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The quality and fermentation of the total diet containing BRS capiaçu or sugarcane with or without urea

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Abstract. The objective of this study was to evaluate total diet silages in PVC silos, containing BRS Capiaçu or sugarcane with or without urea. The experimental design was completely randomized with a 2x2 factorial arrangement with ten replicates, with one of the roughages (sugarcane or Capiaçu) being with or without urea (2% urea on a dry matter basis). No interaction was observed between the bulk and urea factors for the variables analyzed. Diets containing sugarcane presented greater gas losses and lower pH. In contrast, effluent losses and dry matter recovery were higher in diets containing BRS Capiaçu. Ammonia nitrogen levels were influenced by both the type of roughage and the addition of urea, being higher in diets with urea and those containing BRS Capiaçu. Diets with BRS Capiaçu also resulted in higher levels of ash, NDF, ADF, hemicellulose, cellulose, and lignin. The addition of urea, in turn, reduced lignin levels. The levels of ADIN and NDIN were higher in diets with BRS Capiaçu and lower in those with sugarcane, while urea reduced ADIN. For NFC, there was an effect of roughage and urea, with higher values observed in diets with sugarcane and with the addition of urea. The highest iNDF levels were found in diets with BRS Capiaçu and without urea. Protein fractionation showed that the addition of urea increased fraction A and reduced fractions B1+B2 and C. Fraction B3 was higher in diets with sugarcane. Fraction C showed higher levels with BRS Capiaçu and when urea was not added. Sugarcane resulted in higher levels of fractions A+B1 and lower levels of B2 and C, a pattern opposite to that observed with BRS Capiaçu. Similarly, urea increased fractions A+B1 and decreased fractions B2 and C. The addition of urea to the diets promoted changes in the carbohydrate and protein fractions, improving the composition. Regarding the roughage used, sugarcane presented better fermentation parameters when purchased from Capiaçu.

Key words: chemical composition, conservation, fermentation, total mixed ration.

INTRODUCTION

Ruminant production is an important factor in the Brazilian agricultural economy, exerting a significant impact on the national livestock scenario. Production systems, which vary in intensity (extensive, semi-intensive, and intensive), share a dependence on the use of forage as the basis of animal feed. In this context, the efficient management of forage plants, from soil preparation for planting to maintenance, conservation, and utilization of surpluses, emerges as a critical factor for the success of the activity.

Forages are fundamental for providing essential nutrients to ruminants, and Brazil holds a climatic diversity that allows the cultivation of various species. However, forage production faces challenges such as degradation and seasonality, requiring the adoption of effective and sustainable techniques to optimize production. Among the various methods of forage conservation, silage stands out for its growing popularity, especially among small and medium-sized producers. The technique consists of conserving the material through anaerobic fermentation, presenting a low implementation cost and high efficiency in the production of silage, a conserved and high-quality bulky feed.

Total diet silage, which involves ensiling all the ingredients that make up the animals' diet, represents a promising advancement in the field. This technology, as highlighted by Sá et al. (2023), offers the flexibility to use a variety of ingredients, from fresh forages and concentrates to agro-industrial by-products, which can result in reduced feeding costs. The search for methods that conserve forages by optimizing animal feed supply has driven interest in the ensiling process. The addition of concentrated feeds to the process, as demonstrated by McDonald et al. (1991), can bring benefits such as reduced effluent losses, improved fermentation, and increased silage nutritional value, due to its ability to act as moisture sequestrants.

Sugarcane and capiaçu (*Pennisetum purpureum Schumach*), tropical grasses with high green matter production per hectare, stand out as promising roughage feeds for total diet silage. In addition to distinct nutritional characteristics, such as sugarcane's high total digestible nutrient content (60.57%) and the potential to improve the nutritional quality of diets, both grasses are well accepted by animals, even after ensiling, making them viable components for diets (Karimuna et al., 2023). The addition of urea to the silage production process brings benefits due to its role as a microbial additive and the improvement in the fermentation process, which can reduce dry matter and soluble sugar losses, as highlighted by Zanella et al. (2024). Low levels (2%) of urea are recommended because they balance nitrogen supply without causing toxicity and also prevent excessive nitrogen loss through urine, as discussed by Martins et al. (2021). Furthermore, urea can partially or completely replace protein concentrates used in diets, contributing to cost reduction and improving the quality of the final material.

With this objective, the present work aims to evaluate the fermentative and nutritional parameters of tropical grass silage, such as sugarcane and BRS Capiaçu, with the addition of urea.

MATERIALS AND METHODS

The experiment was conducted in the Forage and Pasture sector, located at the State University of Southwest Bahia, Juvino Oliveira Campus. The research was conducted in a completely randomized design, in a 2×2 factorial arrangement, with two sources of bulk feed (BRS Capiaçu or sugarcane) with and without urea (0 and 2% urea on a dry matter basis) with ten replicates. The sugarcane used was of the RB72454 variety, from the third cut and with a Brix degree of 20. The grass, in turn, was cut 80 days after regrowth.

Following harvest, the bulk materials underwent a disintegration process, utilizing equipment adjusted to produce particles with a uniform size of 30 mm. This step aims to optimize compaction and fermentation during ensiling. Subsequently, the material was carefully homogenized and weighed, following the proportions established in the experimental diets. Compaction was carried out in PVC mini-silos, with dimensions of 50 m in height and 10 m in diameter, using concrete tampers to ensure adequate density. The target compaction densities were 925 kg m^{-3} for sugarcane-based diets and 860 kg m^{-3} for those with Capiaçu. The compaction process was conducted to the maximum limit of each silo, seeking to optimize storage capacity and simulate real-scale ensiling conditions.

After a 42-day anaerobic fermentation period, the experimental silos were carefully opened for evaluation. The opening methodology involved the decompaction of the ensiled material and the collection of representative samples for subsequent laboratory analysis. It is important to emphasize that the integrity of the fermentation process was ensured by the use of 40 PVC mini-silos, each equipped with a Bunsen valve for internal pressure control and gas escape.

Additionally, each silo contained a 10 m layer of dehydrated sand at the bottom, previously sterilized in a forced-air circulation oven at 55 °C for 72 hours, with the aim of creating a dry environment and preventing the proliferation of undesirable microorganisms, as well as for quantifying effluent losses. The separation of sand from forage was carried out using a screen with dimensions corresponding to the internal diameter of the PVC tubes, ensuring the absence of contamination of the ensiled material during the ensiling and silo opening process.

Analysis

After opening the silos, representative samples from each diet were carefully collected for the evaluation of their chemical composition. The analysis process began with the pre-drying of the samples in an oven, followed by mechanical processing to homogenize the material. The analytical determinations rigorously followed the methodologies described by Detmann et al. (2021), aiming to ensure the accuracy and reliability of the results.

The following contents were determined: dry matter (DM); crude protein (CP); neutral detergent fiber (NDF); acid detergent fiber (ADF); acid detergent fiber corrected for ash and protein (NDFap); ether extract (EE); neutral detergent insoluble nitrogen (NDIN); and acid detergent insoluble nitrogen (ADIN). The analysis of chemical composition, following the methodologies of Detmann et al. (2021), allows for the creation of a complete nutritional profile of the evaluated diets, providing crucial information for the interpretation of results and the planning of optimized feeding strategies for the animals. In Tables 1, 2 and 3 it is possible to observe the bromatological composition of the ingredients used and the percentage of each ingredient in the experimental diets.

In diets that did not contain urea in their formulation, NFC were calculated by difference, using the following equation proposed by Detmann et al. (2021): NFCap = 100 – MM – EE – NDFap – CP. In diets that incorporated urea as an additive, the calculation of NFC required an adaptation of the equation, considering the contribution of non-protein nitrogen (NPN) from urea. The equation used, as recommended by Hall (2001), was as follows: NFCap = 100 – MM – EE – NDFap – (CP – CPU + U). In this formulation, CPU represents the crude protein from urea, while U corresponds to the urea content in the diet. The inclusion of these parameters in the equation ensures that the NFC estimate accurately reflects the nutritional composition of diets with urea, considering urea nitrogen as part of the protein fraction.

The determination of total digestible nutrients (TDN), in turn, was carried out according to the methodology proposed by Capelle et al. (2001). TDN represents an estimate of the energy available in the feed for the animals, taking into account the digestibility of the different components of the diet.

Table 1. Chemical composition of the ingredients used in the experimental diets

Itens	Ingredients			
	Sugar cane	BRS Capiaçu	Ground corn	Soybean meal
Dry matter (%)	27.9	28.3	89.0	88.0
Mineral matter[1]	2.9	10.1	1.3	6.8
Crude protein[1]	3.5	7.0	9.8	51.0
Ethereal extract[1]	1.8	2.3	4.3	2.3
Neutral detergent fiber cp	51.5	69.1	16.0	18.3
Acid detergent fiber cp	30.1	45.1	8.5	11.9
NDIN[2]	27.8	31.0	14.2	8.5
ADIN[2]	17.3	19.8	4.5	0.8
Cellulose [1]	35.6	30.0	7.8	11.0
Hemicellulose [1]	13.7	28.0	5.1	5.4
Lignin [1]	7.1	7.9	2.1	1.2
Non-fibrous carbohydrates cp[1]	40.3	11.5	68.6	21.6
Total digestible nutrients [1]	68.7	54.6	91.2	82.4

[1]percentage of dry matter; [2] percentage of total nitrogen; NDIN: Neutral detergent insoluble nitrogen; ADIN: Acid detergent insoluble nitrogen, cp: corrected for ash and protein.

Table 2. Proportion of ingredients in the total experimental diets

Ingredients	Diets			
	Sugarcane without urea	Sugarcane with urea	BRS Capiaçu without urea	BRS Capiaçu with urea
Sugar cane[1]	60.0	60.0	0.0	0.0
Capiaçu[1]	0.0	0.0	60.0	60.0
Ground corn[1]	19.3	29.7	25.5	36.0
Soybean meal[1]	18.7	6.30	12.5	0.0
Urea[1]	0.0	2.0	0.0	2.0
Mineral[1]	2.0	2.0	2.0	2.0
Water [2]	508	508	437	437

[1] Percentage; [2]mL.

Table 3. Composition of experimental diets

Itens	Diets			
	Sugarcane without urea	Sugarcane with urea	BRS Capiaçu without urea	BRS Capiaçu with urea
Dry matter (%)	31.5	31.0	30.8	31.1
Mineral matter[1]	5.2	3.6	8.5	8.9
Crude protein[1]	14.7	13.3	14.0	15.8
Ethereal extract[1]	1.9	2.0	2.3	2.4
Neutral detergent fiber[1]	48.7	50.1	55.4	54.7
Neutro detergent fiber$_{cp}$[1]	45.9	48.6	53.7	52.8
Acid detergent fiber$_{cp}$[1]	29.1	27.8	37.2	36.9
Cellulose[1]	21.7	22.3	23.5	24.8
Hemicellulose[1]	17.9	17.1	19.7	18.6
Lignin[1]	6.0	5.8	4.8	4.4
Non-fibrous carbohydrates$_{cp}$[1]	29.5	36.3	21.5	23.6

[1] Dry matter percentage.

Fermentation profile

The ensiling step involved the packing of approximately 2.9 kg of green material per silo for the sugarcane-based diets and 2.7 kg per silo for the diets containing BRS Capiaçu. These values correspond to the compaction densities of 925 kg m^{-3} and 860 kg m^{-3}, respectively, previously established as parameters to optimize the ensiling process.

After a 42-day storage period, the silos were carefully opened and subjected to a controlled aeration process for 30 minutes. This step, crucial for allowing the volatilization of gases resulting from anaerobic fermentation, aims to minimize dry matter losses and ensure a safe working environment.

The weighing of the silos with and without lids, performed before and after the aeration period, allowed for the measurement of gas losses. Effluent losses, gas losses, and the determination of dry matter recovery were calculated by the weight difference of the silos, using the methodologies proposed by Jobim (2007).

The calculation of gas losses (GL) was performed using the following equation: GL = [(WSf – WSa)/(FMf x DMf)] x 100, where WSf represents the weight of the silo with silage, WSa the weight of the silo at opening, FMf the forage mass in silage, and DMf the dry matter content of the forage in silage. This equation allows gas losses to be expressed as a percentage of dry matter.

Effluent losses (EL) were calculated using the equation: EL = [(WE x 1000)/FMi], where WE represents the weight of the effluent (obtained by the difference between the weight of the empty set after opening and the weight of the empty set before filling) and FMi the amount of ensiled green mass. This equation expresses effluent losses in kg per ton of green mass.

Dry matter recovery (DMR) was determined by the equation: DMR(%NM) = (FMf x DMf)/(FMi x Dmi) x 100, where FMf represents the forage mass at opening, DMf the dry matter content of the forage at opening, FMi the forage mass at closing, and Dmi the dry matter content of the forage at closing. This equation expresses the dry matter recovery rate as a percentage of natural matter.

Total dry matter losses (TDML) were calculated using the equation: TDML = [(DMi – DMf)/DMi] x 100, where DMi represents the initial dry matter amount and DMf the final dry matter amount. This equation expresses dry matter losses as a percentage of initial dry matter.

The evaluation of silage quality, carried out on the day the silos were opened, included the determination of pH and ammoniacal nitrogen content ($N\text{-}NH_3$), crucial parameters for characterizing the fermentative profile and nutritional value of the ensiled material. The pH was determined in 15g samples from each silo, following the methodology described by Detmann et al. (2021).

The pH analysis, a fundamental indicator of medium acidity, provides valuable information about fermentation quality, influencing silage preservation and nutrient availability for animals. Ammoniacal nitrogen (N-NH3) analysis, in turn, was performed using silage juice, obtained from 25 g samples from each silo. The samples were immediately treated with 200 mL of 0.2N sulfuric acid solution, aiming to solubilize N-NH3 and preserve sample integrity. The acidified samples were placed in capped jars and stored under refrigeration for a period of 48 hours (Bolsen et al., 1992).

After the solubilization period, the samples were filtered through filter paper to remove solid particles and then subjected to distillation with 2N potassium hydroxide (KOH), using the Kjeldahl method, and titrated with 0.1N hydrochloric acid (HCl), as recommended by Detmann et al. (2021).

The data were subjected to analysis of variance using the Statistical Analysis System (SAS) program. Significant interactions were unfolded and the means compared using the F test at 5% probability.

RESULTS AND DISCUSSION

Regarding dry matter losses and recovery, the interaction was not significant ($P > 0.05$). However, a significant effect ($P < 0.05$) of the bulk feed type was observed, with diets containing sugarcane showing higher gas losses (Table 4). This result can be attributed to alcoholic fermentation, a metabolic process predominant in sugarcane, due to its high concentration of soluble sugars, mainly sucrose. During ensiling, the action of microorganisms results in the production of ethanol as a byproduct, accompanied by the release of carbon dioxide. The gas losses observed in the present study exceeded the values reported by Pedroso et al. (2007), who obtained an average of 10.3% for sugarcane silage without additives. This discrepancy may be attributed to variations in experimental conditions or the specific composition of the silage used.

Table 4. Losses and dry matter recovery (DMR) of total diet silagens of different roughages with or without urea

Variable	Bulky		Urea		CV	*P*- value		
	Cane	Capiaçu	Without	With		Bulk	Urea	Bulk * Urea
Gas loss [1]	14.8	2.9	9.22	8.51	46.2	0.0000	0.5799	0.7830
Effluent loss [2]	59.4	75.9	70.5	64.7	10.3	0.0000	0.0121	0.1608
DMR[1]	83.9	89.7	87.0	86.6	2.1	0.0000	0.7893	0.2381

[1]Percentage of DM; [2]kg per ton, Bulky:Bulky.

Thus, the average values of 14.8% found in the present study (Table 4) are higher than expected, considering total diet silages that contain dry feeds (corn meal and soybean meal) which act as moisture sequestrants, which should reduce overall losses.

The comparative analysis of effluent losses and dry matter recovery revealed that diets containing BRS Capiaçu presented higher values in relation to these parameters. The higher effluent production can be attributed to a combination of factors intrinsic to the plant's composition. The leaf/stem ratio of Capiaçu, for example, is significantly higher than that of sugarcane, which contributes to the greater release of sap during the ensiling process. The rupture of the ensiled material, resulting from cutting and compaction, also plays a crucial role in effluent production. In the case of Capiaçu, the higher proportion of leaves makes the material more susceptible to rupture when compared to stems, resulting in greater liquid release. The observation of these factors demonstrates the complexity of ensiling and the importance of considering the specific characteristics of each bulk product to optimize the process and minimize losses.

The data analysis revealed a significant effect of urea addition on effluent losses. It was observed that diets without urea had the highest average losses. This result corroborates the findings of Santos et al. (2021), who, in studies with corn silage, found a reduction in losses from 160 kg per ton to 109 kg per ton with the addition of 2% urea, on a dry matter basis. The efficacy of urea in modulating fermentative processes and minimizing effluent losses is therefore evident.

Additionally, fermentation efficiency emerges as a determining factor in effluent losses. Although both diets presented pH values within the range considered ideal by McDonald (1991), between 3.8 and 4.2, sugarcane showed a slightly lower pH (3.8) compared to BRS Capiaçu (4.0), as demonstrated in Table 5. This subtle difference suggests that the fermentation process was more efficient in sugarcane, which may have contributed to the lower production of effluents. The complexity of the interactions between urea addition, pH, and the specific characteristics of each forage highlights the importance of a careful analysis to optimize silage conservation.

Table 5. pH and ammoniacal nitrogen of total diet silages of different roughages with or without urea

Variable	Bulky		Urea		CV	*P*- value		
	Cane	Capiaçu	Without	With		Bulk	Urea	Bulk * Urea
pH	3.8	4.0	3.9	3.9	1.3	0.0000	0.0510	0.4470
[1]N-NH_3	8.6	10.6	8.4	10.8	23.4	0.0077	0.0015	0.2107

[1]Percentage of total nitrogen, bulky: Bulk.

The evaluation of dry matter recovery revealed a significant effect ($P < 0.05$) of the bulk feed type, with silages containing sugarcane showing a lower recovery rate. This result can be interpreted in light of the observed gas losses, establishing a correlation between these parameters. Siqueira et al. (2007) corroborate this association, highlighting the preponderant role of carbon dioxide (CO_2) production by yeasts during fermentation in reducing dry matter recovery. The release of CO_2, a byproduct of alcoholic fermentation, contributes to the loss of dry mass and, consequently, to the decrease in the recovery rate. The joint analysis of gas losses and dry matter recovery provides a comprehensive view of the fermentative processes and losses associated with sugarcane ensiling.

The analysis of pH levels revealed the absence of a significant interaction ($P > 0.05$) between the studied factors. However, a significant effect ($P < 0.05$) of the bulk feed type was observed, as shown in Table 5. Diets containing sugarcane showed lower pH values compared to BRS Capiaçu, although both bulk feeds were within the range considered ideal for ensiling, between 3.8 and 4.2, as established by McDonald (1991). The lower acidity observed in sugarcane diets can be attributed to the higher concentration of soluble carbohydrates (CHOs) in this bulk feed, compared to BRS Capiaçu (3.8 and 4.0, respectively). During fermentation, microorganisms utilize CHOs, resulting in the production of organic acids and, consequently, a reduction in pH. The addition of urea did not exert a significant influence ($P > 0.05$) on pH levels, with diets showing an average value of 3.9 (Table 5). The measurement of pH is a crucial indicator of fermentation quality, being one of the parameters used in silage classification (Jobim et al., 2007).

The analysis of ammoniacal nitrogen (NH3-N) values, presented in Table 5, revealed a significant effect ($P < 0.05$) of both the bulk feed type and urea addition. Diets containing BRS Capiaçu showed the highest $N\text{-}NH_3$ levels, as did those that received urea in their composition. The influence of urea on N-NH3 concentration lies in its non-protein nitrogenous nature. When hydrolyzed by urease, urea is converted to ammonia (NH_3), which, in contact with water, forms ammonium hydroxide, resulting in an increase in $N\text{-}NH_3$ content. This mechanism explains the higher $N\text{-}NH_3$ values observed in diets supplemented with urea (10.8%). The interpretation of $N\text{-}NH_3$ levels allows inferences about fermentation quality. Levels below 10% of total nitrogen indicate moderate protein degradation, while values above 15% indicate more intense proteolysis (AFRC, 1998). Based on this criterion, diets composed of sugarcane and those that did not receive urea can be classified as silages with more efficient fermentation and conservation.

Although diets containing BRS Capiaçu and those supplemented with urea showed ammoniacal nitrogen (NH_3-N) levels above 10% (Table 5), these values should be interpreted with caution. Despite indicating more intense proteolysis compared to the other diets, the observed NH3-N levels are still within a reasonable range. It is important to emphasize that excessive proteolysis in silages is generally characterized by NH_3-N/TN values above 15%, which was not observed in the present study.

According to Restelatto (2019), proteolytic activity in silages is the result of the combined action of proteolytic enzymes present in plants and the activity of microorganisms, such as bacteria from the genera *Clostridium* and Enterobacter. The presence of these microorganisms, in particular, can intensify protein degradation, resulting in increased N-NH3 levels. Therefore, the careful analysis of N-NH3 levels allows inferences about fermentation quality and the extent of proteolysis in the evaluated silages.

The analysis of the chemical composition of the diets revealed the absence of significant effects ($P > 0.05$) for the interaction between the studied factors, as demonstrated in Table 6. Additionally, no significant effects ($P > 0.05$) of the bulk feed type or urea addition on dry matter and crude protein contents were observed (Table 6). This absence of variations in crude protein contents can be attributed to the standardization of the diets, which were formulated to be isoproteic. The homogenization of dry matter contents, with an average value of 30%, was also intentional, aiming to optimize the conditions for the silage fermentation process. The addition of water, performed according to the dry matter content of the ingredients, aimed to standardize the moisture of the silages, a crucial factor for the quality of fermentation and preservation of the ensiled material.

The analysis of ash contents in the diets revealed a significant effect ($P < 0.05$) of both the bulk feed type and urea addition. Diets containing BRS Capiaçu showed the highest ash contents, which can be justified by the higher concentration of mineral matter in this culture, as demonstrated in Table 1. The comparison with the results of Retore et al. (2020), who worked with BRS Capiaçu silages at different cutting ages, reveals that the ash contents observed in the present study, 7.9% (Table 6), were higher than those reported by these authors (7.4% for grass cut at 60 days). This difference can be attributed to the contribution of the other diet ingredients, which also present significant mineral contents.

Table 6. Bromatological composition of total diet silages from different bulk feeds with or without urea

Variable	Bulky		Urea		CV	P- value		
	Cana	Capiaçu	Sem	Com		Bulky	Urea	Bulky * Urea
Dry matter %	29.4	30.4	29.9	29.8	2.9	0.0610	0.7597	0.4567
Crude protein [1]	14.3	13.9	13.9	14.3	7.7	0.0652	0.2143	0.1363
Gray [1]	5.1	7.9	6.9	6.1	8.6	0.0000	0.0001	0.4517
Ethereal extract [1]	2.9	2.4	2.6	2.7	19.2	0.0512	0.2126	0.4200
NDF[1]	44.9	50.7	48.3	47.3	5.1	0.0000	0.1761	0.2641
ADF[1]	29.7	34.1	32.3	31.5	8.1	0.0000	0.3526	0.0895
Hemicellulose [1]	15.3	16.6	16.1	15.8	10.5	0.0185	0.5793	0.2998
Cellulose [1]	24.8	29.0	27.1	26.7	6.5	0.0000	0.5372	0.1293
Lignin [1]	4.9	5.5	5.6	4.8	12.9	0.0107	0.0004	0.3689
ADIN [2]	5.4	6.3	6.4	5.3	13.1	0.0001	0.0000	0.0571
NDIN [2]	14.9	12.6	17.1	10.4	14.5	0.0008	0.0000	0.3806
NDFcp[1]	39.1	44.2	41.7	41.6	5.71	0.0000	0.9440	0.4311
NFCcp[1]	39.8	33.9	35.1	38.7	8.70	0.0000	0.0003	0.4559
NDFi[1]	11.7	13.1	13.4	11.4	13.1	0.0107	0.0004	0.3689
TDN[1]	78.3	73.8	73.1	79.0	30.33	0.0001	0.0001	0.4223

[1]Percentage of dry matter; [2]Percentage of total nitrogen; Bulky:Bulky; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; NIDA: NFC: Non-fibrous carbohydrates; TDN: Total digestible nutrients.

Regarding urea addition, the highest ash values were observed in diets that did not contain urea. This is because the diet without urea had a higher amount of soybean meal and consequently a higher mineral content.

The analysis of the fibrous composition of the diets revealed a significant effect ($P < 0.05$) of the bulk feed type on the contents of neutral detergent fiber (NDF), acid detergent fiber (ADF), protein-corrected neutral detergent fiber (NDFcp), hemicellulose, cellulose, and lignin. Diets containing BRS Capiaçu showed the highest values for these parameters, as observed in Table 6. This higher concentration of fibrous components can be attributed to the structural characteristics and bromatological composition of BRS Capiaçu, compared to sugarcane.

The comparison with the results of Retore et al. (2021), who worked with BRS Capiaçu silages, reveals that the NDF, ADF, hemicellulose, and cellulose contents observed in the present study were lower than those reported by these authors (67.33, 39.35, 27.98, and 35.08, respectively). This discrepancy can be explained by the inclusion of concentrated feeds in the diets of the present study, which have lower fiber contents, diluting the fiber concentration in the silages.

Urea addition did not exert a significant influence ($P > 0.05$) on hemicellulose and cellulose contents, with diets showing similar values (16.1% and 15.8% for hemicellulose, and 27.1% and 26.7% for cellulose, respectively).

The analysis of lignin contents revealed a significant effect ($P < 0.05$) of urea addition, with diets without urea showing values of 5.6% and diets with urea showing 4.8%. The reduction in lignin contents with urea addition can be attributed to the fermentation and degradation of urea during ensiling, resulting in the release of ammonia. Ammonia, in turn, can modulate the activity of ligninolytic enzymes, altering the lignin composition and, consequently, reducing its contents. This effect is considered beneficial, since the reduction of lignin is directly correlated with the improvement of diet digestibility.

The higher concentration of lignin observed in the diets containing BRS Capiaçu follows the same trend observed for NDF and ADF contents, reflecting the intrinsic characteristics of this culture. Total digestible nutrients (TDN), as demonstrated in Table 6, showed the highest values in diets containing sugarcane and in diets supplemented with urea. This result was expected, considering the higher nutritional quality of sugarcane and the positive effect of urea on feed digestibility.

The analysis of acid detergent insoluble nitrogen (ADIN) and neutral detergent insoluble nitrogen (NDIN) levels revealed a significant effect ($P < 0.05$) of the bulk feed type. Diets containing BRS Capiaçu showed the highest ADIN values (6.3%) and the lowest NDIN values (12.6%), in contrast to diets containing sugarcane, which exhibited the lowest ADIN values (5.4%) and the highest NDIN values (14.9%). The interpretation of these results lies in the understanding that lower ADIN levels are desirable in ruminant diets. ADIN represents the nitrogen fraction that is unavailable for utilization by microorganisms present in the rumen, limiting the availability of nitrogen for microbial protein synthesis. Therefore, sugarcane, with its lower ADIN levels, has an advantage in terms of nitrogen availability for rumen microorganisms.

The analysis of the effect of urea on acid detergent insoluble nitrogen (ADIN) levels revealed a significant reduction ($P < 0.05$) in values for diets supplemented with urea (5.3%), compared to diets without urea (6.4%). This decrease in ADIN is a positive effect of urea, indicating the solubilization of part of the nitrogen that would otherwise be unavailable to rumen microorganisms.

Regarding neutral detergent insoluble nitrogen (NDIN), an even more significant reduction was observed with urea addition, with values of 10.4% for supplemented diets, compared to 17.1% for diets without urea. This result was expected, considering the high solubility of nitrogen present in urea, which results in a lower NDIN fraction.

The analysis of ash and protein-corrected non-fibrous carbohydrates (NFCap) revealed a significant effect ($P < 0.05$) of the bulk feed type. Diets containing sugarcane showed the highest NFCap contents, which can be attributed to the higher concentration of soluble carbohydrates in this culture. Urea addition also exerted a significant effect ($P < 0.05$), with supplemented diets showing the highest NFCap contents. This effect can be explained by the action of urea in the solubilization of carbohydrates bound to the cell wall, increasing NFCap availability.

The analysis of insoluble neutral detergent fiber (iNDF) revealed a significant effect ($P < 0.05$) of both the bulk feed type and urea addition. Diets containing BRS Capiaçu showed the highest iNDF contents, as did diets that did not receive urea in their formulation. These results can be interpreted in light of lignin contents, since iNDF represents the least soluble fiber fraction, and lignin is the most resistant component. Therefore, the higher the lignin content in the bulk feed, the lower the fiber availability.

Urea addition resulted in a significant reduction in lignin levels (5.6% for diets without urea and 4.8% for diets with urea), demonstrating the ability of urea to modify lignin structure and reduce its levels. This effect is crucial for improving fiber availability and digestibility in diets, which is reflected in the higher total digestible nutrient (TDN) values observed in diets containing sugarcane and urea (Table 6).

The protein fractionation analysis revealed the absence of a significant interaction ($P > 0.05$) between the studied factors. However, significant effects of the isolated factors on the individual fractions were observed.

Urea addition had a significant effect on fraction A, resulting in higher values in urea-supplemented diets. This result was expected, considering the non-protein nitrogen (NPN) nature of urea, which directly influences fraction A, characterized by its high solubility. Urea, being a readily available nitrogen source for rumen microorganisms, contributes to the increase of this fraction.

The B1+B2 fraction was also significantly influenced ($P < 0.05$) by urea addition. The justification lies in the fact that urea, a nitrogen-rich source, partially replaces the protein concentrate in the diets (soybean meal). This substitution results in a reduction of true protein sources (fraction B1+B2), so that the increased addition of NPN in replacement of protein concentrates leads to a decrease in the amount of true protein.

Fraction B3 showed significant effects of both the bulk feed type and urea addition, with the highest values observed in sugarcane-containing diets and in urea-supplemented diets. Daniel & Jobim (2022) discuss these changes in the composition of protein fractions in total diet silages, observing an increase in fractions A1 (ammonia) and A2 (amino acids and peptides) and a reduction in fractions B1 (true protein), B2 (cell wall-bound protein), and C (indigestible protein). These changes are characteristic of diets supplemented with NPN sources, such as urea.

The higher concentration of fraction B3, corresponding to slowly degraded proteins, observed in sugarcane can be attributed to the specific composition of this culture, characterized by a lower proportion of leaves and a higher stem/leaf ratio. In contrast, BRS Capiaçu, with a higher proportion of leaves, has lower B3 contents, since leaves have a lower concentration of cell walls compared to stems.

The reduction in B3 levels observed with urea addition can be explained by the solubilizing action of urea, which increases the soluble fraction and decreases the insoluble fraction. This observation is corroborated by the data presented in Table 7.

The analysis of fraction C, corresponding to indigestible protein, revealed a significant effect ($P < 0.05$) of both the bulk feed type and urea addition. Diets containing BRS Capiaçu showed the highest fraction C contents, which is consistent with the highest acid detergent insoluble nitrogen (ADIN) values observed in Table 6, reflecting the structural characteristics of this culture.

Urea addition resulted in a significant reduction of fraction C, with values of 5.3% for supplemented diets, compared to 6.4% for diets without urea (Table 7). This positive effect of urea, which solubilizes part of the unavailable nitrogen, contributes to improving the composition and nutrient availability in the diets.

Table 7. Protein fractionation of total diet silages of different bulky feeds with or without urea

Variable	Bulky		Urea		CV	*P*- value		
	Cana	Capiaçu	Sem	Com		Bulky	Urea	Bulky*Urea
Total nitrogen	2.3	2.2	2.2	2.3	7.7	0.0652	0.2143	0.1363
Fraction A [2]	50.1	52.1	45.4	56.2	12.7	0.3773	0.0000	0.7561
Fraction B1+B2[2]	35.3	35.7	37.6	33.4	19.6	0.8278	0.0439	0.9749
Fraction B3[2]	9.52	6.27	10.7	5.13	23.7	0.0000	0.0000	0.9170
Fraction C[2]	5.37	6.31	6.42	5.27	13.1	0.0005	0.0000	0.0571

[1]Percentage of dry matter; [2]Percentage of total nitrogen.

The carbohydrate fractionation analysis revealed the absence of a significant interaction ($P > 0.05$) between the studied factors, as shown in Table 8. However, significant effects ($P < 0.05$) of the isolated factors, bulk feed and urea, were observed on fractions A+B1, B2, and C.

Table 8. Carbohydrate fractionation of total diet silages of different bulky feeds with or without urea

Variable	Bulky		Urea		CV	*P*- value		
	Cane	Capiaçu	Without	With		Bulky	Urea	Bulky*Urea
Total carbohydrates	77.1	76.3	76.7	76.8	2.09	0.1321	0.7409	0.8388
Fraction A+B1	51.5	44.3	45.4	50.3	7.00	0.0000	0.0001	0.4482
Fraction B2	33.3	38.5	37.1	34.8	6.10	0.0000	0.0032	0.7489
Fraction C	15.2	17.1	17.5	14.8	13.1	0.0063	0.0003	0.3776

[1]Percentage of dry matter; [2]Percentage of total carbohydrates.

Diets containing sugarcane showed the highest A+B1 fraction contents, which can be justified by the higher concentration of soluble carbohydrates in this culture, compared to BRS Capiaçu. Carvalho et al. (2007) highlight the importance of high A+B1 fraction concentrations, since diets rich in this fraction are considered excellent energy sources for rumen microorganisms, positively influencing fermentation end products and, consequently, animal performance. Urea addition also resulted in higher A+B1 fraction values.

In contrast, diets containing BRS Capiaçu and those without urea addition showed the highest contents of fractions B2 and C, the opposite behavior to that observed for fraction A+B1, which represents soluble carbohydrates. The higher concentration of fractions B2 and C in diets with BRS Capiaçu is consistent with the high NDF contents observed in Table 6, confirming the presence of higher hemicellulose contents. Therefore, an inverse relationship is observed between soluble carbohydrate contents (fraction A) and fractions B2 and C.

CONCLUSIONS

The inclusion of urea in total diet silages promotes a restructuring of carbohydrate and protein fractions, resulting in an improvement in the nutritional value of the ensiled feed. Regarding the evaluated forages, sugarcane demonstrated superior performance in terms of composition, fermentation, and conservation when compared to BRS Capiaçu. These results demonstrate the potential of sugarcane as an advantageous component for the production of total diet silages.

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