

PRODUCTION AND COMPOSITION OF MILK FROM COWS FED SUGARCANE SUPPLEMENTED WITH NO PROTEIN NITROGEN SOURCES OF DIFFERENT RUMINAL DEGRADABILITY

PRODUÇÃO E COMPOSIÇÃO DO LEITE DE VACAS ALIMENTADAS COM CANA DE AÇÚCAR SUPLEMENTADA COM FONTES DE NITROGÊNIO NÃO PROTEICO DE DIFERENTES DEGRADABILIDADES RUMINAL

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SUMMARY

Sugarcane (*Sacharum officinarum* L.) has high production potential of dry matter (DM) and energy per unit area; it is also easy to grow and maintains the nutritional value throughout the year, especially during drought periods. Because it has low protein content, this fraction (protein) should be corrected to optimize the energy:ammonia ratio and maximize rumen microbial fermentation processes. The objective of this study was to evaluate the efficiency of sources and levels of NPN (urea and OptigenII®) added to sugarcane (0.3, 0.6 and 0.9% - as fed) in diets with roughage: concentrate ratio 50:50 on dry matter intake (DMI), organic matter (OM), neutral detergent fiber (NDF), crude protein (CP), ether extract (EE), production and composition of milk and plasma urea nitrogen (PUN). Seven multiparous Holstein cows with 45±12 days in milk and 25 kg of milk per day were used. The cows were distributed in seven treatments in a balanced incomplete block design. All the cows, housed in a tie stall barn, were fed diets based on fresh chopped sugarcane with increasing levels of urea or OptigenII®: 0.3%, 0.6%, 0.9% and a control group without urea. The level of urea or OptigenII® in the diets was based on the amount of sugarcane in as fed basis. The dry matter intake (DMI) was not affected by the experimental diets (P>0.05) and ranged from 18.05 to 18.70 kg per day. DMI expressed as percentage body weight ranged from 2.95 to 3.45%. The NDF intake kg per day was higher for the control diet compared to the 0.9% of OptigenII® diet (P<0.05); however, no difference was detected for NDF intake as percentage of body weight. The milk yield and 3.5 % fat corrected milk yield ranged from 22.23 to 25.58 kg per day and from 19.9 to 21.6 kg per day, respectively, without difference between diets. Milk fat percentage and production ranged from 2.92% to 3.4% and from 0.72 to 0.78 kg per day, respectively. Milk protein percentage and production ranged from 2.7 to 3.11% and from 0.73 to 0.77 kg per day, with coefficient of variation of 1.08% and 16.2%, respectively. The inclusion of 0.9% of urea in the diet resulted in the highest level of milk protein. Different sources of non-protein nitrogen can be used in the diets of dairy cows producing up to 25 kg of milk per day without changing milk yield and composition.

KEY-WORDS: *Sacharum officinarum*. Dry mater intake. Urea.

RESUMO

A cana de açúcar (*Sacharum officinarum* L.) apresenta elevado potencial de produção de matéria seca (MS) e energia por unidade de área, fácil cultivo e manutenção do seu valor nutricional durante todo ano e principalmente em períodos de estiagem. Devido ao baixo teor de proteína bruta, a correção desta fração (proteína) deve ser realizada visando otimizar a relação energia:amônia e maximizar os processos fermentativos microbianos no rúmen. O objetivo do presente estudo foi avaliar a eficiência de fontes e níveis de NNP (ureia e OptigenII®), acrescidos na cana de açúcar (0,3; 0,6 e 0,9% - na matéria natural) em dietas com relação volumoso:concentrado 50:50 sobre o consumo de matéria seca (DMI), matéria orgânica (CMO), fibra em detergente neutro (CFDN), proteína bruta (CPB), extrato etéreo (CEE),

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produção e composição do leite e níveis plasmáticos de nitrogênio uréico (NUP). Sete vacas da raça Holandesa com 583 kg de peso corporal mantidas em tie stall com 45 ± 12 dias em lactação e 25 kg de leite foram distribuídas em delineamento tipo blocos incompletos balanceados com os seguintes tratamentos: ureia 0,3; 0,6 e 0,9% ou Optgen II® nos mesmos níveis e uma dieta controle sem adição de N a cana-de-açúcar.(controle). O CMS em kg ou em % PC não apresentou diferença entre os tratamentos. Houve diferença para o CFDN em kg/dia e não houve diferença ($P>0,05$) para o CFDN em % do peso vivo. Não houve diferença ($P>0,05$) para o CMO e CPB. Não houve diferença ($P>0,05$) para produção de leite. Para os teores de gordura, lactose, ESD e ST não houve diferença ($P>0,05$). Houve diferença ($P>0,05$) para o teor de proteína do leite. O nível de 0,3% de ureia é suficiente para vacas produzindo ao redor de 25 kg/leite/dia.

PALAVRAS-CHAVE: Sacharum officinarum. Consumo de matéria seca. Ureia

INTRODUCTION

The idea of using sugar cane in cattle feed is very old. Easy cultivation, harvesting during dry season and great forage production per area (120 ton/ha) make it a food of great interest to researchers and producers. Several studies (CASTRO, 1967; PAIVA et al., 1991; ALONSO & SENRA, 1992; VALVASORI et al., 1998) show the importance of sugar cane inclusion in the diet of dairy cows by assessing productive economic indices with promising and interesting results regarding its use for animal feeding. Because it is poor protein roughage, the addition of urea aims to correct this nutrient deficiency, the most expensive in the formulation of a diet (de SOUZA, 2011; FILGUEIRAS NETO et al., 2009; CARMO et al., 2005).

This study aims to evaluate the effects of using sugar cane supplemented with increasing levels of non-protein nitrogen sources of different degradability on consumption, production and milk composition of Holstein x Zebu cattle.

MATERIAL AND METHODS

The study was conducted at Professor Hélio Barbosa Farm, in Igarapé, a rural property that belongs to the Veterinary School of the Universidade Federal de Minas Gerais - UFMG.

The experiment lasted 84 days divided into four periods of 21 days each with 14 days for adaptation to the diets and seven days for sampling. Seven multiparous Holstein x Zebu cows, weighing 580 kg body weight, producing an average of 25 kg of milk per day at 45 days of lactation were housed in a tie stall type system with individual drinkers and feeding troughs. The experiment was laid out as balanced incomplete block design, so that for the blocks to be balanced, albeit incomplete, it is necessary that any pair of treatments within a block appears the same number of times in the design; therefore, there are t treatments repeated r times distributed evenly in b blocks of size k , then $r \times t = b \times k$. The design consisted of seven treatments, with four replications, 7 blocks

with four treatments each, with 88% efficiency (SAMPAIO, 2007).

The experimental diets were formulated according to the NRC 1989. Only sugarcane was used as roughage added either traditional or protected urea (OptgenII®), as shown in Table 1. OptgenII® (Alltech, Inc., Lexington, KY) is pelletized urea (43% N) coated with a biodegradable polymer, which controls the release of ammonia to the rumen. Two NPN sources (urea and OptigenII®) were used and added to the sugarcane at three levels (0.3, 0.6, and 0.9% - as fed) at the forage:concentrate ratio of 50:50 and a control diet without non-protein nitrogen source, according to Table 3.

Each of the seven cows received a sequence of four treatments for each 21-day experimental period (SAMPAIO, 2007); 14 days of adaptation and seven sampling days. Table 4 shows the chemical composition of sugar cane. The weighing and milk sampling were conducted in the morning and afternoon milkings of the 15th and 16th days of each period.

The milk collected was stored in a flask containing Bronopol and placed in the refrigerator until analysis of protein, fat, lactose, nonfat dry extract (ESD) and total solids (TS), performed by the Milk Quality Laboratory of the Veterinary School, UFMG.

The milk production corrected to 3.5% fat (FCM 3.5%) was obtained by the equation cited by Gravert (1987): $FCM\ 3.5\% = (0.432 \times MP) + (16.2 \times FP)$ where: FCM 3.5% = milk yield adjusted for 3.5% fat (kg/day); MP = milk production (kg/day), FP = fat yield (kg/day).

Individual blood samples were collected from the coccygeal vein or artery, on the last day of each experimental period (day 21), using Vacutainer with anticoagulant (EDTA). Samples were taken immediately before feeding (0:00 hours) and 1:00, 3:00, 5:00 and 8:00 hours after the first feeding. Immediately after collection, the samples were centrifuged at 2500 g for 10 minutes; supernatant (plasma) was collected and transferred to ependorffs (duplicate), and subsequently stored at -20°C. The urea concentration was determined by the colorimetric-enzymatic method using commercial kit (Urea 500, Doles Reagents and Laboratory Equipment LTD, GO).

Table 1 - Diet composition % of DM.

	Treatments ¹						
	C	0.3 U	0.6 U	0.9 U	0.3 OP	0.6 OP	0.9 OP
Sugarcane	54.0	54.0	54.0	54.0	54.0	54.0	54.0
Meal of cotton seed	8.70	8.80	6.40	3.90	8.60	6.00	5.50
Corn, corn flour	10.8	13.9	18.0	22.0	13.6	17.5	20.5
Soybean, soybean meal	22.75	18.96	16.78	14.62	19.40	17.65	14.62
Urea	-	0.50	1.0	1.6	-	-	-
Optigen®	-	-	-	-	0.5	1.0	1.6
SoyNúcleo ²	3.3	3.3	3.3	3.3	3.3	3.3	3.3
High-calcium	0.27	0.27	0.27	0.27	0.27	0.27	0.27
TOTAL %	100	100	100	100	100	100	100

2 – Vitamin and mineral supplement, with yeast and lasalocid: 13.5% Ca; 5.0% P; 2.9% Mg; 4.7% K; 9.3% Na; 4.0% S; 5.3 ppm Co; 300 ppm Cu; 650 ppm Fe; 25.6 ppm I; 1,530 ppm Mn; 12 ppm Se; 2,040 ppm Zn; 165,000 UI Vitamin A; 50,000 UI Vitamin D; 1,000 UI Vitamin E; 28 ppm Biotin; 430 ppm Lasalocid; 2.1×10^{11} UFC Yeast. 1- Treatments= C= control; 0.3U = 0.3% urea; 0.6U = 0.6% urea; 0.9U = 0.9% urea; 0.3 OP = 0.3% OptigenII®; 0.6 OP = 0.6% OptigenII®; 0.9 OP = 0.9% OptigenII®. Formula estimated by the software Spartan.

Food samples were collected in the first five days of each experimental period, as follows: 200 g of sugar cane from each animal at each feeding period and 100 g of each concentrated, the uneaten food was weighed the next day and 500 g were sampled per day. The samples were placed in plastic bags and frozen for later analysis at the Laboratory of Animal Nutrition, of the Escola de Veterinária, UFMG. At the time of the analysis, the concentrate and sugarcane samples were thawed, pre-dried in a forced ventilation oven at 55°C for 72 hours to determine pre-dried matter. Then, the samples were ground in stationary grinding mills with 1-mm diameter sieve for further analysis of dry, organic and mineral matter (MS, MM, OM), crude protein (PB) by the Kjeldahl method (AOAC, 1990), ether extract (EE) by the Soxhlet method (AOAC, 1997). Fiber analyses were performed according to the method proposed by Van Soest et al. (1991) for neutral detergent fiber (NDF) and acid detergent fiber (ADF). The averages for the dependent variables were adjusted according to the mathematical model:

$Y_{ijk} = \mu + T_i + B_j + P_k + e_{ijk}$, where
 Y_{ijk} = treatment i, in block j, for period k;
 μ = general average;
 T_i = effect of treatment i (T = 1, 2, ..., 7);
 B_j = effect of block j (J = 1, 2, ..., 7);
 P_k = effect of period k (P = 1, 2, 3, 4);
 e_{ijk} = random variation attributed to treatment i, for cow j, in period k.

RESULTS AND DISCUSSION

Table 2 shows that there was no difference in dry matter intake (DMI kg/day) and the averages ranged from 18.05 to 18.70 kg/day (CV 14.18%). The

average dry matter intake was 3.2% of live body weight, remaining constant across different levels of urea and slow release urea, with no difference between treatments.

Carmo et al. (2005) reported decreasing MS when conventional urea or starea replaced soybean meal using sugarcane as the only roughage; however, this data was not statistically analyzed since these measurements were made by animal groups and not individually as in this study. Plumer et al. (1971) observed no difference in dry matter intake when soybean meal was partially replaced by urea in the levels of 2 or 3% of the concentrate or 1.1% of diet DM. Oliveira et al. (2001) reported decreasing DMI when urea was included in the diet at levels of 1.4% and 2.1%, but noted that DMI did not change at the inclusion level of 0.7% urea in diet DM. In this study, the addition of 0.3 to 0.9% of non-protein nitrogen source, quick or slow release, did not alter consumption. At the same time, the increase of NPN concentrations at replacement levels of (0, 2.7, 5.2, 7.5%) of total RDP of MS (10.6, 11.1, 11.5, 12.0%) did not affect milk yield ($P < 0.05$). The non-significant effect for DMI, CP and OM (Table 2) may explain why milk production and FCM 3.5% were not affected, since they are more easily changed when they decrease due to intake of nutrients required for production.

The variable NDF/kg/day (Table 2) was significantly different ($P > 0.05$) ranging from 5.00 to 7.80 kg/day with CV of 12.00%. Despite the fact that there was no difference ($P > 0.05$) of NDF as % of body weight, these results corroborate those reported by Mendonça et al. (2004), who worked with sugar cane and roughage:concentrate ratios of 60:40 and 50:50. DMI expressed as % of body weight ranged from 2.95 to 3.45 of live weight with no significant difference

Table 2 – Dry matter intake (DMI), organic matter intake (OMI), crude protein intake (CPI) and neutral detergent fiber intake (NDFI) of dairy cattle fed sugarcane and supplemented with non-protein nitrogen of different rumen degradability.

<i>Variables</i>	Treatments¹								
	C	0.3 U	0.6 U	0.9 U	0.3 OP	0.6 OP	0.9 OP	CV	P
DMI, kg/day	18.33	18.45	18.70	18.53	18.05	18.34	18.21	14.18	>0.05
DMI, % PV	3.40	2.95	3.10	3.10	3.30	3.40	3.20	44.00	>0.05
OMI, kg/day	18.04	15.72	16.40	16.15	16.81	18.30	16.93	5.46	>0.05
CPI, kg/day	3.23	2.87	3.04	3.05	3.40	3.40	3.30	11.72	>0.05
NDFI, kg/day	7.80 ^a	6.00 ^a	6.30 ^a	5.80 ^a	6.70 ^a	6.90 ^a	5.00 ^b	12.00	<0.05
NDFI, %PV	1.30	1.00	1.10	0.97	1.20	1.20	0.98	1.18	>0.05
PV, kg/day	577.0 ^a	614.15 ^a	588.30 ^a	586.30 ^a	585.20 ^a	563.40 ^b	573.50 ^a	2.50	<0.05

Means with different letters in the row differ by Tukey test (p>0,05).

1- C= control, 0.3 U = 0.3% urea, 0.6 U = 0.6% urea, 0.9 U = 0.9% urea, T4= 0.3% OptigenII®, T5= 0.6% OptigenII®, T6= 0.9% OptigenII®, DMI = dry matter intake, OMI = organic matter intake, CPI = crude protein intake, NDFI = intake of NDF, LW = live weight.

Table 3 – Milk production and composition (kg/day), fat corrected milk (FCM 3.5%), protein, lactose, solids and nonfat dry extract of dairy cattle fed sugarcane and supplemented with non-protein nitrogen of different rumen degradability.

<i>Variables</i>	Treatments								
	C	0.3 U	0.6 U	0.9 U	0.3 OP	0.6 OP	0.9 OP	CV	P
milk, kg/d	23.4	24.46	24.17	22.23	24.89	25.58	24.67	11.10	>0.05
FCM 3,5%	22.11	22.88	22.10	21.75	23.38	23.68	23.13	3.00	>0.05
Fat, %	3.18	2.92	2.95	3.40	3.09	3.07	3.06	5.56	>0.05
Fat kg	0.75	0.76	0.72	0.75	0.78	0.78	0.77	11.15	>0.05
Protein %	2.94 ^{ab}	2.83 ^{ab}	2.70 ^b	3.11 ^a	3.02 ^a	2.84 ^{ab}	3.03 ^a	1.08	<0.05S
Protein kg	0.72	0.69	0.63	0.63	0.75	0.73	0.77	16.20	>0.05
Lactose, %	4.43	4.39	4.45	4.39	4.47	4.42	4.43	2.21	>0.05
Lactose kg	1.00	1.10	1.05	1.00	1.10	1.10	1.10	11.80	>0.05
ST, %	11.13	11.09	11.61	11.17	11.16	11.29	11.13	2.07	>0.05
ST kg	2.70	2.70	2.70	2.60	2.80	2.90	2.80	11.60	>0.05
NDM, %	8.23	8.22	8.03	8.31	8.23	8.30	8.21	2.77	>0.05
NDM kg	1.90	1.98	1.95	1.90	2.00	2.10	2.10	10.90	>0.05

Means followed by the same letter in the row differ by Tukey (p>0.05).

CV: coefficient of variance. P: probability. FCM: milk corrected to 3,5% of fat; TS: total solid; NDM: nonfat dry milk. C= control, 0.3 U = 0.3% urea, 0.6 U = 0.6% urea, 0.9 U = 0.9% urea, T4= 0.3% OptigenII®, T5= 0.6% OptigenII®, T6= 0.9% OptigenII®.

(P>0.05) (Table 2). The similar dry matter content (DM) can be related to the animals being in comparable lactation stages (physiological) and originating from similar productions, but the low DM may be due to the fact that sugar cane increases retention time in the rumen. Increased concentrations of blood metabolites stimulate chemical receptors

activating the satiety center causing food intake to cease, according to the chemotactic theory (CHURCH, 1993).

There was no difference (P>0.05) for the consumption of organic matter and crude protein (Table 2), which ranged from 15.72 to 18.30 kg/day and 2.87 to 3.40 kg/day, respectively. Even the

addition of different degradability NPN sources in the diets did not affect the consumption of CP, since crude protein level of all experimental diets ranged from 15.60 to 18.80% with no difference in dry matter intake ($P>0.05$). According Valadares et al. (1999), 35% of non-fiber carbohydrates (NFC) in the diet correspond to the optimum level for using NPN in the diets of dairy cattle that contain only alfalfa silage as forage. The NFC contents of the experimental diets ranged from 33.1 to 38.8%, close to 36%, the optimum value suggested by Valadares et al. (1999). Table 3 shows production and chemical composition of the milk obtained during the experimental period. The milk production and milk yield corrected for 3.5% fat (FCM 3.5%) ranged from 22.23 to 25.58 kg/day and 19.90 to 21.60 kg/day, respectively (Table 3). These two variables were not significantly different among treatments ($P>0.05$), with coefficients of variation of 11.10% and 9.4%, respectively. Therefore, partial replacement of protein nitrogen sources for non-protein ones with different degradability did not affect milk production. This lack of significant effect on milk production (kg/d) 3.5% FCM can probably be attributed to the similar DM content ($P<0.05$). Table 3 shows that there was no effect ($P<0.05$) for the DM, the non-significance for the milk production and FCM 3.5% may be related to similar DMI ($P>0.05$), which can be explained by the animals (blocks) being in near physiological stages, as required by the design and production used. Milk fat content and production ranged from 2.92 to 3.40% and from 0.72% to 0.78 kg/day, respectively (Table 3), but were not different among treatments ($P>0.05$). The replacement of true protein sources for non-protein nitrogen sources did not affect fat content and production, showing that rumen function did not affect the production of milk fat precursors, despite the low digestibility of NDF from the single roughage source of the diet.

The milk protein content and production ranged from 2.70 to 3.11 and from 0.630 to 0.770 kg/day with CV of 1.08% and 16.2%, respectively, as shown in Table 3. Crude protein milk content was significantly different ($P>0.05$). The urea inclusion level of 0.9% resulted in a higher milk protein content (3.11%) in relation to 0.6 U, which can be explained by the greater availability of nitrogen that replaced 7.5% of total RDP in this treatment along the higher NFC content (38.8%). Generally, it was less than 3.2%, which shows lower efficiency of microbial protein production in the rumen and subsequent supply of amino acids in the intestine. The efficiency of crude protein diet use for conversion into milk protein ranged from 20 to 25%. Although the 0.9 OP experimental group displayed higher protein content in the milk, the level is still below the 3.2% mean value used for differential payment for milk quality. According to Brito and Broderick et al. (2007), this diet may have had a more balanced amino acid profile formed by microbial protein, especially the most limiting (lysine and methionine), which may help to reduce nitrogen excretion and thereby did not compromise milk and protein production. However, Broderick et al. (2009), working with high-producing cows (40.0 kg milk),

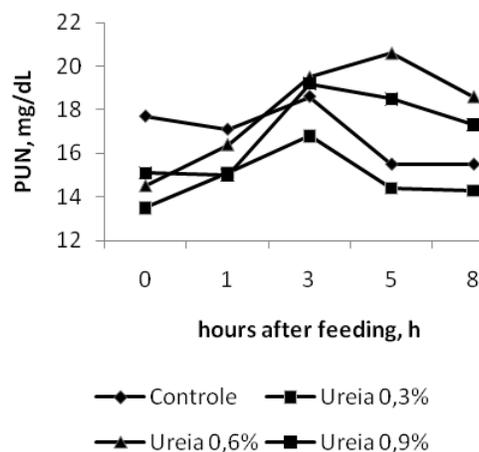
showed that replacing soybean meal RDP with conventional urea RDP decreased milk and other components production, which can reduce microbial protein production in the rumen. In addition, the use of NPN source as RDP source it is not as efficient as true protein sources to optimize protein production.

In the present study milk lactose content and production (Table 3) ranged from 4.47 to 4.39% and between 1.00 and 1.10 kg/day with CV of 2.21% and 11.8%, respectively. Broderick et al. (2009), working with high-producing cows (40.0 kg milk) and a diet with 16.1% crude protein and 10.5% RDP with urea levels of 0, 1.2, 2.4 and 3.7% of diet dry matter observed linear decrease in lactose content of milk, dry matter intake and corrected milk yield (FCM 3.5%) while the percentages of protein, fat and milk solids were not affected.

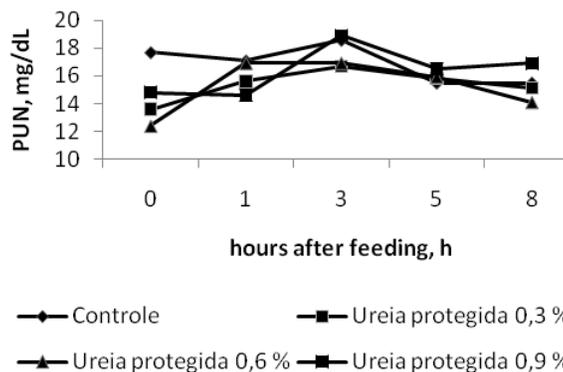
The percentage and production mean values for total solids ranged from 11.29% to 11.09% and from 2.90 to 2.7 kg/day while for nonfat dry extract varied from 8.03 to 8.31% and from 1.90 to 2.10 kg/day, respectively. These milk components were not different for the different treatments ($P>0.05$). Several other studies using NPN in the diet found no statistical difference ($P>0.05$) for the total solids and nonfat dry extract (BRITO & BRODERICK., 2007; IMAIZUMI et al., 2003; CARMO et al., 2005; AQUINO et al., 2009).

Figure 1 shows the mean values for plasma urea nitrogen (PUN mg/dL) at different sampling times, which were not significantly different ($P>0.05$) for PUN levels ranging from 12.40 to 17.70 mg/dL before the first feeding (time zero; CV=15.34%), 16.70 to 19.50 mg/dL three hours after the first feeding (CV=12.90%) and 14.10 to 18.60 mg/dL, eight hours following the first feed (CV=17.72%).

These results agree with those found by Claypool et al. (1980) and Howard et al. (1987), who observed higher PUN values two and three hours after feeding; however, these authors found difference ($P<0.001$) for the levels of 12.7, 16.3 and 19.3% crude protein in the diet and associated this PUN increase with the supply of highly degradable protein and a high crude protein intake from the diet. The authors found serum levels of 7.5, 10.9 and 17.9 mg/dL when protein levels in the diet were 12.7, 16.3 and 19.3%, respectively, and emphasized that the PUN is strictly associated with the intake of dietary nitrogen. As seen in Table 2, the crude protein intake did not differ ($P>0.05$) for the PUN values among experimental treatments, regardless of non-protein nitrogen source. At three hours after feeding (time at which PUN levels are higher), PUN ranged from 15.0 to 19.2 mg/dL for the 0.9 U treatment probably due to the greatest replacement degree of true RDP source for NPN at 7.5% of total 12.0% of the total RDP in this treatment, which indicates loss of dietary nitrogen. As the animals were fed twice a day at eight-hour intervals, it is important to divide the diets with this urea content in at least three, since at this time (the first 3 hours after feeding) where PUN is higher (19.2 mg/dL) synchronization of nitrogen utilization and CNF could occur, reducing this waste.



Control, Urea 0.3%, Urea 0.6%, Urea 0.9%



Control, Protected Urea 0.3%, Protected Urea 0.6%, Protected Urea 0.9%

Figure 1 - Plasma urea nitrogen (PUN mg/dL) at different sampling times.

PUN average values for the experimental diets are similar to those found in the literature, where PUN varies between 14.9 and 21.0 mg/dL (FERGUSON et al., 1989; LARSON et al., 1997; RAJALA-SCHULTZ et al., 2001), indicating loss of dietary nitrogen. PUN values are highly correlated with those of ammonia nitrogen in the rumen fluid and the milk urea nitrogen levels (MUN) (CLAYPOOL et al., 1980). Researchers have mentioned the high correlation (r) between the MUN and PUN contents, ranging from 0.88 to 0.96 (CLAYPOOL et al. 1980; HOWARD et al. 1987; BAKER et al. 1995). Numerically, the highest PUN averages are found between three and five hours after feeding. According to Howard et al. (1987), the highest PUN concentrations at these times are probably due to the high rate of degradation of concentrates, concluding that this is when the ammonia nitrogen levels are higher.

CONCLUSION

For cows with daily milk production of 25.0 kg, several non-protein nitrogen sources can be used in the diet, slow release or not, as they do not alter production or composition of milk regarding fat, lactose, nonfat dry extract and total solids, when NPN sources at the three inclusion levels were replaced by true protein sources. Given that milk production and composition were not affected by NPN level, except for protein content, the level of inclusion is going to be defined by the cost of the daily diet. Treatments 0.3 U and 0.6 U have better economic results when payment is made by produced milk volume. As for quality, the milk composition of treatment 0.6 U was more inefficient regarding the amount of proteins synthesized in the mammary gland, which is determined by the amount of amino acids absorbed in the small intestine from the microbial protein produced in the rumen and the

quality of ruminal non degradable protein (RNDP) that reach the small intestine largely intact. Thus to achieve maximum revenue on feed costs, the diets should contain adequate levels of usable nutrients, thus ensuring higher milk production and composition in accordance with market requirements.

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REFERENCES

- ALONSO, J. R.; SENRA, A. Production system for dairy cows without irrigation an whole sugar cane forage supplied during the dry season. Milk production and composition and live weight. **Cuban Journal Agriculture Science**, n.26, p.123, 1992.
- ASSOCIATION OF AFFICIAL ANALYTICAL CHEMISTS- AOAC. **Official methods of analysis**. 15^a ed. Washington D.C., 1990. 1141 p.
- ASSOCIATION OF AFFICIAL ANALYTICAL CHEMISTS -AOAC. :edited Ig W. Horwitz 16^{ed}. Washington, 1997. p. 850.
- AQUINO, A. A.; JUNIOR PEIXOTO, K. C.; GIGANTE, M. L.; et al..Efeitos de níveis crescentes de ureia na dieta de vacas leiteiras sobre a composição e rendimento da fabricação de queijos minas frescal. **BRAZILIAN JOURNAL VETERINÁRIA. RESEARCH ANIMAL SCIENCE**, São Paulo, v.46, n.4, p.273-279, 2009.
- BAKER, L. D.; FERGUSON, J. D.; CHALUPA, W. Responses in urea and true protein of milk to different protein feeding schemes for dairy cows. **Journal of Dairy Science**, v.78, n.9, p.2424-2434, 1995.
- BRITO, A. F; BRODERICK, G. A. Effects of different protein supplements on milk production and nutrient utilization in lactating dairy cows. **Journal of Dairy Science**, Albany, v.90, n.4, p.1816-1827, 2007.
- BRODERICK, G. A.; REYNAL, S. M. Effect of source of rumen-degraded protein on production and ruminal metabolism in lactating dairy cows. **Journal of Dairy Science**, v.92, p.2822-2834, 2009.
- CARMO, C. A; SANTOS, F. A. P; IMAIZUMI, H; PIRES, A. V; SCOTON, R. A: Substituição do farelo de soja por ureia ou amiréia para vacas em final de lactação. **Acta Scientiarum**. v.27, n.2, p.277-286, 2005.
- CASTRO, A. C. G. **Cana de açúcar versus silagem de milho na produção de leite**. (DISSERTAÇÃO DE MESTRADO EM ZOOTECNIA). Universidade Federal de Viçosa, p.37, 1967.
- CHURCH, D. C. **The ruminant animal digestive physiology and nutrition**. New Jersey: Waveland Press, 1993.
- CLAYPOOL, D. W; PANGBORN, M. C; ADAMS, H. P: Effect of dietary protein on high-producing dairy cows in early lactation. **Journal of Dairy Science** , v.63, p.833-837, 1980.
- DE SOUZA, R. C. **Adição Crescente de Uréia na Cana de Açúcar (SACHARUM OFFICINARUM L.) em Dietas de Vacas em Lactação** (TESE DE DOUTORADO). Universidade Federal de Minas Gerais. p.122, 2011.
- FILGEIRAS NETO, G.; REIS, R. B.; SOUSA, B. M.; et al. Efeito da substituição parcial do farelo de soja por ureia de liberação controlada ou não no consumo e produção de leite para vacas em lactação alimentadas com dietas baseadas em cana-de-açúcar. In: REUNIÃO ANUAL DA SOCIEDADE BRASILEIRA DE ZOOTECNIA, 46, 2009, Maringá-PR.
- FERGUSON, J. D.; CHALUPA, W. Impact of protein nutrition in dairy cows. **Journal of Dairy Science**. v.72, n.3, p.746-766, 1989.GRAVERT, H. O. Dairy cattle production. Nova York: Elsevier Science, 1987, 234 p.
- HOWARD, H. J; AALETH, E. P; ADAMS, G. D; BUSH, L. J: Ifluence of dietary protein on reproductive performance of dairy cows. **Journal of Dairy Science** , v.70, p.1563-1571,1987.
- IMAIZUMI, H; SANTOS, F. A. P; PIRES, A. V; NUSSIO, C. M. B; BARNABE, E. C; JUCHEM, S. O. Avaliação de diferentes fonte e teores de proteína na dieta sobre o desempenho, fermentação ruminal e parâmetros sanguíneos de vacas da raça Holandesa em final de lactação. **Acta Scientiarum**, Maringá, v.25, n.4, p.1031-1037, 2003.
- LARSON, S. F; BUTLER, W. R; CURRIE, W. B. Reduced fertility associated with low progesterone postbreeding and increase milk urea nitrogen in lactating cows. **Journal of Dairy Science** , v.80, n.7, p.1288-1295, 1997.
- MENDONÇA, S. S; CAMPOS, J. M. S; VALADARES FILHO, S. C. Consumo, digestibilidade aparente, produção e composição do leite e variáveis ruminais em vacas leiteiras alimentadas com dietas a base de cana de açúcar. **Revista Brasileira de Zootecnia**, v.33, n.2, p.481-492, 2004.
- NUTRIENT REQUERIMENTS OF DAIRY CATTLE. Sixth Revised Edition Update, 1989, p.157.
- OLIVEIRA, A. S.; VALADARES, R. F. D.; VALADARES, S. C. Consumo, digestibilidade aparente, produção e composição do leite em vacas alimentadas

com quatro níveis de compostos nitrogenados não proteicos. **Revista Brasileira de Zootecnia**, v.30, n.4, p.1358-1366, 2001.

PAIVA, J. A. J.; MOREIRA, H. A.; CRUZ, G. M.; et al. Cana-de-açúcar associada à ureia/sulfato de amônio como volumoso exclusivo para vacas em lactação. **Revista Brasileira de Zootecnia**, v.20, n.1, p.90-99, 1991.

PLUMER, J. R.; MILES, J. T.; MONTGOMERY, M. J.; Effect of urea in the concentrate mixture on intake and production of cows fed corn silage as the only forage. **Journal of Dairy Science**, v.54, p.1861-1865, 1971.

RAJALA-SHULTZ, P. J; SAILLE, W. J. A; FRAZER, G. S. Association between milk urea nitrogen and fertility in Ohio dairy cows. **Journal of Dairy Science**, v.84, n.2, p.482-489, 2001.

SAMPAIO, I. B. M: Estatística Aplicada a Experimentação Animal. 3ª Ed. Belo Horizonte: **Fundação de Estudo e Pesquisa em Medicina Veterinária e Zootecnia**, 2007. 264p.

VALADARES FILHO, S. C; BRODERICK, G. A; VALDARES, R. F. D. Effect of replacing alfafa silage with high moisture corn on ruminal protein synthesis estimated from excretion of total purine derivatives. **Journal of Dairy Science**, v.82, p.2686-2696, 1999.

VALVASORI, E.; LAVEZZO, W.; LUCCI, C. A.; et. Al. Alteração na fermentação de bovinos fistulados alimentados com cana-de-açúcar em substituição à silagem de milho. In: REUNIÃO ANUAL DA SOCIEDADE BRASILEIRA DE ZOOTECNIA, 35., 1998, Botucatu. Anais... Botucatu: SBZ, 1998 (Disponível em CD-ROM).

VAN SOEST, P. J.; ROBERTSON, J. B. et al. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. **Journal of Dairy Science**, v.74, p.3583-3598, 1991.