). (experimento 2).

PER	AN	TMT	DMS	DMO	DPB	DFDN	DEE	DCNF	NDT
1	21	0	75.31	75.02	73.57	63.10	70.08	87.30	72.32
2	8	0	66.03	67.17	64.44	43.23	85.73	86.77	66.08
3	18	0	69.31	70.75	64.50	53.34	90.01	88.07	69.21
4	5	0	65.43	67.13	55.09	52.54	68.61	85.01	65.17
1	5	15.5	70.46	71.35	63.70	58.46	80.73	84.41	69.40
2	21	15.5	67.67	70.28	71.51	54.34	71.39	84.47	68.98
3	8	15.5	67.30	68.70	62.94	53.45	85.73	83.49	67.63
4	18	15.5	65.45	67.95	65.35	53.06	92.64	81.74	67.17
1	18	31	67.14	68.82	68.21	52.68	64.56	82.02	67.11
2	5	31	71.40	73.16	70.69	58.35	84.44	85.34	72.40
3	21	31	64.90	65.32	64.91	50.70	79.28	76.14	63.91
4	8	31	64.99	66.28	64.36	52.94	72.02	78.22	65.41
1	8	46.5	71.35	72.74	70.31	55.66	75.11	86.51	71.11
2	18	46.5	74.74	76.42	76.75	62.61	88.76	86.65	75.46
3	5	46.5	63.31	64.98	59.97	50.18	75.99	77.64	63.85
4	21	46.5	69.23	70.84	69.48	56.50	67.39	83.90	69.23

Valores expressos em %.

Tabela 5. Período (PER), animal (AN), tratamento (TMT), digestibilidade ruminal da matéria seca (DRMS), matéria orgânica (DRMO), proteína bruta (DRPB), fibra em detergente neutro (DRFDN), extrato etéreo (DREE) e carboidratos não-fibrosos (DRCNF) (experimento 2).

PER	AN	TMT	DRMS	DRMO	DRPB	DRFDN	DREE	DRCNF
1	21	0	79.99	84.28	42.28	86.70	-43.97	97.92
2	8	0	80.07	84.13	38.81	93.65	30.29	94.30
3	18	0	62.69	68.23	21.41	94.37	1.82	67.41
4	5	0	76.20	78.13	38.88	81.44	-10.34	87.54
1	5	15.5	76.57	78.68	59.09	95.13	-39.81	77.91
2	21	15.5	75.67	77.35	39.56	90.37	23.67	82.64
3	8	15.5	80.39	82.11	48.18	96.40	24.01	84.93
4	18	15.5	78.87	83.32	43.03	92.18	9.13	92.08
1	18	31	80.75	82.54	69.85	94.73	-59.22	83.49
2	5	31	81.35	81.54	61.57	91.30	13.74	84.47
3	21	31	71.39	76.21	35.41	84.35	8.84	85.23
4	8	31	68.25	72.93	37.78	91.19	6.98	73.22
1	8	46.5	66.04	71.10	54.79	77.20	-39.28	75.45
2	18	46.5	71.46	72.08	62.04	92.11	16.98	66.21
3	5	46.5	78.78	80.75	58.02	96.36	9.07	80.84
4	21	46.5	73.40	75.17	58.03	94.59	-19.27	70.72

Valores expressos em %.

Tabela 6. Período (PER), animal (AN), tratamento (TMT), digestibilidade intestinal da matéria seca (DIMS), matéria orgânica (DIMO), proteína bruta (DIPB), fibra em detergente neutro (DIFDN), extrato etéreo (DIEE) e carboidratos não-fibrosos (DICNF) (experimento 2).

PER	AN	TMT	DIMS	DIMO	DIPB	DIFDN	DIEE	DICNF
1	21	0	20.01	15.72	57.72	13.30	143.97	2.08
2	8	0	19.93	15.87	61.19	6.35	69.71	5.70
3	18	0	37.31	31.77	78.59	5.63	98.18	32.59
4	5	0	23.80	21.87	61.12	18.56	110.34	12.46
1	5	15.5	23.43	21.32	40.91	4.87	139.81	22.09
2	21	15.5	24.33	22.65	60.44	9.63	76.33	17.36
3	8	15.5	19.61	17.89	51.82	3.60	75.99	15.07
4	18	15.5	21.13	16.68	56.97	7.82	90.87	7.92
1	18	31	19.25	17.46	30.15	5.27	159.22	16.51
2	5	31	18.65	18.46	38.43	8.70	86.26	15.53
3	21	31	28.61	23.79	64.59	15.65	91.16	14.77
4	8	31	31.75	27.07	62.22	8.81	93.02	26.78
1	8	46.5	33.96	28.90	45.21	22.80	139.28	24.55
2	18	46.5	28.54	27.92	37.96	7.89	83.02	33.79
3	5	46.5	21.22	19.25	41.98	3.64	90.93	19.16
4	21	46.5	26.60	24.83	41.97	5.41	119.27	29.28

Valores expressos em %.

Tabela 7. Período (PER), animal (AN), tratamento (TMT), nitrogênio microbiano (N mic), carboidratos degradados no rúmen (CHODR) e matéria orgânica degradada no rúmen (MODR) expressos em g/d, e eficiência de síntese microbiana expressa em g PB microbiana/ kg NDT (EFPBNDT), g N mic /kg CHODR, e g N mic/ kg MODR, (experimento 2)

PER	AN	TMT	Nmic	EFPBNDT	CHODR	MODR	NmicCHODR	NmicMODR
1	21	0	104.34	10.07	5.07	5.36	20.56	20.07
2	8	0	116.89	12.46	4.32	4.73	27.44	24.70
3	18	0	154.26	13.66	4.44	4.86	34.77	31.41
4	5	0	100.28	11.79	3.94	4.13	25.44	23.65
1	5	15.5	74.73	9.81	3.47	3.73	21.51	20.02
2	21	15.5	140.96	13.49	5.25	5.75	26.86	24.50
3	8	15.5	131.69	11.46	5.28	5.68	24.93	23.18
4	18	15.5	123.58	11.83	5.18	5.81	23.87	21.25
1	18	31	87.89	8.53	4.53	5.06	19.40	17.36
2	5	31	81.63	9.38	3.91	4.29	21.01	19.05
3	21	31	151.71	12.60	5.09	5.59	29.80	27.13
4	8	31	148.63	12.47	4.81	5.39	30.93	27.56
1	8	46.5	103.95	9.80	4.17	4.80	24.92	19.06
2	18	46.5	98.25	9.05	4.26	4.78	23.06	20.54
3	5	46.5	100.10	12.43	3.68	4.06	27.20	24.67
4	21	46.5	124.46	9.85	5.27	5.81	23.59	22.80

Tabela 8. Blocos (BL), silagem (SIL), concentrado (CONC), ganho médio diário (GMD), consumo de matéria seca (MS), matéria orgânica (MO), proteína bruta (PB), fibra em detergente neutro (FDN), extrato etéreo (EE), carboidratos não-fibrosos (CNF), expressos em kg/dia e consumo de matéria seca e fibra em detergente neutro expressos em % do peso vivo, (experimento 3).

BL	SIL	CONC	ADG	CMS	CMS	CMO	CPB	CFDN	CFDN	CEE	CCNF
1	A	25	1.19	9.39	2.20	9.15	1.26	3.73	0.88	0.19	3.89
2	A	25	1.06	9.45	2.33	9.11	1.26	3.84	0.95	0.19	3.84
3	A	25	1.21	9.49	2.46	9.09	1.25	3.89	1.01	0.20	3.84
4	A	25	1.12	8.06	2.11	7.77	1.10	3.23	0.85	0.16	3.31
5	A	25	0.96	8.77	2.49	8.43	1.19	3.56	1.01	0.18	3.54
6	A	25	0.88	7.86	2.35	7.44	1.05	3.08	0.92	0.16	3.17
1	A	50	1.40	11.37	2.54	10.99	1.51	3.39	0.76	0.20	5.94
2	A	50	1.45	9.60	2.35	9.27	1.19	2.80	0.69	0.16	5.16
3	A	50	0.98	8.53	2.29	8.25	1.11	2.39	0.64	0.15	4.64
4	A	50	0.86	7.71	2.19	7.47	1.00	2.13	0.60	0.14	4.23
5	A	50	1.08	8.76	2.53	8.47	1.12	2.54	0.73	0.14	4.71
6	A	50	1.00	8.02	2.34	7.77	0.99	2.36	0.69	0.14	4.28
1	B	25	1.00	8.37	2.07	8.06	1.10	3.42	0.84	0.15	3.57
2	B	25	0.79	8.29	2.15	7.95	1.08	3.45	0.90	0.15	3.48
3	B	25	1.08	9.23	2.37	8.91	1.19	3.82	0.98	0.17	3.91
4	B	25	1.27	9.24	2.47	8.91	1.18	3.90	1.04	0.15	3.86
5	B	25	1.17	8.42	2.33	8.12	1.09	3.48	0.96	0.15	3.53
6	B	25	1.10	7.89	2.42	7.61	1.03	3.28	1.01	0.14	3.30
1	B	50	1.11	9.97	2.19	9.63	1.26	2.89	0.63	0.14	5.51
2	B	50	1.18	10.03	2.51	9.70	1.30	2.96	0.74	0.16	5.45
3	B	50	1.23	9.53	2.51	9.23	1.14	2.74	0.72	0.15	5.35
4	B	50	1.07	8.01	2.23	7.76	0.92	2.22	0.62	0.12	4.51
5	B	50	1.07	8.60	2.49	8.33	1.04	2.76	0.80	0.13	4.53
6	B	50	1.18	7.91	2.28	7.66	0.96	2.39	0.69	0.12	4.30

Tabela 9. Blocos (BL), silagem (SIL), concentrado (CONC), digestibilidade total da matéria seca (DMS), matéria orgânica (DMO), proteína bruta (DPB), fibra em detergente neutro (DFDN), extrato etéreo (DEE), carboidratos não-fibrosos (DCNF) e teor de nutrientes digestíveis totais (NDT). (experimento 3).

BL	SIL	CONC	DMS	DMO	DCP	DNDF	DEE	DCNF	NDT
1	A	25	61.89	63.24	63.62	40.55	78.70	81.62	62.11
2	A	25	61.31	62.26	63.84	40.65	78.11	80.91	61.49
3	A	25	65.67	66.33	64.76	51.78	76.63	80.07	65.76
4	A	25	64.52	65.41	67.78	48.01	79.07	79.41	64.60
5	A	25	68.21	68.90	64.21	53.67	86.31	83.73	68.35
6	A	25	69.99	70.23	67.11	55.19	85.27	83.72	68.24
1	A	50	62.29	63.32	63.08	39.31	69.10	75.81	62.51
2	A	50	71.35	72.11	64.33	54.89	79.97	82.22	71.26
3	A	50	72.23	73.02	67.97	52.50	84.15	83.67	72.46
4	A	50	73.34	74.13	68.13	56.62	88.69	83.07	73.59
5	A	50	67.58	68.51	58.27	42.93	82.95	83.28	67.64
6	A	50	69.81	70.71	67.82	53.92	74.85	79.48	69.74
1	B	25	66.92	68.07	59.11	53.51	76.19	83.78	68.56
2	B	25	63.56	64.65	66.59	49.17	81.32	78.21	65.26
3	B	25	60.78	62.19	65.52	51.18	75.21	70.36	62.50
4	B	25	59.97	61.37	64.78	47.83	74.71	72.40	61.43
5	B	25	61.53	62.89	59.60	42.08	77.68	82.49	62.71
6	B	25	66.74	68.27	62.63	50.69	85.19	85.00	68.32
1	B	50	64.25	66.32	66.98	55.62	77.79	77.35	69.84
2	B	50	70.49	71.94	67.12	52.92	79.18	81.84	71.60
3	B	50	66.44	68.11	64.68	48.67	79.01	80.27	69.62
4	B	50	63.89	65.70	66.36	31.02	77.46	79.50	63.66
5	B	50	63.64	65.47	62.84	67.20	77.41	75.18	71.50
6	B	50	69.82	71.33	66.41	46.77	80.66	84.31	70.87

Valores expressos em %.

Tabela 10. Animal (AN), período (Per), silagem (SIL), concentrado (Conc), consumo de matéria seca (CMS), matéria orgânica (CMO), proteína bruta (CPB), fibra em detergente neutro (CFDN), extrato etéreo (CEE), carboidratos não-fibrosos (CCNF) e consumo de nutrientes digestíveis totais (CNDT), expressos em kg/dia, e consumo de matéria seca expresso em % do peso vivo, (experimento 4).

ANI	Per	SIL	Conc	CMS	CMS	CMO	CPB	CFDN	CEE	CCNF	CNDT
47	1	A	25	11.56	2.16	11.05	1.55	4.88	0.25	4.69	8.33
34	2	A	25	12.47	2.15	11.92	1.66	5.32	0.28	5.01	7.16
28	3	A	25	10.49	1.88	10.03	1.42	4.44	0.22	4.24	6.88
25	4	A	25	9.35	1.60	8.95	1.30	3.81	0.21	3.90	7.16
25	1	A	50	11.52	2.30	11.09	1.54	3.49	0.23	6.10	8.15
47	2	A	50	9.83	1.73	9.47	1.29	2.86	0.20	5.36	7.08
34	3	A	50	13.67	2.26	13.17	1.78	4.05	0.27	7.39	10.01
28	4	A	50	9.46	1.63	9.13	1.25	2.51	0.18	5.44	7.27
28	1	B	25	9.52	1.84	9.10	1.25	3.89	0.20	3.86	6.12
25	2	B	25	9.84	1.85	9.43	1.27	4.02	0.22	4.00	7.04
47	3	B	25	11.47	1.91	10.97	1.49	4.58	0.24	4.78	7.06
34	4	B	25	10.05	1.60	9.60	1.27	4.18	0.20	4.05	6.28
34	1	B	50	11.09	2.00	10.81	1.55	2.25	0.18	6.86	7.80
28	2	B	50	11.51	2.15	11.16	1.45	3.58	0.21	5.97	8.00
25	3	B	50	11.95	2.15	11.59	1.48	3.69	0.27	6.20	8.89
47	4	B	50	12.59	2.00	12.19	1.65	3.65	0.26	6.71	9.33

Tabela 11. Animal (AN), período (Per), silagem (SIL), concentrado (Conc), digestibilidade total da matéria seca (DMS), matéria orgânica (DMO), proteína bruta (DPB), fibra em detergente neutro (DFDN), extrato etéreo (DEE), carboidratos não-fibrosos (DCNF) e teor de nutrientes digestíveis totais (NDT). Valores em %. Exp. 4.

ANI	PER	SIL	CONC	DMS	DMO	DCP	DNDF	DEE	DNFC	NDT
47	1	A	25	70.40	70.88	67.89	60.28	80.52	82.90	72.07
34	2	A	25	55.37	56.13	67.62	43.51	67.22	65.90	57.45
28	3	A	25	63.51	64.14	57.94	50.37	82.50	80.35	65.58
25	4	A	25	75.59	76.04	73.62	61.67	85.51	88.29	76.53
25	1	A	50	68.93	69.66	63.26	45.55	80.46	84.86	70.79
47	2	A	50	70.35	71.05	62.78	51.19	73.79	83.68	72.02
34	3	A	50	71.46	72.16	68.51	54.38	78.19	82.75	73.28
28	4	A	50	74.91	75.56	72.96	46.28	74.98	89.92	76.80
28	1	B	25	62.99	63.64	65.84	54.75	81.99	72.66	64.30
25	2	B	25	71.52	72.07	70.83	57.46	77.04	86.06	71.50
47	3	B	25	61.44	62.11	61.35	50.20	78.18	71.97	61.60
34	4	B	25	60.24	60.89	65.45	51.67	78.36	72.41	62.48
34	1	B	50	69.83	70.89	71.91	55.33	77.72	73.48	70.30
28	2	B	50	68.95	69.90	64.35	52.17	84.62	80.45	69.56
25	3	B	50	73.65	74.48	61.00	52.27	79.32	89.91	74.40
47	4	B	50	73.75	74.50	69.99	56.63	76.21	82.59	74.13

Tabela 12. Animal (AN), período (Per), silagem (SIL), concentrado (Conc), digestibilidade ruminal da matéria seca (DRMS), matéria orgânica (DRMO), proteína bruta (DRPB), fibra em detergente neutro (DRFDN), extrato etéreo (DREE) e carboidratos não-fibrosos (DRCNF), (experimento 4).

ANI	PER	SIL	CONC	DRMS	DROM	DRCP	DRFDN	DREE	DRCNF
47	1	A	25	84.10	85.07	53.20	89.06	-42.23	96.33
34	2	A	25	81.12	82.72	52.13	84.43	-10.02	91.36
28	3	A	25	87.34	88.41	53.99	94.59	-16.07	90.22
25	4	A	25	80.08	81.10	63.07	97.03	-7.49	81.58
25	1	A	50	88.05	88.46	60.78	97.09	-23.33	95.03
47	2	A	50	81.69	82.29	60.93	93.28	-43.84	86.79
34	3	A	50	82.64	83.24	58.90	87.89	-80.18	92.15
28	4	A	50	75.83	76.66	51.66	103.37	33.84	76.39
28	1	B	25	86.65	87.01	57.00	96.73	-39.75	91.99
25	2	B	25	78.26	79.48	52.95	95.63	-22.05	78.62
47	3	B	25	88.06	89.17	51.55	102.96	0.15	92.77
34	4	B	25	83.50	83.94	51.91	92.39	7.74	84.20
34	1	B	50	81.01	82.58	65.52	91.63	-31.13	86.98
28	2	B	50	83.73	84.44	44.58	99.67	-60.48	91.04
25	3	B	50	88.09	88.59	75.70	95.74	39.87	89.66
47	4	B	50	76.81	77.70	57.90	96.45	-34.96	79.95

Valores expressos em %.

Tabela 13. Animal (AN), período (Per), silagem (SIL), concentrado (Conc), nitrogênio microbiano (N mic), carboidratos degradados no rúmen (CHODR) e matéria orgânica degradada no rúmen (MODR) expressos em g/d, e eficiência de síntese microbiana expressa em g PB microbiana/ kg NDT (EfPB/NDT), g N mic /kg CHODR (NmicCHO), e g N mic/ kg MODR (NmicMO), (experimento 4)

ANI	PER	SIL	CONC	N mic	EfPB/NDT	NmicMO	MODR	CHODR	NmicCHO
47	1	A	25	130.46	9.78	19.58	6.66	6.26	20.85
34	2	A	25	170.35	14.86	30.77	5.54	4.99	34.15
28	3	A	25	125.49	11.41	22.06	5.69	5.32	23.59
25	4	A	25	92.54	8.08	16.76	5.52	4.99	18.55
25	1	A	50	131.76	10.10	19.28	6.83	6.34	20.78
47	2	A	50	113.68	10.03	20.54	5.53	5.14	22.13
34	3	A	50	144.27	9.00	18.24	7.91	7.42	19.44
28	4	A	50	117.87	10.14	22.28	5.29	4.83	24.42
28	1	B	25	118.85	12.14	23.58	5.04	4.70	25.27
25	2	B	25	102.31	9.09	18.93	5.40	4.99	20.51
47	3	B	25	158.85	14.05	26.14	6.08	5.63	28.21
34	4	B	25	128.43	12.78	26.17	4.91	4.53	28.36
34	1	B	50	114.96	9.21	18.17	6.33	5.53	20.78
28	2	B	50	137.62	10.75	20.89	6.59	6.24	22.05
25	3	B	50	111.10	7.81	14.51	7.65	6.86	16.19
47	4	B	50	153.59	10.28	21.78	7.05	6.42	23.91