

# Nutritive value of elephant grass silage added with cottonseed cake in diet for sheep<sup>1</sup>

## Valor nutritivo de silagem de capim-elefante aditivada com torta de algodão em rações para ovinos

Elyane Cristina Borges Dias<sup>2</sup>, Magno José Duarte Cândido<sup>3</sup>, Rafael Nogueira Furtado<sup>4\*</sup>, Roberto Cláudio Fernandes Franco Pompeu<sup>5</sup> and Leane Veras da Silva<sup>6</sup>

**ABSTRACT** - The objective was to evaluate the intake, nutrient digestibility and nitrogen balance in sheep fed diets containing elephant grass (*Pennisetum purpureum* Schum) silage containing 0; 7; 14 and 21% cottonseed cake. Twenty Morada Nova sheep were used in a completely randomized design with four treatments and five replicates. The diets were isoproteic, isoenergetic and isofibrous. There was no effect of cottonseed cake levels on nutrient intake and digestibility coefficient of ether extract. The digestibility coefficients of dry matter, organic matter, crude protein, total carbohydrates and non-fiber carbohydrates presented a linear reduction with the levels of cottonseed cake varying from, respectively, 537.97 to 421.21; 566.54 to 446.42; 595, 38 to 415.62; 544.90 to 421.84 and 697.85 to 521.03, at levels 0 and 21%. The N intake and the urinary N, in the various forms, as well as the fecal N expressed in g day<sup>-1</sup> and g kg<sup>-0.75</sup>, were not influenced by the cottonseed cake levels in the silages. The fecal N presented a quadratic response with a minimum value of 39.09% NI when 3.20% cottonseed cake was added to elephant grass silage. Nitrogen balance showed a quadratic response with a maximum value of 6.50 g day<sup>-1</sup>; 0.79 g kg<sup>-0.75</sup> and 35.07% NI at the levels of 3.25, 5.00 and 4.95% cottonseed cake, respectively. The inclusion of 5% cottonseed cake in elephant grass silage promotes better efficiency in the use of protein sources when aimed at using silage in the composition of a balanced diet.

**Key words:** Additive. Digestibility coefficient. Intake. Nitrogen balance.

**RESUMO** - Objetivou-se avaliar o consumo, digestibilidade dos nutrientes e balanço de nitrogênio em ovinos alimentados com rações contendo silagem de capim-elefante (*Pennisetum purpureum* Schum) contendo 0; 7; 14 e 21% de torta de algodão. Foram utilizados vinte ovinos Morada Nova em delineamento inteiramente casualizado com quatro tratamentos e cinco repetições. As rações foram isoproteicas, isoenergéticas e isofibrosas. Não houve efeito dos níveis de torta de algodão sobre o consumo de nutrientes e coeficiente de digestibilidade do extrato etéreo. Os coeficientes de digestibilidade da matéria seca, matéria orgânica, proteína bruta, carboidratos totais e carboidratos não fibrosos apresentaram redução linear com os níveis de torta de algodão variando respectivamente, de 537,97 a 421,21; 566,54 a 446,42; 595,38 a 415,62; 544,90 a 421,84 e 697,85 a 521,03, nos níveis 0 e 21%. O N consumido e o N urinário, nas diversas formas expressas, assim como o N fecal expresso em g dia<sup>-1</sup> e g kg<sup>-0.75</sup>, não foram influenciados pelos níveis de torta de algodão nas silagens. O N fecal apresentou resposta quadrática com valor mínimo de 39,09% NC quando 3,20% de torta de algodão foi adicionada a silagem de capim-elefante. O balanço de nitrogênio apresentou resposta quadrática com valor máximo de 6,50 g dia<sup>-1</sup>; 0,79 g kg<sup>-0.75</sup> e 35,07% NC nos níveis de 3,25, 5,00 e 4,95% de torta de algodão, respectivamente. A inclusão de 5% de torta de algodão na ensilagem de capim-elefante promove melhor eficiência da utilização das fontes de proteína quando o objetivo for utilizar a silagem na composição de uma ração balanceada.

**Palavras-chave:** Aditivo. Coeficiente de digestibilidade. Consumo. Balanço de nitrogênio.

DOI: 10.5935/1806-6690.20190038

\*Author for correspondence

Received for publication in 31/05/2017; approved in 04/04/2018

<sup>1</sup>Parte da Dissertação de Mestrado do primeiro autor apresentada na Universidade Federal do Ceará, Fortaleza-CE, Brasil

<sup>2</sup>Faculdade de Ciência e Tecnologia, Universidade de Cabo Verde, Praia-Cabo Verde, Brasil, elyaneleida@gmail.com (ORCID ID 0000-0002-1720-2632)

<sup>3</sup>Departamento de Zootecnia, Universidade Federal do Ceará/UFC, Fortaleza-CE, Brasil, magno@ufc.br (ORCID ID 0000-0003-3573-6053)

<sup>4</sup>Pesquisador Visitante do Departamento de Zootecnia/UFC, Fortaleza-CE, Brasil, rafaelnfurtado@yahoo.com.br (ORCID ID 0000-0002-4502-786X)

<sup>5</sup>Embrapa Caprinos e Ovinos, Sobral-CE, Brasil, roberto.pompeu@embrapa.br (ORCID ID 0000-0002-4099-3575)

<sup>6</sup>Programa de Doutorado Integrado em Zootecnia/UFC/UEPB/UFRPE, Fortaleza-CE, Brasil, leaneveras@zootecnista.com.br (ORCID ID 0000-0002-1610-0212)

## INTRODUCTION

The seasonality in forage production in semi-arid regions has been responsible, among other factors, for the low productivity of the herds. Added to this fact is the variation in the prices of traditional protein and energy ingredients used in animal supplementation during the dry period (FURTADO *et al.*, 2014). In this scenario, the use of alternative foods in feed formulation has been gaining increasing prominence in ruminant nutrition (OLIVEIRA *et al.*, 2013).

Among the grasses, elephant grass (*Pennisetum purpureum*, Schum) stands out as an important forage resource for supplementation of the herds during the dry period, being used mainly for silage production. Its versatility is due to high productivity coupled with good nutritional value (RÊGO *et al.*, 2010b). However, like all grasses, the main limiting factor for the production of good quality silage is the low dry matter content at the appropriate time for cutting. This fact leads to the occurrence of secondary fermentations caused mainly by bacteria of the genus *Clostridium*, generating nutrient losses with effluent production.

Moisture-absorbing additives are an important tool to minimize the effects of low DM of tropical grasses at the time of ensiling. Several moisture-absorbing additives have already been tested: cassava meal, rice bran, cocoa meal, corn meal, dehydrated by-product from mango, acerola, cashew, pineapple, among others (ANDRADE *et al.*, 2010; FERREIRA *et al.*, 2009, 2010; ÍTAVO *et al.*, 2010; NEGRÃO *et al.*, 2016; RÊGO *et al.*, 2010a).

Cottonseed cake is one of the by-products from the cotton production chain, which has the fiber for the textile sector as the main product. Its use in animal feed is of importance due to its high protein content and can reduce the participation of soybean meal in feed.

In this sense, the cottonseed cake can contribute to the improvement of the fermentative characteristics of elephant grass silage. Nevertheless, studies evaluating the use of this by-product in grass silage are scarce. As well as evaluations on the intake, digestibility and nitrogen balance in sheep.

This study was conducted with the objective of evaluating the effect of increasing levels of inclusion of cottonseed cake in elephant grass silage on intake, nutrient digestibility and nitrogen balance in sheep.

## MATERIAL AND METHODS

The experiment was carried out at the Núcleo de Ensino e Estudos em Forragicultura NEEF/DZ/CCA/

UFC in the municipality of Fortaleza, State of Ceará. The municipality of Fortaleza is located in the coastal zone at 15.49 m altitude, 3°43'02" South latitude and 38°32'35" West longitude with rainy tropical, Aw' climate, according to Köeppen classification.

Twenty intact male sheep, weighing on average  $17.5 \pm 0.5$  kg ( $\frac{1}{2}$  Morada Nova x  $\frac{1}{2}$  SPRD) with approximately 6 months of age, from the same breeder, were used to evaluate four levels of inclusion (0; 7; 14 and 21%) of cottonseed cake in elephant grass silage (*Pennisetum purpureum* Schum) on a natural matter basis, in a completely randomized design with five replications.

For the preparation of the experimental silages, we used the elephant grass from the grass area already established in the Experimental Farm Curú Valley (FEVC), in Pentecoste, State of Ceará, belonging to the Federal University of Ceará. The grass was cut manually at approximately 70 days of age, after the standardization cut. Then, it was processed in a forage chopper adjusted for cutting to a particle size varying from 1.0 to 2.0 cm, and then mixed to the cottonseed cake in different proportions according to each treatment.

As experimental silos, 210L plastic drums were filled with 126 kg forage each to reach a density of  $600 \text{ kg m}^{-3}$ . After weighing and homogenizing the elephant grass with the cotton seed cake, the material was compacted inside the silo. After filling, the silos were sealed with plastic tarpaulin, fastened with rubber bands.

Experimental diets were prepared based on the recommendations of the National Research Council (2007) according to the nutritional requirement of the category, with the diets being isoproteic, isoenergetic and isofibrous, with different forage: concentrate ratios (40:60, 43:57, 48:52 and 51:49 at levels 0, 7, 14 and 21%, respectively). The proximate composition of the experimental diets is listed in Table 1, the chemical composition of the ingredients in Table 2 and the chemical composition of the total diets in Table 3.

The animals were wormed, received a vitamin complex prior to the onset of the experiment and were housed in metabolic cages containing feeder, drinker and salt, with cages equipped with collectors and separators of feces and urine. The animals were weighed before the start of the experiment and each week.

The experiment lasted 21 days, 14 days for adaptation of the animals to the diets and the experimental environment, and seven days for collection of food, leftovers, feces and urine to determine the voluntary intake, apparent digestibility of nutrients and nitrogen balance. Silage and concentrate were given daily in two equal meals in the morning (at 8:00h) and in the afternoon

**Table 1** - Proximate composition of ingredients in the experimental diets, on a dry matter basis

| Ingredient          | Levels of cottonseed cake (%) |       |       |       |
|---------------------|-------------------------------|-------|-------|-------|
|                     | 0                             | 7     | 14    | 21    |
| Silage              | 40.00                         | 43.00 | 48.00 | 51.00 |
| Corn                | 43.13                         | 45.53 | 46.94 | 46.88 |
| Soybean meal        | 15.04                         | 9.59  | 3.15  | 0.17  |
| Urea                | 0.15                          | 0.14  | 0.13  | 0.12  |
| Ammonium sulfate    | 0.07                          | 0.06  | 0.06  | 0.05  |
| Common salt         | 0.44                          | 0.44  | 0.49  | 0.46  |
| Calcitic limestone  | 0.66                          | 0.61  | 0.49  | 0.49  |
| Dicalcium phosphate | 0.50                          | 0.62  | 0.74  | 0.82  |

**Table 2** - Chemical composition of ingredients and the silages

| Nutrient (g kg <sup>-1</sup> ) | Elephant grass | Cottonseed cake | Levels of cottonseed cake (%) |        |        |        |
|--------------------------------|----------------|-----------------|-------------------------------|--------|--------|--------|
|                                |                |                 | 0                             | 7      | 14     | 21     |
| Dry matter                     | 176.00         | 882.80          | 183.37                        | 244.00 | 342.60 | 422.70 |
| Organic matter                 | 902.70         | 951.10          | 879.10                        | 895.30 | 907.90 | 926.20 |
| Crude protein                  | 55.80          | 357.30          | 48.50                         | 98.30  | 163.80 | 176.30 |
| Ether extract                  | 15.90          | 90.10           | 23.90                         | 33.40  | 46.00  | 57.00  |
| Neutral detergent fiber        | 717.10         | 498.40          | 741.10                        | 683.60 | 678.40 | 640.80 |
| Acid detergent fiber           | 445.70         | 358.40          | 498.50                        | 484.80 | 465.60 | 431.00 |
| Cellulose                      | 346.90         | 298.60          | 346.50                        | 327.00 | 325.00 | 287.60 |
| Lignin                         | 50.11          | 50.53           | 58.80                         | 48.30  | 50.30  | 50.50  |
| Total carbohydrates            | 831.00         | 503.70          | 806.70                        | 763.60 | 698.10 | 693.10 |
| Non-fiber carbohydrates        | 142.20         | 17.60           | 82.70                         | 106.80 | 48.10  | 85.30  |
| Total digestible nutrients     | 520.19         | 701.88          | 475.59                        | 541.48 | 566.45 | 605.50 |

**Table 3** - Chemical composition of total diets

| Nutrient (g kg <sup>-1</sup> ) | Levels of cottonseed cake (%) |        |        |        |
|--------------------------------|-------------------------------|--------|--------|--------|
|                                | 0                             | 7      | 14     | 21     |
| Dry matter                     | 583.35                        | 592.61 | 615.29 | 644.33 |
| Organic matter                 | 835.00                        | 843.37 | 859.02 | 872.64 |
| Crude protein                  | 158.54                        | 152.91 | 143.88 | 143.62 |
| Ether extract                  | 39.50                         | 47.76  | 51.72  | 60.82  |
| Neutral detergent fiber        | 397.24                        | 370.33 | 394.43 | 389.63 |
| Acid detergent fiber           | 243.26                        | 247.22 | 252.76 | 241.62 |
| Cellulose                      | 165.84                        | 233.61 | 236.52 | 48.07  |
| Lignin                         | 28.26                         | 21.46  | 42.97  | 77.28  |
| Total carbohydrates            | 726.96                        | 725.01 | 732.58 | 729.56 |
| Non-fiber carbohydrates        | 354.02                        | 374.70 | 361.56 | 362.44 |
| Total digestible nutrients     | 703.83                        | 722.48 | 718.36 | 732.99 |

(at 16:00h), the amount offered being calculated daily from the previous consumption so as to allow leftovers of approximately 15% of the food offered.

During the collection period, intake and digestibility determinations were performed by daily weighing and sampling 10% of the food supplied, leftovers and feces from each animal. Samples were packed in plastic bags, identified and stored at -10 °C. At the end of the experiment, the samples for each animal were thawed and homogenized, and a sample of approximately 300 g was taken, weighed and pre-dried in a forced ventilation oven at 55 °C to constant weight to determine the pre-dried matter. They were then ground in a knife mill with a 1mm sieve and placed in plastic containers for further analysis.

The determination of dry matter (DM), organic matter (OM), ether extract, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), neutral detergent fiber corrected for ash and protein (NDFap) and neutral detergent insoluble nitrogen (% of total N), acid detergent insoluble nitrogen (% of total N), followed methodologies compiled in Silva and Queiroz (2002). Total carbohydrates (TC) were obtained according to Sniffen *et al.* (1992), non-fiber carbohydrates (NFC) were determined according to Hall (2000) and the total digestible nutrients content (NDT) was obtained according to Weiss (1999).

With the values obtained from the chemical analyses of the diet supplied and the leftovers, we determined the consumption of DM, OM, EE, CP, NDF, ADF, TC and NFC, which were expressed as percentage of body weight (% BW) and gram per kg metabolic weight ( $\text{g kg}^{-0.75}$ ). The digestibility coefficient (DC) of nutrients was determined according to equation 1.

$$DC (\%) = [IN (g) - NEF (g) / IN (g)] \times 100 \quad (\text{Eq.1})$$

Where: NI = ingested nutrient (g) and NEF = nutrient excreted in feces (g).

On the 21<sup>st</sup> day, urine samples were collected in plastic containers over 24 hours using funnel collectors in the metabolic cages, which poured the urine into plastic containers containing 20 mL of 1: 1 hydrochloric acid solution. After collection, the containers containing urine were duly weighed to determine the total volume produced, homogenized and gauze-filtered. Subsequently, aliquots of approximately 10% of the total volume were taken, properly identified and stored at -5 °C, to determine the nitrogen contained in urine according to the methodology described in Silva and Queiroz (2002). Nitrogen balance (NB) was determined by the difference between ingested and excreted nitrogen in feces and urine.

Data were tested by analysis of variance and regression. The choice of the models was based on the significance of the linear and quadratic coefficients, using the Student's t-test, at the 5% probability level and in the determination coefficient using the SAS computational package (SAS INSTITUTE, 2003).

## RESULTS AND DISCUSSION

Dry matter intake (DMI), organic matter intake (OMI), crude protein intake (CPI) and ether extract intake (EEI) were not influenced by the levels of inclusion of cottonseed cake in elephant grass silages when expressed in grams ( $\text{g day}^{-1}$ ), grams per kg of metabolic weight ( $\text{g kg}^{-0.75}$ ) and percentage of body weight (% BW), with mean values presented in Table 4.

DMI increases with increasing DM content of the silage (VAN SOEST, 1994). In this sense, it was expected an increase in DMI with the increment of the cottonseed cake levels due to the increase in DM content promoted by this additive due to the differences between DM contents of elephant grass ( $176.00 \text{ g kg}^{-1}$ ) and cottonseed cake ( $882.80 \text{ g kg}^{-1}$ ). However, the lack of effect is justified by the differences in the forage: concentrate ratio between the diets (40:60, 43:57, 48:52 and 51:49 at the 0, 7, 14

**Table 4** - Nutrient intake in sheep fed diets containing elephant grass silage added with different levels of cottonseed cake

| Variable                                 | Regression equation        | R <sup>2</sup> | CV (%) |
|--|----------------------------|----------------|--------|
| Dry matter (% BW)                        | $\hat{Y} = 2.83 \pm 0.38$  | -              | 14.30  |
| Dry matter ( $\text{g kg}^{-0.75}$ )     | $\hat{Y} = 57.54 \pm 7.49$ | -              | 13.84  |
| Organic matter (% BW)                    | $\hat{Y} = 2.65 \pm 0.36$  | -              | 14.32  |
| Organic matter ( $\text{g kg}^{-0.75}$ ) | $\hat{Y} = 53.87 \pm 7.07$ | -              | 13.85  |
| Crude protein (% BW)                     | $\hat{Y} = 0.44 \pm 0.06$  | -              | 13.53  |
| Crude protein ( $\text{g kg}^{-0.75}$ )  | $\hat{Y} = 8.83 \pm 1.13$  | -              | 13.21  |
| Ether extract (% BW)                     | $\hat{Y} = 0.15 \pm 0.02$  | -              | 14.03  |
| Ether extract ( $\text{g kg}^{-0.75}$ )  | $\hat{Y} = 2.96 \pm 0.42$  | -              | 14.28  |

and 21% levels, respectively) that were necessary for the proper balancing and culminated in similar supply of DM in the total diet.

DMI expressed in %BW and  $\text{g kg}^{-0.75}$  was higher than reported by Teles *et al.* (2010), who found that DMI of 2.22% BW and  $46.38 \text{ g kg}^{-0.75}$  when sheep fed exclusively elephant grass silage containing different levels of dehydrated cashew by-product. However, Carvalho Júnior *et al.* (2009) verified DMI in sheep of 3.59% BW and  $79.8 \text{ g kg}^{-0.75}$  when using elephant grass silage with 15% coffee husk and 3.85% BW and  $86.9 \text{ g kg}^{-0.75}$  when using elephant grass silage with 15% cassava meal, both supplemented with concentrate. These results were higher than those obtained in the present study, which presented mean values of 2.83% BW and  $57.54 \text{ g kg}^{-0.75}$  and showed higher potential of coffee husk and cassava meal as additives in elephant grass silage when supplemented.

The lack of effect of cottonseed cake levels in elephant grass silages on OMI, CPI and EEI is because diets are isoprotein and isoenergetic, associated with high correlation between DMI and OMI ( $r = 0.99$ ,  $p < 0.0001$ ), DMI and CPI ( $r = 0.85$ ,  $p < 0.0001$ ) and DMI and EEI ( $r = 0.93$ ,  $p < 0.0001$ ). It should be noted that the inclusion of cottonseed cake increased the contents of organic matter (OM), crude protein (CP) and ether extract (EE) of the silages. Results corroborated by Viana *et al.* (2013), who also verified increase in CP and EE contents when evaluating the chemical composition and fermentation profile of elephant grass silages supplemented with cottonseed cake. However, the balancing of the total diets in the present study eliminated the effects of the chemical composition of silages on the intake. The CPI expressed in % BW was not enough to meet the requirements proposed by the National Research Council (2007) for lambs finished early, with eight months, weighing 20 kg BW, ranging from 0.52 to 0.56% BW as a result of the proportion of CP of the diet that is degradable in the rumen.

There was no effect of cottonseed cake levels on the intake of neutral detergent fiber (NDFI), acid detergent fiber (ADFI), total carbohydrates (TCI) and non-fiber carbohydrates (NFCI), expressed as % BW and  $\text{g kg}^{-0.75}$  (Table 5). This response was due to the balance of the diets, since they were formulated to be isoenergetic and isofibrous, reducing possible variations in the contents of neutral and acid detergent fibers, total and non-fiber carbohydrates, resulting from the inclusion of cottonseed cake in elephant grass silages. The average NDFI of 1.07% BW is within the limits of 0.8 to 1.2% BW recommended by Van Soest (1994) for a good ruminal functioning.

The dry matter digestibility coefficient (DMDC) decreased with the increasing levels of cottonseed cake in elephant grass silages (Table 6). Each percentage point of added cottonseed cake reduced DMDC by  $5.56 \text{ g kg}^{-1}$ . This reduction can be attributed to the increase in silage participation in the total diet with consequent reduction of the concentrate, increasing the rate of passage and consequently reducing the DMDC. Similar results were reported by Medeiros *et al.* (2007), when evaluating castrated sheep fed total diets containing different ratios of concentrate. The authors verified a DMDC increase of  $2.66 \text{ g kg}^{-1} \text{ DM}$  for each percentage point of concentrate added to the feeds.

Although organic matter content of the diets and the OMI by the animals in the different levels of inclusion of the cottonseed cake were not varied with the diets, there was a linear reduction of the organic matter digestibility coefficient (OMDC) with the levels of cottonseed cake in the silages. This result is attributed to a high correlation between OMDC and DMDC ( $r = 0.99$ ;  $p < 0.0001$ ) and suggests that the factors responsible for DMDC reduction also impacted the OMDC reduction.

Diets with higher levels of cottonseed cake had a reduction in crude protein digestibility coefficients (CPDC), with decreases of  $8.56 \text{ g kg}^{-1} \text{ DM}$  for each increase of one percentage point of cottonseed cake in

**Table 5** - Fiber and carbohydrate intake in sheep fed diets containing elephant grass silage supplemented with different levels of cottonseed cake

| Variable  | Regression equation        | R <sup>2</sup> | CV (%) |
|---|----------------------------|----------------|--------|
| Neutral detergent fiber (% BW)                    | $\hat{Y} = 1.07 \pm 0.15$  | -              | 15.87  |
| Neutral detergent fiber ( $\text{g kg}^{-0.75}$ ) | $\hat{Y} = 21.78 \pm 3.02$ | -              | 15.48  |
| Acid detergent fiber (% BW)                       | $\hat{Y} = 0.63 \pm 0.10$  | -              | 16.43  |
| Acid detergent fiber ( $\text{g kg}^{-0.75}$ )    | $\hat{Y} = 12.77 \pm 1.92$ | -              | 16.05  |
| Total carbohydrates (% BW)                        | $\hat{Y} = 2.04 \pm 0.28$  | -              | 14.5   |
| Total carbohydrates ( $\text{g kg}^{-0.75}$ )     | $\hat{Y} = 41.54 \pm 5.60$ | -              | 14.01  |
| Non-fiber carbohydrates (% BW)                    | $\hat{Y} = 1.05 \pm 0.16$  | -              | 15.94  |
| Non-fiber carbohydrates ( $\text{g kg}^{-0.75}$ ) | $\hat{Y} = 21.29 \pm 3.16$ | -              | 15.4   |

the silages. This result is due to the lower participation of soybean meal in the total diet with the higher levels of cottonseed cake in the silages. Moreover, in the higher levels of cottonseed cake there was an increase in the  $\text{NH}_3\text{-N}$  content of the silages (unpublished data) that impacted the proportion of soluble protein remaining for ruminal digestion and consequently for CPDC. The similar contribution of CP of the diets, with similar CPI, allows to evidence possible differences in the quality of the nitrogen source (GERON *et al.*, 2015), which in the present study showed that the soybean meal had higher quality as a nitrogen source composing the concentrate compared to the cottonseed cake used as an additive in elephant grass silage.

The ether extract digestibility coefficient (EEDC) was not affected by the different levels of cottonseed cake, with a mean value of  $735.92 \text{ g kg}^{-1} \text{ DM}$ . Lower values were observed by Teles *et al.* (2010), who also found no effect of EEDC when added dehydrated cashew peduncle in elephant grass silage with mean values of  $589.70 \text{ g kg}^{-1} \text{ DM}$ .

The neutral detergent fiber digestibility coefficient (NDFDC) showed quadratic response with the increment of the cottonseed cake in the silages. The lowest NDFDC ( $351.68 \text{ g kg}^{-1} \text{ DM}$ ) was obtained when 17.36% cottonseed cake was added. The reduction in NDFDC to this level is attributed to the greater participation of the cottonseed cake in the silages and the silages in the total diet. From the inclusion of 16.15% cottonseed cake, there was an increase in hemicelluloses content and a decrease in cellulose content (unpublished data), and the interaction of these two components was responsible for the increase in the NDFDC. The lignin together with the cellulose and hemicelluloses represent the NDF fraction and when it is present in large quantities, it is more likely to form complexes with hemicellulose and cellulose, making them unavailable for microbial growth (VAN SOEST, 1994) with consequent reduction in NDFDC.

There was a linear reduction in total carbohydrates (TCDC) and non-fiber carbohydrates (NFCDC) digestibility coefficients with addition of the cottonseed cake. TCDC ranged from  $544.90$  to  $421.84 \text{ g kg}^{-1}$  and NFCDC ranged from  $697.85$  to  $521.03 \text{ g kg}^{-1}$  at levels 0 and 21%, respectively. This shows the negative effect of inclusion of cottonseed cake on elephant grass silage, since NFC are easy to ferment, thus ensuring a higher energy intake in the ruminal environment (NATIONAL RESEARCH COUNCIL, 2001).

The nitrogen (N) intake and fecal N, expressed as  $\text{g}^{-1}$  and  $\text{g kg}^{-0.75}$ , and urinary nitrogen expressed as  $\text{g day}^{-1}$ ,  $\text{g kg}^{-0.75}$  and percentage of N intake (% NI) were not influenced by the inclusion levels of cottonseed cake (Table 7).

There was a quadratic response for fecal N expressed as % NI as a function of cottonseed cake levels. The fecal N presented a minimum value of 39.09% NI when 3.20% cottonseed cake was added to the silage, rising again from that value. This fact was due to the reduction of the CPDC in the higher levels of cottonseed cake, with consequent increase in the excretion of N in the feces. Even at the level of cottonseed cake that promoted lower fecal N value (3.20% cottonseed cake), there was a greater loss of N through the fecal route compared to the urinary route, which presented an average value of 28.41% NI. Results similar to the present study were reported by Moreno *et al.* (2010), who evaluated the nitrogen balance in lambs fed corn or sugar cane silage and two concentrate levels and verified losses of fecal and urinary N of 40.74 and 22.20% of N intake, respectively.

The nitrogen balance (NB) expressed in  $\text{g day}^{-1}$ ,  $\text{g kg}^{-0.75}$  and % NI presented a quadratic response with a maximum value of  $6.50 \text{ g day}^{-1}$ ,  $0.79 \text{ g kg}^{-0.75}$  and 35.07% NI at the levels of 3.25, 5.00 and 4.95% cottonseed cake, respectively. The maximum NB obtained with the inclusion

**Table 6** - Nutrient digestibility coefficient in sheep fed diets containing elephant grass silage supplemented with different levels of cottonseed cake

| Variable                                       | Regression equation                                   | R <sup>2</sup> | CV (%) |
|--|---|----------------|--------|
| Dry matter ( $\text{g kg}^{-1}$ )              | $\hat{Y} = 537.97 - 5.56\text{TA}$                    | 0.53           | 8.93   |
| Organic matter ( $\text{g kg}^{-1}$ )          | $\hat{Y} = 566.54 - 5.72\text{TA}$                    | 0.51           | 9.18   |
| Crude protein ( $\text{g kg}^{-1}$ )           | $\hat{Y} = 595.38 - 8.56\text{TA}$                    | 0.80           | 6.69   |
| Ether extract ( $\text{g kg}^{-1}$ )           | $\hat{Y} = 735.92 \pm 31.39$                          | -              | 3.95   |
| Neutral detergent fiber ( $\text{g kg}^{-1}$ ) | $\hat{Y} = 469.20 - 13.54\text{TA} + 0.39\text{TA}^2$ | 0.33           | 18.4   |
| Total carbohydrates ( $\text{g kg}^{-1}$ )     | $\hat{Y} = 544.90 - 5.86\text{TA}$                    | 0.46           | 10.8   |
| Non-fiber carbohydrates ( $\text{g kg}^{-1}$ ) | $\hat{Y} = 697.85 - 8.42\text{TA}$                    | 0.78           | 6.07   |

**Table 7** - Balance of nitrogen compounds in sheep fed diets containing elephantgrass silage supplemented with different levels of cottonseed cake

| Variable                           | Regression equation                     | R <sup>2</sup> | CV (%) | P      |
|------------------------------------|---|----------------|--------|--------|
| N intake (g day <sup>-1</sup> )    | $\hat{Y} = 17.90 \pm 2.03$              | -              | 10.87  | NS     |
| N intake (g kg <sup>-0.75</sup> )  | $\hat{Y} = 2.13 \pm 0.26$               | -              | 11.70  | NS     |
| Fecal N (g dia <sup>-1</sup> )     | $\hat{Y} = 8.51 \pm 2.82$               | -              | 32.78  | NS     |
| Fecal N (g kg <sup>-0.75</sup> )   | $\hat{Y} = 1.01 \pm 0.33$               | -              | 33.12  | NS     |
| Fecal N (% NI)                     | $\hat{Y} = 39.83 - 0.461TA + 0.072TA^2$ | 0.42           | 24.18  | 0.0098 |
| Urinary N (g day <sup>-1</sup> )   | $\hat{Y} = 4.99 \pm 1.36$               | -              | 31.82  | NS     |
| Urinary N (g kg <sup>-0.75</sup> ) | $\hat{Y} = 0.59 \pm 0.16$               | -              | 30.71  | NS     |
| Urinary N (% NI)                   | $\hat{Y} = 28.41 \pm 8.86$              | -              | 36.08  | NS     |
| N balance (g day <sup>-1</sup> )   | $\hat{Y} = 6.29 + 0.13TA - 0.02TA^2$    | 0.57           | 49.29  | 0.0008 |
| N balance (g kg <sup>-0.75</sup> ) | $\hat{Y} = 0.74 + 0.02TA - 0.002TA^2$   | 0.55           | 50.06  | 0.0011 |
| N balance (% NI)                   | $\hat{Y} = 32.37 + 1.09TA - 0.11TA^2$   | 0.54           | 47.69  | 0.0012 |

of approximately 5% cottonseed cake can be attributed to lower fecal N loss when 3.20% cottonseed cake was added to elephant grass silage. The high correlation between fecal N and NB expressed in g day<sup>-1</sup>, g kg<sup>-0.75</sup> and % NI ( $r = 0.76$ ,  $p < 0.0001$ ;  $r = 0.76$ ,  $p = 0.0001$ , and  $r = 0.84$ ,  $p < 0.0001$ , respectively) corroborate the negative impact of fecal N expressed as % NI on NB, since the higher retrieval of N in feces represents lower NB. In association, the reductions in CPDC and NFCDC with the levels of cottonseed cake in the silages, reduced the availability of protein and digestible energy, impairing the synchronization of these nutrients, maximizing the microbial synthesis and consequently increasing the nitrogen losses by the fecal route. Thus, protein metabolism of sheep was impaired in levels of addition of cottonseed cake in elephant grass silage higher than 5%, which reflected a reduction in the efficiency of the protein fraction of the diet.

## CONCLUSIONS

1. The addition of cottonseed cake in elephant grass silage does not alter nutrient intake, but negatively affects nutrient digestibility coefficients;
2. The inclusion of 5% cottonseed cake in elephant grass silage promotes better efficiency in the use of protein sources when the objective is to use silage in the composition of a balanced diet.

## REFERENCES

ANDRADE, I. V. O. *et al.* Perdas, características fermentativas e valor nutritivo da silagem de capim-elefante contendo

subprodutos agrícolas. **Revista Brasileira de Zootecnia**, v. 39, n. 12, p. 2578-2588, 2010.

CARVALHO JUNIOR, J. N. *et al.* Desempenho de ovinos mantidos com dietas com capim-elefante ensilado com diferentes aditivos. **Revista Brasileira de Zootecnia**, v. 38, n. 6, p. 994-1000, 2009.

FERREIRA, A. C. H. *et al.* Consumo e digestibilidade de silagens de capim-elefante com diferentes níveis de subproduto da agroindústria da acerola. **Revista Ciência Agrônômica**, v. 41, n. 4, p. 693-701, 2010.

FERREIRA, A.C. H. *et al.* Desempenho produtivo de ovinos alimentados com silagens de capim-elefante contendo subprodutos do processamento de frutas. **Revista Ciência Agrônômica**, v. 40, n. 2, p. 315-322, 2009.

FURTADO, R. N. *et al.* Balanço de nitrogênio e avaliação ruminal em ovinos machos e fêmeas alimentados com rações contendo torta de mamona sob diferentes tratamentos. **Semina: Ciências Agrárias**, v. 35, n. 6, p. 3237-3248, 2014.

GERON, L. J. V. *et al.* Balanço de nitrogênio em cordeiros alimentados com rações contendo diferentes teores de concentrado. **Semina: Ciências Agrárias**, v. 36, n. 3, p. 1609-1622, 2015.

HALL, M. B. **Neutral detergent-soluble carbohydrates: nutritional relevance and analysis.** Gainesville: University of Florida, 2000. 76 p.

ITAVO, L. C. V. *et al.* Composição química e parâmetros fermentativos de silagens de capim-elefante e cana-de-açúcar tratadas com aditivos. **Revista Brasileira de Saúde e Produção Animal**, v. 11, n. 3, p. 606-617, 2010.

MEDEIROS, G. R. *et al.* Efeito dos níveis de concentrado sobre o desempenho de ovinos Morada Nova em confinamento. **Revista Brasileira de Zootecnia**, v. 36, n. 4, p. 1162-1171, 2007. Suplemento.

- MORENO, G. M. B. *et al.* Desempenho, digestibilidade e balanço de nitrogênio em cordeiros alimentados com silagem de milho ou cana-de-açúcar e dois níveis de concentrado. **Revista Brasileira de Zootecnia**, v. 39, n. 4, p. 853-860, 2010.
- NATIONAL RESEARCH COUNCIL. **Nutrient requirements of small ruminants: sheep, goats, cervides, and world camelides**. Washington: National Academy Press, 2007. 362 p.
- NATIONAL RESEARCH COUNCIL. **Nutrient requirements of the dairy cattle**. 7. ed. Washington: National Academy Press, 2001. 363 p.
- NEGRÃO, F. M. *et al.* Perdas, perfil fermentativo e composição química das silagens de capim *Brachiaria decumbens* com inclusão de farelo de arroz. **Revista Brasileira de Saúde e Produção Animal**, v. 17, n. 1, p. 13-25, 2016.
- OLIVEIRA, R. L. *et al.* Alimentos alternativos na dieta de ruminantes. **Revista Científica de Produção Animal**, v. 15, n. 2, p. 141-160, 2013.
- RÊGO, M. M. T. *et al.* Intake, nutrients digestibility and nitrogen balance of elephant grass silages with mango by-product addition. **Revista Brasileira de Zootecnia**, v. 39, n. 1, p. 74-80, 2010a.
- RÊGO, M. M. T. *et al.* Nutritional evaluation of elephant-grass silages with byproduct of annatto. **Revista Brasileira de Zootecnia**, v. 39, n. 10, p. 2281-2287, 2010b.
- SAS INSTITUTE. **SAS system for windows**. Version 9.1. Cary, 2003.
- SILVA, D. J.; QUEIROZ, C. **Análise de alimentos: métodos químicos e biológicos**. 3. ed. Viçosa, MG: Editora UFV, 2002. 235 p.
- SNIFFEN, C. J. *et al.* A net carbohydrate and protein system for evaluation cattle diets II. Carbohydrate and protein availability. **Journal of Animal Science**, v. 70, n. 11, p. 3562-3577, 1992.
- TELES, M. M. *et al.* Consumo, digestibilidade de nutrientes e balanço de nitrogênio da silagem de capim-elefante com adição de pedúnculo de caju desidratado. **Ciência Rural**, v. 40, n. 2, p. 427-433, 2010.
- VAN SOEST, P. J. **Nutritional ecology of the ruminant**. 2. ed. New York: Cornell University Press, 1994. 476 p.
- VIANA, P. T. *et al.* Losses and nutritional value of elephant grass silage with inclusion levels of cottonseed meal. **Acta Scientiarum. Animal Sciences**, v. 35, n. 2, p. 139-144, 2013.
- WEISS, W.P. Energy prediction equations for ruminant feeds. *In: CORNELL NUTRITION CONFERENCE FOR FEED MANUFACTURERS*, 61., 1999, Ithaca. **Proceedings...** Ithaca: Cornell University, 1999. p. 176-185.



This is an open-access article distributed under the terms of the Creative Commons Attribution License