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Elephant grass cv. BRS capiaçu silage with inclusion of different proportions of silk cotton

Silagem de capim-elefante cv. BRS capiaçu com inclusão de diferentes proporções do algodão-de-seda

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Highlights

Silk cotton increased the crude protein content of BRS capiaçu silage.
Silk cotton improved the energy content of grass silage.
BRS capiaçu silage with silk cotton has potential for feeding ruminants.

Abstract

The objective was to evaluate the qualitative and nutritional aspects of elephant grass cv. BRS capiaçu silage with increasing proportions of silk cotton. A completely randomized design was used with five (0, 15, 30, 45, and 60% on natural matter basis) different inclusion proportions of silk cotton to elephant grass silage and four repetitions. Silages were prepared in PVC minisilos and remained ensiled for 42 days. The inclusion of silk cotton resulted in a linear reduction in dry matter (DM), neutral detergent fiber (NDF), acid detergent fiber (ADF), cellulose, and indigestible neutral detergent fiber (iNDF) contents at 0.06, 0.30, 0.23, 0.20, and 0.09 percentage points for each 1% inclusion, respectively. There was an increasing linear effect of crude protein (CP) and total digestible nutrients (TDN), with an increase of 39.32 and 20.89% from the lowest to the highest inclusion of silk cotton to elephant grass cv. BRS capiaçu silage. Values of pH, gas losses (GL), effluent losses (EL), dry matter recovery (DMR), hemicellulose, and lignin were not influenced by the evaluated inclusion levels, showing estimated averages of 3.87, 1, 42%, 11.55kg t-1 natural matter, 97.36%, 19.30%, and 7.20%, respectively. An increasing linear effect was found for fraction a, c, effective degradability (ED)

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(2% and 5%) dry matter, with an increment of 0.05, 0.0005, and 0.11 percent points, respectively. There was a quadratic effect for fraction b, potential degradability (PD), ED (2%), with minimum points of 23.72%, 50.52%, (at 45% inclusion level) 39.69%, (at 15% inclusion level) respectively. The indigestible fraction (IF) had a quadratic effect with a maximum of 49.48% at the 45% inclusion level. The colonization time (CT) linearly reduced by 0.09 percentage points for each 1% inclusion evaluated. According to the parameters evaluated, the inclusion of up to 60% silk cotton improved nutritional aspects and nutritional value of elephant grass cv. BRS capiaçu silage.
Key words: *Calotropis procera.* Chemical composition. *Penisetum purpureum* Schum. Crude protein. Nutritional value.

Resumo

Objetivou-se avaliar aspectos qualitativos e nutricionais da silagem de capim-elefante cv. BRS capiaçu com proporções crescentes de algodão-de-seda. Foi utilizado o delineamento inteiramente casualizado com cinco (0, 15; 30; 45; 60% na base da matéria natural) diferentes proporções de inclusão do algodão-de-seda à silagem de capim-elefante cv. BRS Capiaçu e quatro repetições. As silagens foram confeccionadas em silos de PVC e permaneceram ensiladas por 42 dias. A inclusão do algodão-de-seda resultou em redução linear nos teores de matéria seca (MS), fibra em detergente neutro (FDN), fibra em detergente ácido (FDA), celulose e fibra em detergente neutro indigestível (FDNi) em 0,06, 0,30, 0,23, 0,20 e 0,09 pontos percentuais para cada 1% de inclusão, respectivamente. Houve efeito linear crescente da proteína bruta (PB) e nutrientes digestíveis totais (NDT), com incremento de 39,32 e 20,89% da menor à maior proporção de inclusão do algodão-de-seda à silagem do capim-elefante cv. BRS Capiaçu. O potencial hidrogeniônico (pH), perdas por gases (PG), perdas por efluentes (PE), recuperação de matéria seca (RMS), hemicelulose e lignina não foram influenciados pelos níveis de inclusão avaliados apresentando médias estimadas de 3,87, 1,42%, 11,55kg t-1 de MN, 97,36%, 19,30% e 7,20%, respectivamente. Foi observado efeito linear crescente para fração a, c, degradabilidade efetiva (DE) (2% e 5%) da matéria seca, com incremento de 0,05, 0,0005, e 0,11 pontos percentuais, respectivamente. Houve efeito quadrático para fração b, degradabilidade potencial (DP), DE (2%), com pontos de mínima de 23,72%, 50,52%, (no nível de 45 % de inclusão) 39,69%, (no nível de 15 % de inclusão) respectivamente. A fração indigestível (FI) apresentou efeito quadrático com máxima de 49,48% no nível de 45% de inclusão. O tempo de colonização (TC) reduziu linearmente em 0,09 pontos percentuais para cada 1% de inclusão avaliada. De acordo com os parâmetros avaliados, a inclusão de até 60% de algodão-de-seda à silagem de capim-elefante cv. BRS capiaçu favorece os aspectos nutricionais e o valor nutritivo das mesmas.
Palavras-chave: *Calotropis procera.* Composição química. *Penisetum purpureum* Schum. Proteína bruta. Valor nutritivo.

Introduction

Cattle raising is one of the most economically important activities in Brazil. Brazilian cattle raising stands out on the world stage, with the largest commercial herd in the world, with 213.7 million heads, representing 14.8% world herd, according to the Associação Brasileira das Indústrias Exportadoras de Carnes [ABIEC] (2020). A significant portion of this herd is raised on pasture, however, under these circumstances, ruminant production is

limited by the seasonality of forage production, caused by low rainfall, especially in semi-arid climate conditions, limiting the availability of feed to meet the animal nutrition requirements throughout the year.

Feed preservation as silage is a viable alternative in the search for a balance in availability of food for animals in periods of scarcity, as this technique allows for the preservation of surplus forage production during the rainy season, in addition to allowing the use of native forage for animal feeding.

Elephant grass cv. BRS capiaçu (*Pennisetum purpureum* Schum.), developed by the Brazilian Agricultural Research Corporation - National Center for Research in Dairy Cattle (EMBRAPA-CNPGL) as an alternative for roughage supplementation, has a high production potential, reaching an average of 50 t ha^{-1} $year^{-1}$ dry matter and good nutritional quality, which reduces production costs (A. V. Pereira et al., 2016). Also according to these authors, the cultivar stands out from other elephant grass cultivars because it has tolerance to water stress and resistance to lodging, which facilitates mechanical harvesting, and can be used to produce good quality silage.

According to Monção et al. (2019), for BRS capiaçu grass silage, harvest is recommended at an age between 90 and 120 days (presenting 9.75 and 7.88% crude protein, respectively), or between 3 and 4.2 meters in height, period where a better relationship between nutritional composition and dry matter content is found.

Silk cotton (*Calotropis procera*) is a plant native to semi-arid climate regions and has been studied as a nutritional alternative for ruminants. Belonging to the family Asclepiadaceae, it is adapted to semi-arid and arid regions, it develops satisfactorily in degraded soils and in places with low rainfall (Andrade, M., Silva, Andrade, Medeiros, & Pinto, 2005). This species also stands out for its permanence capacity of the leaves and vigorous regrowth, high availability of seeds and excellent germination; good digestibility and dry matter intake. Andrade et al. (2008) obtained for fresh *C. procera* the following chemical composition values: 23.25 % dry matter (DM); 19.44% crude protein (CP); 42.17% neutral detergent fiber (NDF); 28.41% acid detergent fiber (ADF); 25.22 % non-fiber carbohydrates (NFC) and 65.5% total carbohydrates. Furthermore, studies such as Lima et al. (2006) demonstrate the possible antifungal capacity of silk cotton, acting in a favorable way to the silage preservation process.

The presence of toxic substances in the latex produced by silk cotton limits its supply in fresh form for animal consumption. Mello et al. (2001) evaluating the composition of the latex produced by silk cotton, detected glycosidic, flavonic, cardiotonic and steroid substances in leaves of the plant. These authors also state that after chopping and drying the leaves, they lose some of these active substances by volatilization, which makes them less toxic, favoring animal intake.

Thus, the possibility of including fresh silk cotton in capiaçu silage can improve the nutritional value of conventionally used feed silages, preserve alternative forage sources for periods of food shortage, maintaining the ideal quality parameters of silages, and thus increase feed availability and, consequently, animal performance, with favorable effects throughout the production chain.

With this, the objective was to evaluate the effect of different levels of inclusion of silk cotton in capiaçu silage on the qualitative and nutritional aspects.

Material and Methods

The experiment was conducted at the Department of Agricultural Sciences at the State University of Montes Claros (Unimontes), Campus Janaúba, northern Minas Gerais. According to Koppen and Geiger (1928), the climate in the region is Aw (savannah climate with dry winter). It presents an average annual rainfall of 700 mm, with an average temperature of 28°C and relative humidity around 65%.

The treatments consisted of five levels of inclusion of silk cotton (0, 15, 30, 45 and 60%) in elephant grass cv. BRS capiaçu silage, and each treatment contained 10% corn meal ground in a 3mm sieve, as a moisture sequestering, in order to prevent a significant decline in the DM content of the silages to be evaluated. The treatments consisted of: T1 (90% BRS capiaçu + 10% corn meal); T2 (75% BRS capiaçu + 15% silk cotton + 10% corn meal); T3 (60% BRS capiaçu + 30% silk cotton + 10% corn meal); T4 (45% BRS capiaçu + 45% silk cotton + 10% corn meal); T5 (30% BRS capiaçu + 60% silk cotton + 10% corn meal), in a completely randomized design with four replications, totaling 20 experimental units.

Elephant grass cv. BRS capiaçu was mechanically harvested (New Holland TL 75 tractor (New Holland Agriculture®, Paranavaí - PR, Brazil) and JF-90 Z10 forage machine (JF Agricultural Machinery, SP, Brazil)), in an area at the Experimental Farm of the State University of Montes Claros - Unimontes, harvested at 10 cm from the ground, after the regrowth period of 120 days. Silk cotton was manually harvested at an average of 30 cm from the ground, in pasture areas in the region, in June 2019, with the plants presenting an approximate height of 1.5 m, and average stalk thickness of 1.0 cm in diameter, being subsequently subjected to dehydration through exposure to the sun for 48 hours, in order to facilitate the chopping process, in addition to prior inactivation of toxic substances present in its composition (Melo, Vaz, Gonçalves, & Saturnino, 2001). Elephant grass cv. BRS capiaçu and silk cotton were ground in a stationary forage machine (JF brand, 40P, Itapura, São Paulo, Brazil), to a particle size of approximately 2.0cm.

For silage making, experimental polyvinyl chloride (PVC) silos were used, with dimensions of 50 cm in height and 10 cm in diameter, fitted with a lid with a Bunsen valve. At the bottom of the silos, 0.4 kg dry sand was added to drain the effluents produced, as well as a foam to avoid contact of the forage with the sand. The set composed of silo, lid, sand and foam were previously weighed to determine the weight of the empty silo. Then, the mass to be ensiled was properly homogenized, according to the proportions of each treatment, deposited in the respective silos, and compacted with the aid of a wooden piston to obtain a density of 550 kg $(m3)^{-1}$. After the end of the ensiling process, silos were sealed with adhesive tape, weighed and stored in the Laboratory of Bromatology at Unimontes, kept at room temperature.

After 42 days of ensiling, silos were weighed to quantify gas losses (Equation 1), calculated according to the methodology described by Jobim, Nussio, Reis, & Schmidt, (2006). After weighing the closed silos, they were opened, with subsequent sampling of forage for further analysis. Next, the set

containing silo, lid, sand and foam were weighed to quantify the effluent produced, determined through the equation proposed by Jobim, Nussio, Reis, & Schmidt, (2006) (Equation 2).

Equation 1.

$$G = [(PCen - Pen) * MSen] - [(PCab - Pen) * MSab] \times 100$$

$$[(PCen - Pen) * MSen]$$

Where:

G = Gas losses in % DM;

PCen = Silo full weight in silage (kg);

Pen = Weight of the set (silo + lid + sand + foam) in the silage (kg);

MSen = Forage DM content in silage (%);

PCab = Full silo weight at opening (kg);

MSab = Forage DM content at opening (%).

Equation 2.

$$E = \frac{(Pab-Pen)x1000}{(MVfe)}$$

Where:

E = Effluent production (kg t-1 natural matter);

Pab = Weight of the set (silo + lid + sand + foam) at opening (kg);

Pen = Weight of the set (silo + lid + sand + foam) in the silage (kg);

MVfe = Ensiled forage green mass (kg).

The pH was determined by silage extract, where after removing the silage from each silo, the material was homogenized, and part of it was pressed with the aid of a hydraulic press to extract the "juice". In the silage juice, immediately after extraction, pH values were determined using a digital potentiometer (Ak 90, Akso Measuring Instruments, São Leopoldo, RS, Brazil) (Wilson & Wilkins, 1972).

The remainder of the silage taken from the silos was pre-dried process in a forced ventilation oven at 55 °C for 72 hours. Subsequently, part of the pre-dried silage was ground in a Wiley mill with 1 mm diameter sieve sieves for chemical analysis, and a second part was ground in 2 mm sieves, in order to enable the digestibility test.

Samples were analyzed for dry matter content (INCT-CA G-001/1 and G-003/1), crude protein (INCT-CA N-001/1) and ash (INCT-CA M-001/1), neutral detergent fiber (INCT-CA F-002/1) and acid detergent fiber (INCT-CA F-003/1), lignin (INCT-CA F-005/1) and non-fiber carbohydrates, according to Detmann et al. (2012). The content of total digestible nutrients (TDN) was estimated according to the following equation (Undersander, Mertens, & Thiex, 1993): TDN (%DM) = 88.9 - [ADF(%DM) x 0.779]

The chemical composition of silk cotton, BRS capiaçu and corn meal can be found in Table 1.

Table 1
Chemical composition of silk cotton, BRS capiaçu grass and mixtures related to the respective experimental treatments, immediately before ensiling

Item (% DM)	Silk cotton	BRS-capiaçu (120 days)	Ground corn [1]
Dry matter	22.81	27.92	87.96
Ash	7.94	7.93	1.64
Crude protein	8.89	6.90	9.01
Neutral detergent fiber	38.92	72.46	13.05
Acid detergent fiber	33.89	49.05	5.03
Lignin		7.11	1.21

[1]Valadares et al. (2016).

To evaluate the ruminal degradation kinetics, two castrated crossbred steers (Holstein/Zebu) were used, equipped with ruminal cannulas, with an average weight of 580kg, receiving diets based on elephant grass cv. BRS capiaçu. The in situ degradability technique was performed in 7.5 x 15 cm (weight 100g) non-woven fabric (NWF) bags with approximately 60 µm mesh size, according to Casali et al. (2009); the number of samples was determined from the ratio of 20 mg DM cm-2 bag surface area (Nocek, 1988).

Twenty samples (referring to four replicates x five treatments) were placed in NWF bags. These twenty NWF bags were then placed in 20 x 30 cm filo bags, together with 100 grams lead, and each filo bag corresponded to an evaluation time. The fillet bags were tied with nylon thread, leaving a length of 1 m so that the bags could move freely in the solid and liquid phases of the rumen. Bags were then deposited in the ventral sac region of the rumen with the tip of the remaining nylon thread attached to the cannula for 0, 3, 6, 12, 24, 48, 72, 96, 120, and 144 hours in reverse order, i.e., starting with the duration of 144 hours. Bags related to time zero were not incubated in the rumen, but were washed in running water, similar to the incubated bags. To estimate the indigestible fraction of NDF, samples were incubated in a bovine for 288 hours (Detmann et al., 2012).

Subsequently, samples were taken to an oven at 55 °C for 72 hours and then cooled in a desiccator and weighed. Residues remaining in BWF bags collected from the rumen were analyzed for DM content. Degradation percentage was calculated by the proportion of feed remaining in the bags after ruminal incubation. Data obtained were fitted to a nonlinear regression by the Gauss-Newton method, using the SAS software (SAS Institute Inc., Cary, NC), according to the equation proposed by Orskov & McDonald (1979): $Y = a + b (1-e^{-ct})$, where: Y = accumulated degradation of the analyzed nutrient component after time t; a = intercepted degradation curve when t = 0, which corresponds to the water-soluble fraction of the analyzed nutrient; b = potential for degradation of the water-insoluble fraction of the analyzed nutrient component; a + b = potential degradation of the analyzed nutrient component when time is not a limiting factor; c = rate of degradation by fermentation of b; t =

incubation time. The disappearance values at time zero ("fraction a") were used to estimate the DM colonization time (CT), where the parameters "a", "b", and "c" were obtained by the Gauss-Newton algorithm: CT = [-ln(a'-a-b)/c].

After calculation, the coefficients a, b and c were applied to the equation proposed by Orskov & McDonald (1979): ED = a + (b x c / c + k), where: ED = effective ruminal degradability of the analyzed nutrient; k = feed through rate. Rates of passage of particles through the rumen were estimated at 2, 5 and 8% h^{-1} as suggested by the AFRC (1993).

Data were tested by analysis of variance using REG procedures of SISVAR, version 5.6 (Ferreira et al., 2014). Variables related to the fermentation profile and chemical composition were analyzed according to the model:

$$Y\ ij = \mu + Trati + eij$$

Where:

Yijk = observation regarding treatment "i" in repetition "j";

μ = constant associated with all observations.

Trati = effect of treatment "i", with i = 1, 2, 3,4, and 5;

eij = experimental error associated with plots that hypothetically have normal distribution with zero mean and variance δ^2.

The DM ruminal degradability test was carried out in a split plot randomized block design with five treatments (plots) and 10 incubation times (subplots) and two blocks. Variation in body weight of each animal was the blocking factor. The following statistical model was used:

$$Y\ ijk = \mu + Ti + Bj + eij + Pk + Ti \times Pik + eijk$$

Where:

Yk(ij) = observation regarding the time (P) in subplot k of treatment (T) i in block j;

μ = constant associated with all observations;

Ti = effect of treatment "i", with i = 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, and 12;

Bj = effect of block j, with j = 1, 2, 3, and 4;

eij = experimental error associated with plots that hypothetically have normal distribution with zero mean, and variance δ2;

P = effect of incubation time k, with k=1, 2, 3, 4, 5, 6, 7, 8, 9, and 10;

TPik= effect of the interaction of the i treatment level with the k level of the incubation time;

eijk = experimental error associated with all observations that hypothetically have normal distribution with zero mean and variance δ2.

When significant by F-test, treatment means and incubation times were compared by decomposing the sum of squares into linear orthogonal contrasts and quadratic effects, with subsequent adjustments to the regression equations. For all statistical procedures, α = 0.05 was used as the maximum tolerable limit for type I error.

Results and Discussion

The inclusion of silk cotton in capiaçu silages linearly reduced the dry matter (DM) content of the silages (Table 2). For each 1% addition of silk cotton to the BRS capiaçu silage, a decrease of 0.06 percentage points (p.p.) of DM in the mixed silages was observed. Based on the estimated DM contents, which varied from 31.81 to 27.02%, in the silage of

elephant grass cv. BRS capiaçu alone and with 60% of silk cotton, respectively, it was possible to observe a reduction of 15.06% DM content of the silages, in the evaluated inclusion range.

Table 2
Average values observed for dry matter (DM), potential of hydrogen (pH), gas losses, effluent losses, and dry matter recovery (DMR) of BRS-capiaçu silage according to different levels of inclusion of silk cotton

Item	Sil Inclusion levels of silk cotton (%) k cotton					CV (%)	p-value	
	0	15	30	45	60		Linear	Quad
Dry matter (%)[1]	30.84	29.89	28.44	28.71	26.69	4.18	<0.01	0.86
pH	3.98	3.86	3.75	3.83	3.94	3.23	0.04	0.01
Gas losses (% DM)	1.04	1.52	1.24	1.74	1.58	28.69	0.06	0.52
Effluent losses (kg t^{-1} NM)	8.97	15.13	13.68	10.49	9.50	48.28	0.69	0.14
DMR (%)	98.21	96.54	96.92	97.44	97.69	1.19	0.94	0.08

CV- Coefficient of variation; NM - Natural matter; P - probability; Equation: 1Ŷ=30.8150-0.0632*X, R2=0.91; *significant by t-test at 5% probability.

The lower DM content in silk cotton implied in these observed results, denoting that pre-drying the material for 48 hours, together with the addition of corn meal in the roughage mixture, at the proportion of 10% natural matter, were determining factors to avoid a decrease in the DM content to critical levels at the highest levels of inclusion of silk cotton. Ramos Neto et al. (2020) evaluated the effect of including different levels of corn meal (0 to 20%) in elephant grass ensilage (Pennisetum purpureum Schum, cv. Napier) observed a 110% increase in DM content, from the lowest to the highest level of inclusion. With the addition of 10% corn meal to elephant grass cv. BRS capiaçu silages, Paula et al. (2020) observed that the silages exceeded the minimum limit of 25% DM to reduce fermentation losses.

The estimated DM contents of silages at the different levels of inclusion of silk cotton are within the range reported in the literature as adequate for a good fermentation. According to Kung, Shaver, Grant, & Schmidt (2018), for an adequate silage fermentation, the DM content of the forage should be between 25 and 35%, values close to those reported by Jobim and Nussio (2013), between 28 and 40%.

When evaluating the chemical composition of the silage of elephant grass BRS capiaçu at 110 days of cutting ages, A. V. Pereira et al. (2016) found 20.4% DM, that is, high moisture content, which represents an obstacle to good fermentation, which was not observed in the present study, given the value of 30.84% DM in silage of BRS capiaçu alone, harvested at 120 days. This denotes, therefore, that soil fertility and climatic conditions, such as temperature and rainfall, can accelerate plant growth and determine variations in dry matter content as a function of harvest age, in different regions or periods.

In fact, Belém et al. (2016) evaluated the digestibility, fermentation and microbiological characteristics of C. procera silage with different amounts of grape pomace, and reported that fresh silk cotton contributes to a reduction in dry matter content in mixed silages; the silage of C. procera alone had a DM content of 22.7%.

Sometimes, the inclusion of alternative feeds in forage silages commonly used in ruminant feed can negatively influence the DM content of the mixture. Bonfá et al. (2017), evaluating different levels of inclusion (0 to 50%) of pineapple bark to elephant grass silage, observed a reduction in DM content, with values from 33.33 to 21.91%, for treatments with lower and higher level of inclusion of this co-product, respectively.

However, the use of additives can be an alternative to control the reduction in the dry matter content of mixed silages, since this mixture of forages results in gains in nutritional value of the ensiled material and that it is economically viable for the producer, allowing ensiling materials impossible to be ensiled in an exclusive way, guaranteeing additional feed roughage for critical times of the year, such as the dry season, for example. When it is not possible to use additives to increase the dry matter content of the mixed silage cotton to silk, pre-wilting elephant grass BRS capiaçu constitutes an auxiliary measure to be considered and can help in achieving suitable DM contents (Ribas et al., 2021). Thus, Oliveira et al. (2017) analyzed elephant grass silages, with and without pre-wilting, and with and without inclusion of cassava meal, and demonstrated that pre-wilting increases silage DM.

The inclusion of silk cotton in elephant grass cv. BRS capiaçu silage did not influence the silage pH, with an average value of 3.87 (Table 2). The pH is one of the fundamental factors to characterize the preservation of the ensiled product. In general, in the literature, pH values between 3.8 and 4.2 have been attributed as suitable for well-preserved silages, since this interval restricts the action of proteolytic enzymes from the plant, from enterobacteria and clostridia, which promote the processes of silage spoilage (McDonald, Henderson, & Heron, 1991). Based on this parameter, it is clear, a priori, that in the evaluated range of inclusion of silk cotton (0 to 60%), the mixed silages of this plant with BRS capiaçu present adequate pH values.

Taking into account the mean pH value of 3.87, associated with estimated DM contents of 31.81 and 27.02%, in the exclusive BRS capiaçu silage and silk cotton silage replacing 60% BRS capiaçu, respectively, it is noted that within the range of inclusion levels of silk cotton evaluated in this study, good quality silages are produced, based on the silage qualification proposed by Tomich, Pereira, Gonçalves, Tomich, & Borges (2003).

The inclusion of silk cotton in the capiaçu silage did not influence gas losses, effluent losses, and dry matter recovery of the evaluated silages (Table 2), with observed averages of 1.42% DM, 11.55kg t^{-1} natural matter (NM) and 97.36%, respectively. These results show that the inclusion of 10% corn meal in the silages, on a natural matter basis, combined with the dry matter content of BRS capiaçu, as well as the pre-drying of silk cotton for 48 hours, were essential to guarantee proper fermentation in the various treatments evaluated. Additionally, one can also attribute the fungistatic effect shown by silk cotton (Lima et al., 2006) as an additional important factor in preserving the original characteristics

of the ensiled material. According to Muck et al. (2018), the existence of fungi in the silage is undesirable, due to the direct action of these microorganisms in the degradation of sugar and lactic acid through the normal pathway of respiration.

Belém et al. (2016) verified the gas losses equivalent to 5.35% dry matter ensiled in an exclusive material of silk cotton. In the present study, at the highest level of inclusion (60%) of silk cotton in the silage of BRS capiaçu, gas losses were 1.58% of the ensiled dry matter. Tropical forages with DM contents below 25% can present high nutrient losses through gases and/or effluents produced, harming the nutritional value of silages, since part of carbohydrates and proteins are present in the effluents (Kung et al., 2018). Considering these contrasting results and taking into account that in Belém et al. (2016), the dry matter content of silk cotton was 22.70%, close to the value of 22.81% observed in the present study, which further reinforces the hypothesis that the addition of material that increases the dry matter content of silk cotton mixed silages, as well as pre-drying the entire plant before crushing and ensiling, are recommended practices to preserve the quality of the ensiled material. Lima et al. (2006) observed that the pre-drying of silk cotton silage for 24 hours resulted in lower values of toxigenic fungi, microorganisms associated with the degradation of sugar and lactic acid through the normal pathway of respiration.

Belém et al. (2016) reported effluent losses equivalent to 1.53 kg t^{-1} NM in exclusive silk cotton silage, against an average value of 11.55 kg t-1 NM observed in the present study. The smaller percentage participation of silk cotton (maximum 60%) in the mixed silages of BRS capiaçu in the present study, compared to the exclusive silk cotton silage in Belém et al. (2016), may justify the lower effluent losses in the latter, possibly due to the greater fungistatic action at higher levels of silk cotton in the ensiled material. Lima et al. (2006) observed that increasing levels of silk cotton in silages indicated a reduction in the levels of toxigenic fungi. Fungi act by promoting the rupture of the cell wall of the ensiled material, favoring the extravasation of the liquid portion (Cytosol) contained within the plant cell. In this sense, Muck et al. (2018) report as undesirable the presence of fungi in silages, responsible for hydrolysis and metabolization of cellulose and other constituents of the cell wall.

Tropical forages with DM contents below 25% can present high nutrient losses through the effluent produced, harming the nutritive value of silages, since part of the carbohydrates and proteins are present in the effluents (Kung et al., 2018).

The lack of effect of including silk cotton in BRS capiaçu silage is in line with the results of gas and effluent losses, which were also not influenced. Pacheco et al. (2013) highlighted that the dry matter recovery rate (DMR) is directly related to losses due to the production of effluents and gases from silages, with high losses in silages causing a reduction in dry matter recovery. Paula et al. (2020), evaluated the chemical composition of elephant grass BRS Capiaçu silage with inclusion of 0 - 20% corn meal, and observed that the control silage presented a lower percentage of dry matter recovery, compared to the other treatments, with an increase of 0.31 percentage points for each percentage unit of corn meal added to the silage. With this, the corn meal used in elephant grass silages

with high moisture content can increase the DM content of silages and, consequently, control or reduce nutrient losses of silages.

There was an increasing linear effect for crude protein (CP) content with increasing levels of inclusion of silk cotton in elephant grass cv. BRS Capiaçu silage (Table 3), estimating an increase of approximately 0.04 percentage points of CP for each 1% silk cotton inclusion. The estimated CP contents varied from 6.74% to 9.39%, in the exclusive capiaçu silages and with the inclusion of 60% silk cotton, respectively, representing an increase of 39.32%.

Table 3
Chemical composition of elephant grass cv. BRS capiaçu silage with different inclusion levels of silk cotton (*Calotropis procera*)

Item (%)	Inclusion levels of silk cotton (%)					CV (%)	p-value	
	0	15	30	45	60		Linear	Quad
Crude protein[1]	6.82	7.12	8.49	8.44	9.47	8.21	<0.01	0.97
Neutral detergent fiber[2]	67.99	64.10	58.43	53.28	50.80	12.31	<0.01	0.80
Acid detergent fiber[3]	45.95	43.37	40.59	36.27	31.89	11.89	<0.01	0.56
Total digestible Nutrients[4]	53.10	55.11	57.27	60.64	64.05	6.32	<0.01	0.56
Cellulose[5]	37.69	35.82	34.21	27.87	26.42	11.34	<0.01	0.57
Hemicellulose	22.03	20.73	17.84	17.01	18.91	23.09	0.17	0.32
Lignin	8.25	7.55	6.37	8.39	5.46	25.38	0.12	0.71
Indigestible neutral detergent fiber[6]	26.54	23.36	23.09	22.60	20.12	6.90	<0.01	0.70

CV - Coefficient of variation; Equations: [1]$\hat{Y}$=6.7430+0.0442*X, R^2= 0.92; [2]$\hat{Y}$=67.9635-0.3013*X, R2= 0.98; 3$\hat{Y}$=46.6635-0.2348*X, R^2= 0.98; [4]$\hat{Y}$=52.5495+0.1859*X, R^2=0.98; [5]$\hat{Y}$=38.5070-0.2032*X, R^2=0.93; [6]$\hat{Y}$=25.8635-0.0906*X, R^2=0.88; *significant by t-test at 5% probability.

Considering that silk cotton did not influence the qualitative parameters of losses by gases and effluents, as well as the recovery of dry matter, when added to BRS capiaçu silage, it can be inferred, based on the results related to the content of crude protein from mixed silages, that *C. procera* is a viable forage alternative to increase the protein content of silages made with forage with low protein value, paying attention to the average dry matter content of the forage mixture. Thus, S. Santos et al. (2012) reported that the inclusion of 30% mesquite meal (*Prosopis julifora*) in elephant grass silage increased CP contents by 61.8%, whereas Garcez et al. (2018) found an increase of 56.66% CP, with the inclusion of 24% of faveleira pod (*Cnidoscolus quercifolius*) in elephant grass silage, thus showing that unconventional forage species can present potential to increase the CP content of elephant grass silages.

Regarding NDF content, there was a decreasing linear behavior (P<0.01), as silk cotton was added to the silage (Table 3). For each 1% silk cotton inclusion to elephant grass cv. BRS capiaçu silages NDF value

was reduced by 0.30 percentage points. The estimated values of NDF varied from 67.96% in silage of elephant grass cv. BRS capiaçu alone, to 49.88%, in silages with 60% inclusion of silk cotton, representing a reduction of 26.60%. The effect found for this variable can be explained by the lower content of NDF present in silk cotton compared to elephant grass BRS capiaçu.

According to the results obtained, it can be estimated with the regression equation that with the inclusion of approximately 26 and 43% silk cotton in the elephant grass cv. BRS capiaçu silage, it is already possible to obtain between 60 and 55% NDF, respectively, in the roughage mixture, since NDF contents higher than these can compromise the feed intake.

Silk cotton inclusion levels linearly reduced the ADF content of capiaçu silages (Table 3), where, for each 1% silk cotton inclusion to elephant grass cv. BRS capiaçu silages reduced the ADF values by 0.23 percentage points. Considering the estimated ADF values, 46.66% in the silage of elephant grass cv. BRS capiaçu alone, and 32.57% in silages with the highest level of inclusion of silk cotton, there was a 30.20% decrease in the content of this component in the dry matter, a result that is in line with the lower content of ADF present in the silk cotton compared to BRS capiaçu.

It is noteworthy that the reduction in ADF and NDF contents found in silages with the inclusion of silk cotton indicates improvement in the nutritive value and quality of the silages. High NDF content may limit forage DM intake by ruminants due to low degradation rates (National Research Council [NRC], 2001), and high ADF values indicate loss of nutritional quality of silages (Van Soest, 1994), as there is a negative correlation between the content of ADF and the degradation rate of the food, that is, with a reduction in the content of ADF, there is an increase in DM digestibility (Berchielli, Pires, & Oliveira, 2011).

Evaluating elephant grass silages combined with 0, 20, 40 and 60% wedge (*Clitoria ternatea*), Oliveira et al. (2017) found a reduction in NDF contents from 67.72 to 56.04%, as well as in NDF contents, from 44.57 to 42.03% assuming a decreasing linear effect, corroborating the results obtained herein.

Total digestible nutrients were influenced by the inclusion of silk cotton in elephant grass cv. BRS capiaçu silage increasing TDN contents by 0.18 percentage points for each 1% inclusion (Table 3). The estimated TDN contents ranged from 52.55% to 63.53%, considering the exclusive capiaçu silages and those including 60% silk cotton, respectively, representing an increase of 20.89%. This result may be directly related to the ADF contents obtained, since the estimate of total digestible nutrients in silages was obtained by an equation based on the contents of this constituent in roughage.

The relationship between TDN and CP content, of 6.76, considering the values of 64.05 and 9.47%, respectively, observed in the mixed silage containing 60% silk cotton inclusion, denotes a relationship that approximates the 5.91 ratio, resulting from the ratio of TDN to CP of 59.02%, and 9.97%, respectively, required by a 400 kg castrated crossbred steer in feedlot and with daily body weight gain of 0.50 kg, contained in the manual of nutritional requirements of purebred and crossbred zebu cattle - BR Corte 2016 (Valadares et al., 2021). This observation demonstrates that the inclusion of silk cotton

in BRS capiaçu silage, in addition to increasing the crude protein content in the mixed silage, also has a balance of TDN and CP closer to the nutritional recommendations for feedlot beef cattle.

A decreasing linear effect was observed for the cellulose content in the evaluated silages (Table 3). Cellulose values were reduced by 0.20 percentage points for each 1% increase in silk cotton and capiaçu silages. According to the estimated values, there was a variation from 38.51 to 26.31% of the cellulose content in silages of elephant grass cv. BRS capiaçu alone and in the silages with the highest level of inclusion of silk cotton, respectively. The effect observed for this variable is directly related to the reduction in levels of the fibrous portion of silages, since cellulose is the main component of the fiber fraction of foods and is obtained by a difference in the fiber content. Hemicellulose and lignin contents were not influenced by the inclusion of different levels of silk cotton in the elephant grass BRS capiaçu (Table 3) silage, with an estimated average of 19.30 and 7.20%, respectively.

Although the lignin content of capiaçu shows 40% superiority when compared to the contents of this variable in the composition of silk cotton, no significant effect was detected on the silages, according to the different inclusion levels.

A linear reduction of indigestible neutral detergent fiber (iNDF) was observed (Table 3). The levels found for iNDF reduced by 0.09 percentage points, for each 1% increment of silk cotton to elephant grass cv. BRS capiaçu silage. iNDF consists of the undigested portion of the plant cell wall along the gastrointestinal tract of ruminants, called fraction "C" in carbohydrate fractionation (Viana et al., 2012). It should be noted that silk cotton, until then considered a weed in pastures, presents morphophysiological characteristics of adaptation to drought better than cultivated grasses, which may imply in its lower susceptibility to oxidation processes and consequent concentration of the indigestible fiber fraction.

The highest lignin content of elephant grass cv. BRS capiaçu (71.1 g kg^{-1}) compared to silk cotton (42.2 g kg^{-1}) could justify the reduction found for NDF with increasing inclusion levels, since lignin makes up the fraction C of fiber carbohydrates, but no significant effect was detected for lignin content. Thus, it can be seen that silk cotton favored the levels of degradability of capiaçu silages, in view of the determined estimates.

Regarding the ruminal kinetics, it was possible to observe an increasing linear effect of the soluble fraction ("a") and the degradation rate of fraction "b" ("c") with an increase of 0.05 and 0.0005 percentage points, respectively, for each 1% inclusion of silk cotton in BRS capiaçu silage (Table 4). These increases observed for fraction "a" and "c" may be related to the reduction in NDF content in mixed silages, as the percentages of participation of silk cotton in the mixture were increased. For fraction "b" and for potential degradability (PD), a quadratic effect was found, obtaining minimum points of 23.72, and 50.52%, respectively, in the proportion of 45% inclusion of silk cotton to BRS capiaçu silage.

Table 4
Kinetic parameters of in situ dry matter (DM) degradation of elephant grass cv. BRS Capiaçu silage with different inclusion levels of silk cotton (*Calotropis procera*)

Item (%)	Inclusion levels of silk cotton (%)					CV (%)	p-value	
	0	15	30	45	60		Linear	Quad
Soluble fraction (a)[1]	24.24	24.85	26.00	26.89	27.60	4.23	<0.01	0.98
Potentially degradable fraction (b)[2]	49.07	27.88	26.92	27.34	25.53	14.26	0.00	<0.01
Degradation rate of fraction b (c), in %/h[3]	1.00	2.00	2.00	3.00	5.00	21.36	<0.01	0.11
CT, h[4]	11.59	13.67	12.05	9.48	6.41	24.44	<0.01	0.03
PD, %[5]	73.30	52.73	52.91	54.23	53.14	7.82	0.00	<0.01
ED, k=2%[6]	42.60	37.95	40.17	42.96	45.44	4.98	0.00	<0.01
ED, k=5%[7]	33.71	32.22	34.39	36.83	39.88	4.01	<0.01	0.00
ED, k=8%[8]	30.52	29.99	31.97	34.09	36.97	3.57	<0.01	0.00
If, %9	26.70	47.27	47.08	45.77	46.86	10.47	0.00	<0.01

CV - coefficient of variation; c - rate of degradation of fraction b; CT - colonization time; PD- potential degradability; ED - effective degradability; If - indigestible fraction. Equations: [1]$\hat{Y}=24.16+0.0584*X$, $R^2= 0.99$; [2]$\hat{Y}=46.60-1.08*X+0.0127*X^2$, $R^2= 0.86$; [3]$\hat{Y}=0.009+0.0005*X$, $R^2= 0.89$;[4]$\hat{Y}=13.55-0.0969*X$, $R^2=0.67$; [5]$\hat{Y}=70.75-1.02*X+0.0127*X^2$, $R^2=0.82$; [6]$\hat{Y}=41.80-0.21*X+0.0047*X^2$, $R^2=0.82$; [7]$\hat{Y}=32.02+0.1129*X$, $R^2=0.79$; [8]$\hat{Y}=29.37+0.1119*X$, $R^2=0.87$; [9]$\hat{Y}=29.24+1.02*X-0.0127*X^2$, $R^2=0.82$; *significant by t-test at 5% probability.

The colonization time (CT) linearly reduced according to the levels of inclusion of silk cotton to the capiaçu silage. For each 1% inclusion, a reduction of 0.09 percentage points was verified. The colonization time is directly related to the potential and effective degradation of food (Silva et al., 2012). The smaller the accessibility of microorganisms to the feed, the greater the amount of slowly degrading components, thus compromising the adequate supply of nutrients for microbial metabolism, altering ruminal parameters (Campos, Bose, Boin, Lanna, & Morais, 2000).

In the effective degradability (ED) at a rate of passage of 2%, it was possible to observe a quadratic effect, reaching a minimum point of 39.69% when 15% silk cotton were added to the capiaçu silage. The ED with 5 and 8% was linearly influenced by the evaluated inclusion levels, where, for each 1% inclusion of silk cotton to the capiaçu silage, an increase of 0.11 percentage points was observed for both rates of passage. In the inclusion level of 45% silk cotton in the capiaçu silage, it was possible to observe a maximum percentage of the indigestible fraction (49.48%) in silages, with a quadratic effect.

In general, the inclusion of silk cotton in the silage of elephant grass BRS capiaçu, at the evaluated levels, did not change the fermentation parameters of the mixed silages, considering the results of pH, losses by gases and effluents, as well as the recovery of the ensiled dry matter.

The chemical composition of the mixed silages showed better results with higher levels of inclusion of silk cotton, increasing the content of CP and TDN and reducing the content of fiber components (NDF, ADF, cellulose), as well as ruminal kinetics parameters.

These results indicate that silk cotton is a roughage material that can be strategically used, especially in semi-arid regions, where the production of DM for animal feed, in quantity and quality, is compromised in a certain period of the year.

Conclusion

The inclusion of silk cotton in up to 60% natural matter during the BRS capiaçu ensiling process improves the indicators of the nutritional value of silages, increasing the crude protein contents, reducing the components of the fiber fraction of the feed, and favoring the degradability parameters.

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