



## Sugar cane fresh or ensiled with or without bacterial additive in diets for dairy cows

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**ABSTRACT.** This study evaluated the effect of using fresh sugar cane, sugar cane silage with or without *Lactobacillus buchneri*, and burnt sugar cane silage with or without *L. buchneri* on ingestive behavior, nitrogen balance and synthesis of microbial nitrogen compounds of dairy cows. Five  $\frac{3}{4}$  Holstein x Gir crossbred cows, assigned to a 5 x 5 Latin square design, were given diets with a 60:40 forage: concentrate ratio on a dry matter basis, to meet an average body weight of 550 kg and production of 15 kg of milk per day. The treatment with fresh sugar cane showed higher values ( $p < 0.05$ ) for dry matter feed efficiency (3,133.3 vs 2,234.47 g DM h<sup>-1</sup>) and rumination (1,642.3 vs 1,222.93 g DM h<sup>-1</sup>) compared to the silages. There was a shorter total chewing time (58.7 vs 81.5 min. kg<sup>-1</sup> DM) for fresh sugar cane when compared to silages without inoculum. The addition of microbial additive during ensiling of sugar cane did not alter ( $p > 0.05$ ) the nitrogen intake and balance, but led to a greater ( $p < 0.05$ ) synthesis of microbial nitrogen. Microbial efficiency was not affected ( $p > 0.05$ ), and showed an average value of 204.32 g microbial crude protein kg<sup>-1</sup> total digestible nutrients.

**Keywords:** *Lactobacillus buchneri*, microbial synthesis, nitrogen balance, *Saccharum officinarum*.

## Cana-de-açúcar in natura e ensilada com ou sem aditivo bacteriano em dietas para vacas lactantes

**RESUMO.** Objetivou-se avaliar o efeito da utilização de cana-de-açúcar *in natura*, silagem de cana sem ou com *Lactobacillus buchneri* e cana queimada sem ou com *L. buchneri* sobre o comportamento ingestivo, balanço de nitrogênio e síntese de compostos nitrogenados microbianos em vacas lactantes. Foram utilizadas cinco vacas mestiças,  $\frac{3}{4}$  Holandês x Gir, distribuídas em um quadrado latino 5 x 5 e alimentadas com dietas compostas por 60% de volumoso e 40% de concentrado na base da matéria seca, para atender um peso corporal médio de 550 kg e produção média de 15 kg de leite por dia. A cana-de-açúcar *in natura* proporcionou maiores valores ( $p < 0,05$ ) para eficiência de alimentação da matéria seca (3.133,3 vs 2.234,47 g MS h<sup>-1</sup>) e de ruminação (1.642,3 vs 1.222,93 g MS h<sup>-1</sup>) em relação às silagens. Observou-se menor tempo de mastigação total (58,7 vs 81,5 min. kg<sup>-1</sup> de MS) para cana-de-açúcar *in natura* quando comparada às silagens sem inóculo. A utilização do aditivo microbiano na ensilagem da cana-de-açúcar não alterou ( $p > 0,05$ ) o consumo e o balanço de nitrogênio, porém propiciou maior ( $p < 0,05$ ) síntese de nitrogênio microbiano. Não houve diferença entre as dietas ( $p > 0,05$ ) para eficiência microbiana, cujo valor médio foi de 204,32 g PB microbiana kg<sup>-1</sup> de TDN.

**Palavras-chave:** *Lactobacillus buchneri*, síntese microbiana, balance de nitrogenio, *Saccharum officinarum*.

### Introduction

For a long time, farmers and researchers have sought improvements in the production of forages, which is a major factor responsible for the production of milk and meat in Brazil. Such improvements drive the search for the economic balance of the activity, beyond the aspect related to the production and yield.

In Brazil, sugar cane is a major forage resource for feeding dairy and beef cattle, given the availability during periods of shortage of forage in pastures, which explains the great interest for its high potential

production than for its nutritional value, when compared to corn or sorghum.

The use of fresh sugar cane requires daily cuts and high investments, due to hand labor that increases its cost, transportation and shredding, which may cause limitations to feed larger herds. Moreover, the daily cut becomes problematic when one wants to use sugarcane as forage throughout the year, due to the difficulty of harvesting on rainy days and loss of nutritional value during the summer.

The controlled burning of sugar cane aimed at facilitating harvesting is a common practice in Brazil,

especially in non-mechanized areas and in places whose soil structure prevents mechanization.

Sugar cane harvesting for ensiling allows the release of the area for homogeneous regrowth of plants, resulting in better soil cover and higher leaf area index for the rainy season and hence lower spending on weed control (FREITAS et al., 2006). For these reasons, there has been growing search for sugar cane silage production technologies, as well as for the improvement in animal performance with its use in ruminant feed.

The preservation of sugarcane by ensiling provides homogenization of the ensiled material, but it has been shown that ensiling of sugar cane alone causes marked reduction in its nutritional value due to the rapid fermentation of soluble sugars into ethyl alcohol by yeasts. Thus, chemical additives and microbial inoculants have been used to improve the fermentation and preservation of silages, since they promote the growth of beneficial microorganisms, such as lactic acid producing bacteria and inhibit of undesirable ones, such as yeast and clostridia (FERREIRA et al., 2007), but its application has resulted in variable effects (PEDROSO et al., 2008).

The present study aimed to evaluate the use of silages of fresh and burnt sugar cane with or without *L. buchneri* and fresh sugar cane in diets for crossbred dairy cows on ingestive behavior, nitrogen balance and synthesis of microbial protein.

## Material and methods

The experiment was conducted at the Polo Regional de Desenvolvimento Tecnológico da Alta Mogiana – APTA in Colina, São Paulo State, Dairy Cattle sector. We used five crossbred cows, 3/4 Holstein x Gir, with an average of 100 days of lactation at the beginning of the experiment. Tables 1 and 2 show the ingredients of the concentrate and chemical composition of the diets formulated according to NRC (2001) to meet the requirements of mid lactation cows, mean body weight of 550 kg and production of 15 kg day<sup>-1</sup> of milk.

**Table 1.** Percentage composition of the concentrate, expressed on natural matter basis.

Ingredient	%
Corn	56.41
Soybean meal	18.08
Cottonseed meal	17.49
Urea + Ammonium sulfate (90:10)	3.06
Dicalcium phosphate	1.55
Limestone	0.92
Common salt	0.36
Sulfur Flower	0.12
Fosbovi 20 <sup>1</sup>	2.01

<sup>1</sup>Amount per kg of product (calcium 120 g, iodine 75 mg, phosphorus 88 g, manganese 1300 mg, sodium 126 g, selenium 15 mg, sulfur 12 g, zinc 3630 mg, cobalt 55.5 mg, fluorine 880 mg, copper 1530 mg, vehicle q.s. 1,000 mg, iron 1,800 mg).

Animals were assigned to a 5 x 5 Latin square to evaluate the effects of fresh sugar cane, sugar cane silage with *Lactobacillus buchneri*; sugar cane silage without *Lactobacillus buchneri*; burnt sugar cane without *L. buchneri* and burnt sugar cane with *L. buchneri* (Table 2).

**Table 2.** Chemical composition of the concentrate, roughage and experimental diets, on a dry matter basis.

Nutritional component (%)	Roughages					Concentrate
	Fresh	SCSI	BSCSWI	SCSWI	BSCI	
Dry matter	25.6	24.1	24.1	24.8	24.9	94.6
Organic matter	97.0	95.8	97.2	95.7	97.1	92.8
Crude protein	2.4	3.5	3.2	3.1	3.0	29.4
Ether extract	1.7	3.5	3.1	2.8	2.8	2.5
Neutral detergent fiber	51.5	67.4	65.6	72.9	61.6	28.5
NDFcp <sup>1</sup>	47.1	62.8	60.6	67.4	57.9	17.5
Acid detergent fiber	39.1	47.8	52.5	53.4	47.4	19.2
NDIN <sup>2</sup>	43.2	35.1	41.8	38.8	39.4	12.7
ADIN <sup>3</sup>	13.0	13.9	13.4	16.2	12.7	9.6
Non-fiber carbohydrates	45.9	26.3	30.5	22.5	33.8	43.3
Total carbohydrates	93.0	89.2	91.1	89.8	91.7	60.8
Ashes	3.0	4.2	2.8	4.3	2.9	7.2
Lignin	7.6	8.6	8.9	9.2	8.0	2.7
	Diets					
Dry matter	53.2	52.3	52.3	52.7	52.8	-
Organic matter	95.3	94.6	95.4	94.5	95.4	-
Crude protein	13.2	13.9	13.7	13.6	13.5	-
Ether extract	2	3.1	2.9	2.7	2.7	-
Neutral detergent fiber	42.3	51.8	50.7	55.1	48.3	-
NDFcp <sup>1</sup>	35.3	44.7	43.4	47.4	41.7	-
Acid detergent fiber	31.1	36.4	39.2	39.7	36.1	-
NDIN <sup>2</sup>	31.0	26.1	30.2	28.4	28.7	-
ADIN <sup>3</sup>	11.6	12.2	11.9	13.6	11.5	-
Non-fiber carbohydrates	44.8	33.1	35.6	30.8	37.6	-
Total carbohydrates	80.1	77.8	79	78.2	79.3	-
Ashes	4.6	5.4	4.5	5.5	4.6	-
Lignin	5.7	6.3	6.4	6.6	5.9	-
TDN	60.0	59.0	56.8	54.9	55.0	-

SCSI – sugar cane silage with *L. buchneri*; BSCSWI – burnt sugar cane silage without *L. buchneri*; SCSWI – sugar cane silage without *L. buchneri* and BSCI – burnt sugar cane silage with *L. buchneri*. <sup>1</sup>Neutral detergent fiber corrected for ash and protein (FDNcp); <sup>2</sup>neutral detergent insoluble nitrogen (NDIN); <sup>3</sup>acid detergent insoluble nitrogen (ADIN); <sup>2,3</sup>expressed as percentage of total N.

The variety of cane sugar IAC-86-2480 composed the forages, which constituted 60% of the dry matter of the diets. The experiment consisted of five experimental periods, lasting 15 days each, the first 10 days for adaptation and 5 days for data collection. Animals were weighed at the beginning and end of each period after the morning milking, to measure the body weight variation (VPC). Animals were kept in roofed individual pens, equipped with trough and drinking fountain, and received food in the form of complete mixture twice daily, after the morning and afternoon milkings, with free access to diets and water. The leftovers of the morning and afternoon were weighed daily and diets were adjusted to correspond to 10% leftovers of the total supplied. The voluntary intake was calculated as the difference between the supplied and leftovers.

The feeding behavior the animals was assessed on the 13<sup>th</sup> day of each period, by observing every 10 minutes for 24 hours, to determine the time spent eating, ruminating and idling. At night, animals were

observed in artificial lighting. On the 14<sup>th</sup> day, we counted the number of cud chews and the time spent ruminating each cud, using a digital stopwatch. Feeding and rumination efficiencies, number of cuds per day, total chewing time per day and number of cud chews per day were obtained according to Burger et al. (2000).

Cows were mechanically milked twice a day, and we recorded the milk production. We collected milk samples on day 11 in the afternoon and on day 12 in the morning of each period, making composite samples, with 1% of production for analysis of allantoin and total nitrogen. The milk was deproteinized with trichloroacetic acid (10 mL milk were mixed with 5 mL 25% trichloroacetic acid, filtered through filter paper and stored at -20°C). Allantoin analysis was performed with the deproteinized filtrate according to Valadares et al. (1999).

Feces samples were taken on the 11<sup>st</sup> and 15<sup>th</sup> day, of each period, one in the morning and afternoon of each day, respectively, according to Itavo et al. (2002). After collection, 10% aliquots were frozen for later analysis of total nitrogen.

The nitrogen balance was obtained by subtracting the total nitrogen intake from the total nitrogen excreted in feces, urine and milk.

Urine samples were collected on day 12, 4 hours after the morning feeding (spot urine), massaging the vulva and collecting the total volume of urine with a container to obtain a representative sample, for a later sampling of 10 mL urine, which where diluted with 40 mL 0.036 N sulfuric acid to prevent decomposition of metabolites, and immediately frozen for analysis of creatinine, total nitrogen, uric acid and allantoin.

Daily urine volume of each animal was quantified by multiplying the respective body weight by the amount of creatinine excreted daily and dividing the product by creatinine concentration (mg L<sup>-1</sup>) in the spot urine sample. The average daily creatinine excretion was 24.31 expressed in mg kg<sup>-1</sup> BW<sup>-1</sup> (OJEDA et al., 2005).

The analysis of allantoin in the urine and milk were carried out following the methodology of Chen and Gomes (1992), and the determination of uric acid and creatinine in urine were performed using commercial kits (Bioclin®).

Total excretion of purine derivatives was calculated by summation of the amounts of allantoin and uric acid excreted in the urine and the amount of allantoin secreted in milk, expressed in mmol day<sup>-1</sup>. Absorbed purines (X, mmol day<sup>-1</sup>) were calculated from the excretion of purine derivatives, (Y, mmol day<sup>-1</sup>), with the equation  $Y = 0.85X + 0.385 BW^{0.75}$ , where 0.85 is the recovery of purines absorbed as purine derivatives and 0.385 BW<sup>0.75</sup> is the endogenous contribution for purine excretion (VERBIC et al., 1990). The synthesis

of microbial nitrogen compounds in the rumen (Y, g N day<sup>-1</sup>) was calculated on the basis of absorbed purines (X, mmol day<sup>-1</sup>) by the equation:

$$Y(\text{g N day}^{-1}) = \frac{70X}{0.83 \times 0.116 \times 1000}$$

In which, 70 is the N content in purines (mg N mmol<sup>-1</sup>), 0.83 is digestibility of microbial purine and 0.116 is the purine N:total N ratio in bacteria (CHEN; GOMES, 1992).

Results were subjected to analysis of variance and comparison of means by Tukey's test at 5% significance, with the aid of the System for Genetic Analysis and Statistics - SAEG 8.0 (UFV, 1999).

## Results and discussion

Time spent in feeding, ruminating and idling were not significantly different ( $p > 0.05$ ) between diets used (Table 3).

**Table 3.** Time spent (hours day<sup>-1</sup>) in feeding, ruminating and idling by dairy cows in different experimental diets.

Activity	Diets					CV(%)
	Fresh	SCSI	BSCSWI	SCSWI	BSCI	
	Feeding					
(h day <sup>-1</sup> )	5.10	5.37	5.07	5.57	5.80	11.4
min. Kg <sup>-1</sup> DM	20.2 <sup>b</sup>	25.4 <sup>a</sup>	27.5 <sup>a</sup>	28.2 <sup>a</sup>	27.3 <sup>a</sup>	9.58
min. kg <sup>-1</sup> NDF	51.1	54.0	55.9	52.6	58.2	9.82
	Ruminating					
(h day <sup>-1</sup> )	9.75	10.13	10.0	10.20	9.27	11.8
min. kg <sup>-1</sup> DM	38.5 <sup>b</sup>	48.7 <sup>ab</sup>	54.2 <sup>a</sup>	53.1 <sup>ab</sup>	43.7 <sup>ab</sup>	15.24
min. Kg <sup>-1</sup> NDF	97.4	104.3	110.5	100.1	93.4	17.31
	Idling					
(h Day <sup>-1</sup> )	9.16	8.50	9.40	8.67	9.17	16.9

SCSI – sugar cane silage with *L. buchneri*; BSCSWI – burnt sugar cane silage without *L. buchneri*; SCSWI – sugar cane silage without *L. buchneri* and BSCI – burnt sugar cane silage with *L. buchneri*. Means followed by the same letter in the row are not significantly different by Tukey's test ( $p > 0.05$ ); CV: Coefficient of variation.

Similarly, Schmidt et al. (2007) worked with chemical and biological additives in the ensiling of sugar cane and observed no change in feeding behavior of dairy cows, and reported average daily values of 3.84 and 8.66h for feeding and rumination, which are lower compared to those obtained in this study (5.4 and 9.9h day<sup>-1</sup>, respectively). Diets composed of sugar cane silage showed the highest NDF content, possibly due to fluid loss in the effluent, and had the same total time spent eating and ruminating in relation to the diet with fresh sugar cane, which can be explained by the increased frequency of visits of animals to the trough within 24 hours (Table 3).

The fresh sugar cane resulted in less time spent on intake per unit kg of dry matter (20.2 min. kg<sup>-1</sup> DM) and also on rumination (38.5 min. kg<sup>-1</sup> DM) ( $p < 0.05$ ) probably due to the effect of ensiling, which increased the NDF content of the diets in association with altered palatability.

The diet with fresh sugar cane was significantly different from the others, with higher mean value for efficiency of feeding and rumination of dry matter in g DM h<sup>-1</sup>, for increasing the intake, once the time spent on feeding was similar to those obtained with diets with silages containing or not the inoculum (Table 4).

This increased intake of DM may be related to better palatability of fresh sugarcane allied to lower content of NDF. Thus, even with the lowest NDF of the diet with fresh sugar cane, its consumption approached the other diets due to increased DM intake (Table 4).

On the other hand, Sousa et al. (2009) examined diets containing corn and fresh cane sugar silages with 0, 7, or 14% cottonseed, and observed a value of 3101.70 g DM hour<sup>-1</sup> for EAL in cows fed chopped sugarcane without cottonseed in a forage: concentrate ratio of 60:40, similar to the presented study. In this work, authors reported that both intake and efficiencies of feeding and ruminating DM and NDF were higher for diets with corn silage compared with those containing sugar cane, emphasizing that the NDF content was higher for corn silage, which showed a better digestibility.

In this way, it can be inferred that higher feeding efficiency of DM for the diet with fresh sugar cane resulted from the higher CMS (kg day<sup>-1</sup>), and consequently the increased rumination efficiency (g DM h<sup>-1</sup>) reflected the ruminal capacity of processing feed and thus improving the feeding efficiency.

Feeding and rumination efficiency of NDF, total chewing time in min. kg<sup>-1</sup> NDF, number of cuds,

number of cud chews per day, number of chews per cud, and rumination time per cud were not different between diets ( $p > 0.05$ ) (Table 4). Since the total chewing time (TMT) expressed in min. kg<sup>-1</sup> DM was shorter for the diet with fresh sugar cane, and given the similarity in TRB and NBR between diets, the reduction probably occurred in the time spent chewing during feeding.

**Table 4.** Intake of DM (IDM) and NDF (INDF), feeding efficiency of DM (FE<sub>DM</sub>) and of NDF (FE<sub>NDF</sub>), rumination efficiency of DM (RE<sub>DM</sub>) and of NDF (RE<sub>NDF</sub>), total chewing time (TCT), number of cuds (NC), number of cud chews NCCnd), number of chews per cud (NCCnc) and rumination time per cud (RTC) of dairy cows in different experimental diets.

Item	Diets					CV(%)
	Fresh	SCSI	BSCSWI	SCSWI	BSCI	
CMS (kg day <sup>-1</sup> )	15.37 <sup>a</sup>	12.71 <sup>b</sup>	11.15 <sup>b</sup>	11.83 <sup>b</sup>	12.77 <sup>b</sup>	6.6
CFDN (kg day <sup>-1</sup> )	6.06	6.11	5.51	6.36	6.03	11.4
EAL <sub>DM</sub> (g DM h <sup>-1</sup> )	3133.3 <sup>a</sup>	2382.3 <sup>b</sup>	2221.3 <sup>b</sup>	2147.7 <sup>b</sup>	2186.6 <sup>b</sup>	7.6
ERU <sub>DM</sub> (g DM h <sup>-1</sup> )	1642.3 <sup>a</sup>	1243.5 <sup>b</sup>	1115.4 <sup>b</sup>	1160.3 <sup>b</sup>	1372.5 <sup>b</sup>	10.2
EAL <sub>NDF</sub> (g NDF h <sup>-1</sup> )	1197.4	1150.2	1104.4	1149.5	1035.2	11.3
ERU <sub>NDF</sub> (g NDF h <sup>-1</sup> )	626.4	601.6	555.9	628.2	650.2	15.8
TMT (h day <sup>-1</sup> )	14.9	15.5	15.1	15.8	15.1	10.4
TMT (min. kg <sup>-1</sup> DM)	58.7 <sup>b</sup>	74.1 <sup>a</sup>	81.7 <sup>a</sup>	81.3 <sup>a</sup>	70.9 <sup>b</sup>	12.1
TMT (min. kg <sup>-1</sup> NDF)	148.5	158.4	166.4	152.7	151.6	13.9
NBR (n day <sup>-1</sup> )	701.9	722.2	729.6	739.8	678.2	14.8
NMMD (n day <sup>-1</sup> )	37184.9	38136.6	38232.8	38669.5	34686.2	14.1
NMMB (n cud <sup>-1</sup> )	52.9	52.8	52.6	52.3	51.5	5.4
TRB (seg. cud <sup>-1</sup> )	50.4	50.8	50.0	49.8	49.7	6.9

SCSI – sugar cane silage with *L. budmeri*; BSCSWI – burnt sugar cane silage without *L. budmeri*; SCSWI – sugar cane silage without *L. budmeri* and BSCI – burnt sugar cane silage with *L. budmeri*. Means followed by the same letter in the row are not significantly different by Tukey's test ( $p > 0.05$ ); CV: Coefficient of variation.

The production of nitrogen compounds in milk in g day<sup>-1</sup>, % BW and g<sup>-1</sup> BW<sup>0.75</sup> was significantly different ( $p < 0.05$ ) between treatments (Table 5).

**Table 5.** Intake, excretion (N milk, N urine and N feces), and balance of nitrogen and body weight variation (VPC) of dairy cows in different experimental diets.

Items	Diets					CV%
	Fresh	SCSI	BSCSWI	SCSWI	BSCI	
	g day <sup>-1</sup>					
N Intake	353.0	299.4	286.7	294.4	309.2	16.70
Milk N	69.8 <sup>a</sup>	58.6 <sup>bc</sup>	56.2 <sup>bc</sup>	54.7 <sup>c</sup>	63.3 <sup>ab</sup>	6.44
Urine N	122.3	135.8	111.9	119.3	117.6	16.77
Feces N	143.9	93.3	97.1	97.0	117.3	25.50
N balance	16.9	11.7	21.5	23.4	10.9	368.79
	% BW					
N Intake	76.5	64.4	63.2	63.4	67.1	17.24
Milk N	15.2 <sup>a</sup>	12.6 <sup>bc</sup>	12.4 <sup>bc</sup>	11.8 <sup>c</sup>	13.7 <sup>ab</sup>	5.84
Urine N	26.9	29.4	24.7	25.5	25.4	16.14
Feces N	31.5	20.2	21.3	20.9	25.6	26.94
N balance	2.86	2.16	4.86	5.29	2.39	394.51
	g kg <sup>-1</sup> BW <sup>0.75</sup>					
N Intake	3.55	2.99	2.92	2.94	3.11	17.00
Milk N	0.71 <sup>a</sup>	0.59 <sup>bc</sup>	0.57 <sup>bc</sup>	0.55 <sup>c</sup>	0.64 <sup>ab</sup>	5.91
Urine N	1.25	1.36	1.14	1.18	1.18	16.18
Feces N	1.46	0.94	0.98	0.97	1.18	26.62
N balance	0.14	-1.85	-0.90	0.24	-1.23	412.51
	% N intake					
Milk N	20.7	19.7	19.6	18.7	20.6	14.01
Urine N	36.2	45.6	39.2	40.8	39.2	22.42
Feces N	41.2	31.5	34.1	32.7	39.2	28.07
N balance	1.92	3.27	7.11	7.85	1.03	432.41
	VPC					
g day <sup>-1</sup>	280	-320	266.7	360	93.3	-

SCSI – sugar cane silage with *L. budmeri*; BSCSWI – burnt sugar cane silage without *L. budmeri*; SCSWI – sugar cane silage without *L. budmeri* and BSCI – burnt sugar cane silage with *L. budmeri*. Means followed by the same letter in the row are not significantly different by Tukey's test ( $p > 0.05$ ); CV: Coefficient of variation.

N intake, N excretion in urine and feces, and retained N were not affected ( $p > 0.05$ ) by treatments. Although diets presented similar crude protein content, identified by the lack of difference in nitrogen intake between diets, the type of nitrogen source used may influence the proportion between N excreted via urine and feces. For dairy cows, milk N is also affected by these factors.

In this study, there were differences in the mammary secretion of nitrogen. The values obtained with the supply of diets with fresh sugar cane and with burnt sugar cane silage with *L. buchneri* were higher and similar to each other. However the N intake did not differ, so the nitrogen metabolism should have been changed by the type of roughage used.

Moreover, nitrogen balance, in all forms of expression, was not different ( $p > 0.05$ ) between diets. Importantly, there were no negative means for nitrogen balance, which represents a false indication that the consumption of nitrogen compounds in all diets met the individual protein requirements of the cows.

Meyer (1997) argued that dairy cows in good body condition, but not at their maximum production potential, can divert amino acids to increase milk production rather than to synthesis of tissue protein, if diets meet the energy requirements of animals. In this sense, our results corroborate this statement, in which the negative body weight variation with the diet containing cane sugar silage with *L. buchneri* may be possibly because the additive provide better conditions for utilization of protein and energy in the ruminal environment improving milk production with this diet, which was 12.2 kg day<sup>-1</sup>, higher than 11.6 and 11.3 kg day<sup>-1</sup> for respective silages without inoculum (SCSWI and BSCWI) (SANTOS et al., 2012).

The diet that does not provide enough nutrients to meet the demand of mammary glands to produce milk causes mobilization of body reserves. Thus, the retention of N may be related to the restoration of body protein as a result of increased protein turnover. At the beginning of the experimental periods, the mean weight of animals fed the diet consisting of sugar cane silage with *L. buchneri* was always higher, which probably contributed to no impairment of lactation.

In addition, diets containing sugar cane silage, burnt or not, without *L. buchneri* showed lower milk production (11.6 and 11.3 kg day<sup>-1</sup>, respectively) in relation to fresh sugar cane, sugar cane silage with *L. buchneri* and burnt sugar cane silage with *L. buchneri* (13.5, 12.2, and 12.9 kg day<sup>-1</sup>, respectively), due to lower energy consumption (SANTOS et al., 2012)

associated with lower microbial protein synthesis in the rumen (Table 6).

**Table 6.** Mean daily excretion of purine derivatives and microbial nitrogen compounds by dairy cows in different experimental diets.

Items	Diets					CV(%)
	Fresh	SCSI	BSCSWI	SCSWI	BSCI	
Allantoin in the urine (mmol day <sup>-1</sup> )	310.9 <sup>ab</sup>	353.7 <sup>a</sup>	288.5 <sup>ab</sup>	223.0 <sup>b</sup>	355.7 <sup>a</sup>	18.5
Allantoin in the milk (mmol day <sup>-1</sup> )	3.1	2.3	2.9	3.3	4.0	38.2
Uric acid in the urine (mmol day <sup>-1</sup> )	34.7	32.1	26.4	30.3	39.8	28.6
Total purines (mmol day <sup>-1</sup> )	348.5 <sup>ab</sup>	388.2 <sup>a</sup>	317.7 <sup>ab</sup>	256.4 <sup>b</sup>	399.4 <sup>a</sup>	17.2
Absorbed purines (mmol day <sup>-1</sup> )	365.1 <sup>ab</sup>	411.4 <sup>a</sup>	328.9 <sup>ab</sup>	256.6 <sup>b</sup>	424.6 <sup>ab</sup>	19.4
Microbial nitrogen (g day <sup>-1</sup> )	265.4 <sup>ab</sup>	299.1 <sup>a</sup>	239.1 <sup>ab</sup>	186.5 <sup>b</sup>	308.7 <sup>a</sup>	19.4
Urea-N in the urine (g day <sup>-1</sup> )	18.7	15.9	14.9	24.2	31.6	77.0
NUU (mg kg <sup>-1</sup> BW <sup>0.75</sup> )	192.4	161.3	152.3	243.1	319.2	77.3
Efficiency (g CPmic kg <sup>-1</sup> TDN)	165.6	228.6	208.4	161.9	257.1	24.8
Urine volume (liters)	11.1	8.8	14.6	17.9	10.2	68.3

SCSI – sugar cane silage with *L. buchneri*; BSCSWI – burnt sugar cane silage without *L. buchneri*; SCSWI – sugar cane silage without *L. buchneri* and BSCI – burnt sugar cane silage with *L. buchneri*. NUU – urea-N in urine. Means followed by the same letter in the row are not significantly different by Tukey's test ( $p > 0.05$ ); CV: Coefficient of variation.

Nevertheless, for diets made up of burnt sugar cane silage without *L. buchneri* and sugar cane silage without *L. buchneri*, the positive N balance provided conditions for weight gain.

Mean values of excretion of allantoin in the urine, total purine derivations, absorbed purines and microbial nitrogen compounds were significantly different ( $p < 0.05$ ) (Table 6). Higher values were found in diets composed of silages inoculated with *Lactobacillus buchneri*, which probably improved the fermentation profile, favoring the microbial growth in the rumen.

Schmidt et al. (2007) concluded that the formation profile and ruminal environment were satisfactory and similar between diets containing silages inoculated with *L. buchneri* and those with fresh sugar cane. However, according to the results, it can be stated that the microbial growth in the rumen was also favored even with lower levels of NFC of silages containing *L. buchneri*. For the diet containing sugar cane silage without *L. buchneri*, intestinal flow of nitrogen microbial was similar to those of burnt sugar cane silage without *L. buchneri* and fresh sugarcane, which exhibited the lowest values compared to diets with sugar cane silages, burnt or not, with *L. buchneri*.

Urea-N in urine was not different ( $p > 0.05$ ) between diets studied, with a mean value of 21.04 g day<sup>-1</sup> and 213.65 mg kg<sup>-1</sup> BW<sup>0.75</sup>. Also, microbial efficiency did not differ, with mean value of 204.32 g CPmic kg<sup>-1</sup> TDN, well above 130 g CPmic kg<sup>-1</sup> TDN

presented by NRC (2001) as a mean index of well-balanced diet in protein and energy for ruminal synthesis of microbial protein in lactating cows.

The highest value of urea-N in urine was achieved with the burnt sugar cane silage with *L. buchneri* (Table 6), possibly due to the deficiency of energy by the reduced content of CNF with the ensiling process, combined to the lower intake of DM. As in this treatment there was high microbial synthesis, it can be concluded that amino acids provided by rumen microorganisms and/or diet were used to meet milk production, and also to be transformed into glycodes in response to energy requirement, which cannot be supplied by the diet. This evidence is also supported the retention of N without considerable weight gain allied to higher milk production, which was similar to that of fresh sugarcane. Therefore, for cows fed sugar cane silage with *L. buchneri* and burnt sugar cane silage with *L. buchneri*, the higher availability of amino acids from the use of microbial protein resulted in milk production at levels not significantly different from that of fresh sugar cane.

## Conclusion

The fresh sugar cane increases feeding and rumination efficiency of dry matter compared with other silages. The use of the microbial additive in sugar cane ensiling does not change the behavior of animals in relation to time spent on feeding, ruminating and idling, but alters the total chewing time per kg of dry matter, which is intermediate in relation to fresh sugar cane and sugar cane silage without the additive. The addition of *Lactobacillus buchneri* to sugar cane ensiling process does not modify the nitrogen intake of animals and provides greater ruminal synthesis of microbial nitrogen.

## References

BURGER, P. J.; PEREIRA, J. C.; QUEIROZ, A. C.; SILVA, J. F. C.; VALADARES FILHO, S. C.; CECON, P. R.; CASALI, A. D. P. Comportamento ingestivo em bezerros holandeses alimentados com dietas contendo diferentes níveis de concentrado. **Revista Brasileira de Zootecnia**, v. 29, n. 1, p. 236-242, 2000.

CHEN, X. B.; GOMES, M. J. Estimation of microbial protein supply to sheep and cattle based on urinary excretion of purine derivatives – an overview of technical details. **International Feed Research Unit**. Bucksburnd: Rowett Research Institute, 1992.

FERREIRA, D. A.; GONÇALVES, L. C.; MOLINA, L. R.; CASTRO NETO, A. G.; TOMICH, T. R. Características de fermentação da silagem de cana-

deaçúcar tratada com uréia, zeólita, inoculante bacteriano e inoculante bacteriano/enzimático. **Arquivos Brasileiro de Medicina Veterinária e Zootecnia**, v. 59, n. 2, p. 423-433, 2007.

FREITAS, A.W. P.; PEREIRA, J. C.; ROCHA, F. C.; DETMANN, E.; RIBEIRO, M. D.; COSTA, M. G.; LEONEL, F. P. Características da silagem de cana-de-açúcar tratada com inoculante bacteriano e hidróxido de sódio e acrescida de resíduo da colheita de soja. **Revista Brasileira de Zootecnia**, v. 35, n. 1, p. 48-59, 2006.

ÍTAVO, L. C. V.; VALADARES FILHO, S. C.; VALADARES, R. F. D.; PAULINO, M. F.; ÍTAVO, C. C. B.; MORAES, E. H. B. K. Comparação de indicadores e metodologia de coleta para estimativas de produção fecal e fluxo de digesta em bovinos. **Revista Brasileira de Zootecnia**, v. 31, n. 4, p. 1833-1839, 2002.

MEYER, K. Estimates of genetic parameters for weaning weight of beef cattle accounting for direct-maternal environmental covariances. **Livestock Production Science**, v. 52, n. 3, p. 187-199, 1997.

NRC-National Research Council. **Nutrient requirements of dairy cattle**. 7th ed. Washington, D.C.: National Academy Press, 2001.

PEDROSO, A. F.; NUSSIO, L. G.; LOURES, D. R. S.; PAZIANI, S. F.; RIBEIRO, J. L.; MARI, L. J.; ZOPOLLATTO, M.; SCHMIDT, P.; MATTOS, W. R. S.; HORII, J. Fermentation, losses, and aerobic stability of sugarcane silages treated with chemical and bacterial additives. **Scientia Agricola**, v. 65, n. 6, p. 567-691, 2008.

OJEDA, A.; PARRA, O.; BARCELLS, J. Urinary excretion of purine derivative in *Bos indicus* x *Bos taurus* crossbred cattle. **British Journal of Nutrition**, v. 93, n. 6, p. 821-828, 2005.

SANTOS, A. B.; PEREIRA, M. L. A.; AZEVEDO, S. T.; SIGNORETTI, R. D.; SIQUEIRA, G. R.; MENDONÇA, S. S.; PIRES, A. J. V.; PEREIRA, T. C. J.; ALMEIDA, P. J. P.; RIBEIRO, L. S. O.; PEREIRA, C. A. R. Vacas lactantes alimentadas com silagem de cana-de-açúcar com e sem aditivo bacteriano: consumo, digestibilidade, produção e composição do leite. **Revista Brasileira de Saúde e Produção Animal**, v. 13, n. 3, p. 720-731, 2012.

SCHMIDT, P.; MARI, L. J.; NUSSIO, L. G.; PEDROSO, A. F.; PAZIANI, S. F.; WECHSLER, F. S. Aditivos químicos e biológicos na ensilagem de cana-de-açúcar. 1. Composição química das silagens, ingestão, digestibilidade e comportamento ingestivo. **Revista Brasileira de Zootecnia**, v. 36, n. 5, p. 1666-1675, 2007.

SOUSA, D. P.; CAMPOS, J. M. S.; VALADARES FILHO, S. C.; LANA, R. P.; SEDIYAMA, C. A. Z.; NETO, J. M. Comportamento ingestivo, consumo e digestibilidade de nutrientes, produção e composição do leite de vacas alimentadas com silagem de milho ou cana-de-açúcar com caroço de algodão. **Revista Brasileira de Zootecnia**, v. 38, n. 10, p. 2053-2062, 2009.

UFV-Universidade Federal de Viçosa. **SAEG-Sistema de análise estatísticas e genéticas**. Versão 8.0. Viçosa: UFRV, 1999.

VALADARES, R. F. D.; BRODERICK, G. A.; VALADARES FILHO, S. C.; CLAYTON, M. K. Effect of replacing alfalfa with high moisture corn on ruminal protein synthesis estimated from excretion of total purine derivatives. **Journal of Dairy Science**, v. 82, n. 12, p. 2686-2696, 1999.

VERBIC, J.; CHEN, X. B.; MACLEOD, N. A.; ØRSKOV, E. R. Excretion of purine derivatives by ruminants. Effect of microbial nucleic acid infusion on

purine derivative excretion by steers. **Journal of Agricultural Science**, v. 114, n. 3, p. 243-248, 1990.

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