



*Research article*

## **Nutritional parameters of sheep fed diets based on grass silage with brewery residue and a concentrate with rice bran**

**Anderson de Moura Zanine<sup>1</sup>, Breno de Moura Gimenez<sup>2</sup>, Daniele de Jesus Ferreira<sup>1</sup>, Michelle de Oliveira Maia Parente<sup>3</sup>, Henrique Nunes Parente<sup>1</sup>, Edson Mauro Santos<sup>4</sup>, Glayciane Costa Gois<sup>1,\*</sup>, Fleming Sena Campos<sup>1</sup>, Luana Milena Pinheiro Rodrigues<sup>1</sup>, Jessica Maria de Sousa Oliveira<sup>1</sup>, Nelquides Braz Viana<sup>1</sup>, Daimy Salas Aguilar<sup>1</sup>, George de Sousa Lima Paiva<sup>1</sup>, and Dilier Olivera Vicedo<sup>5</sup>**

<sup>1</sup> Postgraduate Program in Animal Science, Universidade Federal do Maranhão, Chapadinha, MA, 65500-000, Brazil

<sup>2</sup> Postgraduate Program in Animal Science, Universidade Federal de Mato Grosso, Cuiabá, MT, 78060-900, Brazil

<sup>3</sup> Postgraduate Program in Tropical Animal Science, Universidade Federal do Piauí, Teresina, PI, 64049-550, Brazil

<sup>4</sup> Postgraduate Program in Animal Science, Universidade Federal da Paraíba, Areia, PB, 58397-000, Brazil

<sup>5</sup> Institute of Agrifood, Animals and Environmental Sciences, Universidad de O'Higgins, Ruta 90 km 3, San Fernando, Chile

\* **Correspondence:** Email: [glayciane\\_gois@yahoo.com.br](mailto:glayciane_gois@yahoo.com.br); Tel: +9832729902.

**Abstract:** The objective was to evaluate the intake, digestibility, and feeding behavior of sheep receiving diets based on marandu grass silage (MGS) with different levels of dehydrated brewery residue (DBR) inclusion and different types of concentrates. Sixteen sheep ( $30 \pm 1.46$  kg and 12 months old) were distributed in a randomized block experimental design. The treatments were: MGS containing 10% DBR + concentrate (100% corn); MGS containing 10% DBR + concentrate (50% corn and 50% rice bran); MGS containing 30% DBR + concentrate (100% corn); MGS containing 30% DBR + concentrate (50% corn and 50% rice bran), with 4 replicates per treatment. The experimental period lasted 21 days. Regardless of the concentrate used, diets containing MGS + 30% DBR provided the animals with higher intakes and digestibility of dry matter (DM) and nutrients, water intake, and urinary pH ( $p < 0.05$ ). Longer feeding and rumination times and periods, and shorter idle times, feeding

efficiency of DM, feeding and rumination efficiency of neutral detergent fiber (NDF), and intakes of DM and NDF per meal were shown by sheep-fed diets containing MGS + 10% DBR ( $p < 0.05$ ). The use of 100% corn concentrate also resulted in lower rumination efficiency of NDF and a higher number of mastic chews associated with MGS + 10% DBR ( $p < 0.05$ ). Diets containing MGS + 30% DBR and concentrate (50% corn + 50% rice bran) resulted in shorter rumination and total chewing times (in min/kg/NDF) ( $p < 0.05$ ). The use of MGS ensiled with 30% BR in sheep diets improves dry matter intake and nutrient digestibility.

**Keywords:** agro-industrial co-products; alternative feeds; ruminants

## 1. Introduction

To make the sheep production chain competitive and sustainable in the market, research is needed to focus on the use of feeds that meet the animals' nutritional requirements while reducing production system costs [1]. One commonly adopted technology to cope with off-season periods is the conservation of food produced during the rainy season [2,3]. Therefore, the practice of animal confinement, combined with dietary planning, becomes an effective strategy to optimize animal production.

According to Ishangulyyev et al. [4], approximately 1.3 billion tons of agro-industrial residues are generated globally each year. This amount would be sufficient to tackle the global challenge of meeting the growing demand for food, which could reach around 160% of the current demand by the year 2050. The use of agro-industrial waste in feed formulation can significantly reduce feeding costs, potentially accounting for up to 80% of the total cost in confined systems [5,6]. Therefore, the use of agro-industrial by-products as ingredients in diets for small ruminants is an option that should be considered by the livestock sector, transforming low-value raw materials into high-quality, low-cost animal feed that does not compete with human food [7].

The utilization of agro-industrial waste in sheep confinement systems can contribute to environmental conservation by providing a better destination for these wastes as food and nutritional inputs [8]. Brewery residue and rice bran are readily available during drought periods in Brazilian regions [9,10], both having great potential for use in animal feeding due to their considerable concentrations of protein and energy, characterizing them as protein and/or energy feeds, capable of replacing soybean meal and corn in the diets offered to small ruminants [11,12].

However, brewery residue has a high moisture content, which, according to Terefe [13], can lead to rapid deterioration and environmental problems after 7 to 10 days of storage. According to Dentinho et al. [7], in agro-industrial wastes with high moisture content, it becomes essential to apply preservation methods to stabilize the product and mitigate seasonal availability. An interesting alternative for utilizing wet brewery residue is its use in a dehydrated form, as an additive for grass silages, reducing losses from gases and effluents, and improving the fermentative profile and nutritive value of the silage, as observed by Ferro et al. [14], Silva et al. [15], and Dai et al. [16]. Regarding rice bran, studies demonstrate that its use has been viable in feeding small ruminants, with this bulky supplement providing dry matter and nutrient intakes similar to corn [10].

However, for these residues to be efficiently used in animal production, it is necessary to evaluate the intake, chemical-bromatological composition, and digestibility of the feed [17]. The feed will only be utilized by the animal if consumed and digested in sufficient quantity to meet its nutritional

requirements [18]. Thus, we hypothesize that the combination of marandu grass with brewery residue in silage production, along with the provision of rice bran as an energy concentrate for confined sheep, will increase dry matter intake and nutrient digestibility while reducing feeding time.

In this context, the aim was to evaluate the intake, digestibility, and feeding behavior of sheep fed diets based on grass silage with different levels of dehydrated brewery residue inclusion and different types of concentrates.

## 2. Materials and methods

### 2.1. Ethical aspects and location of the experiment

The use of animals in this study was approved and certified by the Animal Use Ethics Committee of UFMT (protocol no. 23108.046399/13-4).

The experiment was conducted in the forage crops sector experimental area in the Department of Animal Science, Federal University of Mato Grosso – UFMT, Rondonópolis Campus, Mato Grosso, Brazil (16° 28' South Latitude, 50° 34' West Longitude, 270 m altitude). The region is characterized by an Aw-type tropical climate, with well-defined dry and rainy seasons, hot and humid summers, and cold and dry winters. The average annual temperature is 27.5 °C, with a relative humidity of 60% and average annual precipitation of 1240 mm.

### 2.2. Animals, facilities, and the experimental period

The confinement was carried out in a hollow shed (without side walls), with a ceiling height of 2.5 m, a beaten floor, and covered with metal tiles. Sixteen crossbred Santa Inês lambs, uncastrated males, with an initial average body weight of  $30 \pm 1.46$  kg (mean  $\pm$  standard deviation) and an average age of  $12 \pm 2$  months, were used in the experiment. The animals were previously identified, weighed, treated for endo- and ectoparasites, and distributed in individual pens (1.5 m<sup>2</sup>) equipped with individual feeders, drinkers, and a salt trough. The confinement period lasted 21 days, with 14 days for adaptation to the diets and seven for data collection.

### 2.3. Silage preparation

The marandu grass (*Brachiaria brizantha* cv. Marandu syn. *Urochloa brizantha* cv. Marandu) used in the ensiling process was from an established experimental area, harvested 60 days after regrowth, and cut 5 cm above the ground. The grass was processed in a stationary forage chopper (PP-35, Pinheiro Machines, Itapira, São Paulo, Brazil) to an average particle size of approximately 2.0 cm. The wet brewery residue was sun-dried for 36 hours, reaching a dry matter (DM) content of 90%. The marandu grass silages (MGS) containing 10% and 30% dehydrated brewery residue (DBR) were prepared in 200 L plastic drum silos, with removable lids sealed with a metal ring. Once closed, the silos were kept in a covered shed, where they remained for 45 days. After opening, silage samples were collected, with the top layer (10 cm) discarded from each silo. The silage was removed manually and collected in plastic containers for later chemical analysis (Table 1). The analyses were performed in triplicate.

#### 2.4. Design, treatments, and experimental diets

A randomized block experimental design was adopted, constituting four treatments with four animals per treatment. The initial body weight of the animals was used to define the blocks. The treatments were defined as: MGS containing 10% DBR + concentrate with 100% corn; MGS containing 10% DBR + concentrate containing 50% corn and 50% rice bran; MGS containing 30% DBR + concentrate with 100% corn; and MGS containing 30% DBR + concentrate containing 50% corn and 50% rice bran.

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

Items (%)	Ingredients						
	MGS + 10% DBR	MGS + 30% DBR	DBR	Ground corn	Cottonseed meal	Soybean meal	Rice bran
DM	27.91	39.50	89.96	90.98	89.05	90.02	90.6
Ash	7.13	6.52	-	1.30	3.71	8.16	5.97
CP	11.39	13.50	29.92	8.75	26.39	44.15	13.98
EE	5.08	5.29	6.52	8.95	13.48	2.44	20.78
NDF	64.06	61.60	60.75	10.25	28.87	21.75	24.13
NDF	30.28	32.30	30.09	4.79	47.40	31.40	32.55
TC	76.40	74.69	-	81.00	56.42	45.25	59.27
NFC	12.34	13.09	-	70.75	27.55	23.50	35.14

MGS: Marandu grass silage; DBR: Brewery residue; DM: Dry matter; CP: Crude protein; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; TC: Total carbohydrates; NFC: Non-fibrous carbohydrates.

The ingredients used for the experimental diets included marandu grass silage containing 10% brewery residue, marandu grass silage containing 30% brewery residue, ground corn, cottonseed meal, soybean meal, rice bran, and urea (Table 1). The diets were formulated in a roughage:concentrate ratio of 50:50, based on dry matter (Table 2), and balanced to allow for daily gains of 200 g [19].

#### 2.5. Intake and apparent digestibility

The diet was provided daily at 08:00 and 16:00, and water was offered *ad libitum*. The mixing of the roughage and the concentrate was carried out at the time of feeding. The amount of feed offered was calculated based on the previous day's intake, allowing for up to 15% leftovers. Dry matter intake (DMI) and nutrient intake were determined as the difference between the total dry matter and nutrients present in the consumed diet and in the leftovers.

Water intake was evaluated daily. Water was provided in buckets and weighed before and after a 24-hour period. Three buckets containing water were distributed in the shed, near the animal pens, to determine daily evaporation.

The digestibility trial, using the total feces collection method, was conducted over 6 days. Feces were sampled using collection bags attached to the animals before the sampling period. The bags were weighed and emptied twice a day. A 10% subsample of the total feces from each animal per treatment was collected and stored at  $-20^{\circ}\text{C}$  for later analysis. Urine samples were collected by natural micturition or forced by interrupting respiration through nostril occlusion for 10 to 20 seconds [20].

Urine pH was measured with a portable pH meter, previously calibrated in buffer solutions of pH 4.0 and pH 7.0.

Rumen fluid was collected using an esophageal probe, utilizing a flexible hose, rounded at the tip, with a completely open orifice at the end and no holes on the sides, connected to a vacuum pump. Between collections, the hose was washed and lubricated with petroleum jelly. The rumen fluid was filtered through gauze and the pH was measured [21].

**Table 2.** Proportion of ingredients and the chemical composition of experimental diets.

Ingredients (kg)	MGS + 10% BR		MGS + 30% BR	
	100% corn	50% corn + 50% rice bran	100% corn	50% corn + 50% rice bran
	Concentrate composition (%)			
Ground corn	78.78	39.38	78.75	39.38
Cottonseed meal	0.0	6.19	7.68	11.25
Soybean meal	10.31	4.5	3.38	0.0
Rice bran	0.0	39.38	0.0	39.38
Urea	0.93	0.57	0.19	0.0
	Concentrate chemical composition (% dry matter)			
Dry matter*	91.08	91.64	91.22	91.72
Crude protein	12.53	12.37	12.54	12.47
Ether extract	18.32	20.17	21.34	28.77
Ash	1.51	4.34	1.41	5.50
Neutral detergent fiber	29.62	38.12	28.96	38.67
Acid detergent fiber	14.99	33.21	17.66	31.66

MGS: Marandu grass silage; BR: Brewery residue; \* in % dry matter.

## 2.6. Feeding behavior

The feeding behavior of the animals were evaluated through visual observation [22], starting at eight in the morning. Feeding (A), rumination (R), and idleness (O) behaviors were evaluated using instantaneous and continuous sampling, according to the focal sampling method and sampling intervals of 10 minutes, during 24 hours. The observations were carried out by four trained observers recording animal behavior data, minimizing interference whenever possible. Each observer was responsible for recording the activities of 4 animals (1 observer per treatment). Prior to the analysis, the animals were subjected to nighttime artificial lighting for adaptation. The discretization of the time series was done by counting the discrete periods of feeding, rumination, and idleness. The average duration of each of the discrete periods was obtained by dividing the daily times of each activity by the number of discrete periods of the same activity [23].

The number of mastic chews per bolus (MCPB; n°/bolus), the number of mastic chews per day (MCPD; n°/day), and the time of the mastic chews per ruminated bolus (MCTB; sec/bolus) were measured at three different times of the day, using a digital stopwatch [24]. To obtain the average chewing times, observations of the ruminal boluses were made every 30 minutes, within the 24 hours of evaluation. The time and number of chewing times for each ruminal bolus per animal were

computed within this time period.

Feeding (FE) and rumination (RU) efficiencies for dry matter (DM; g DM ingested/h) and neutral detergent fiber (NDF; g NDF ingested/h) were estimated according to Bürger et al. [25]. The number of feeding, rumination, and idleness periods were counted by the number of observed activity sequences. The average daily duration of these activity periods was calculated by dividing the total duration of each activity (feeding, rumination, and idleness in min/day) by the respective number of discrete periods.

### 2.7. Laboratory analyses

Samples of the offered feed, leftovers, and feces were pre-dried in a forced-air oven (65 °C; 72 h) and then processed in a knife mill (Wiley Mill, Marconi, MA-580, Piracicaba, Brazil) using a 1 mm sieve. Analyses were conducted using the methods described by Silva and Queiroz [26] to determine the contents of the dry matter (DM), ash, crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), and acid detergent fiber (ADF). The analyses were performed in triplicate.

Total carbohydrates (TC) were obtained according to Sniffen et al. [27]:

$$TC = 100 - [CP + EE + MM] \quad (1)$$

Non-fibrous carbohydrates (NFC) for diets containing urea were estimated according to Hall [28]:

$$NFC = 100 - [\%ash + \%EE + \%NDF + (\%CP - \%CP_{urea} + \%Urea)] \quad (2)$$

In urea-free diets, NFC were obtained according to Weiss [29]:

$$NFC = 100 - (CP + NDF + EE + ash) \quad (3)$$

The apparent digestibility coefficient (ADC) of the nutrients was calculated using the equation:

$$ADC(\%) = [(\text{nutrient ingested}(g) - \text{nutrient excreted in feces}(g))/\text{nutrient ingested}(g)] * 100 \quad (4)$$

### 2.8. Statistical analyses

The data were subjected to the Shapiro-Wilk and Levene tests to verify the normality of the residues and homogeneity of the variances, respectively; once the premises were met, they were subjected to ANOVA using the Statistical and Genetic Analysis System—SAEG [30]. Significant probability values were those below 5% using Tukey's test. The following statistical model was used:

$$Y_{ij} = \mu + b_j + t_i + e_{ij} \quad (5)$$

where:  $Y_{ij}$  = Observed value of the variable;  $\mu$  = Overall mean;  $b_j$  = Block effect;  $t_i$  = treatment effect; and  $e_{ij}$  = Residual error.

## 3. Results

Dry matter and nutrient intake, expressed in g/day and as a percentage of body weight, and water intake by animals receiving diets containing MGS + 10% DBR were lower ( $p < 0.05$ ) than those of animals receiving diets containing MGS + 30% DBR (Tables 3 and 4).

**Table 3.** Dry matter and nutrient intake of sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates based on crushed corn and crushed corn + rice bran.

Items	Silages		Concentrates		SEM Silages	SEM Concentrates	P value
	MGS 10% DBR	+ MGS 30% DBR	+ 100% corn	+ 50% corn + 50% rice bran			
Intake (g/day)							
DM	846.34 b	1095.84 a	939.68 a	1002.50 a	124.75	31.41	<0.001
OM	806.49 b	1041.96 a	906.74 a	941.71 a	117.73	17.48	<0.001
CP	145.05 b	197.81 a	166.81 a	176.06 a	26.38	4.62	<0.001
EE	112.82 b	177.98 a	127.49 a	153.31 a	32.58	12.91	<0.001
NDF	363.15 b	483.08 a	371.47 a	474.75 a	59.96	51.64	<0.001
ADF	137.23 b	241.79 a	189.51 a	328.84 a	52.28	69.66	<0.001
TC	598.17 b	716.53 a	651.56 a	663.14 a	59.18	5.79	<0.001
NFC	216.93 b	223.18 a	160.07 a	280.04 a	3.12	59.98	<0.001
Intake (% body weight)							
DM	2.75 b	3.78 a	3.12 a	4.42 a	0.51	0.65	<0.001
OM	2.62 b	3.59 a	3.00 a	3.21 a	0.48	0.10	<0.001
CP	0.47 b	0.68 a	0.56 a	0.60 a	0.10	0.02	<0.001
EE	0.36 b	0.61 a	0.42 a	0.56 a	0.12	0.07	<0.001
NDF	1.18 b	1.66 a	1.53 a	1.69 a	0.24	0.08	<0.001
ADF	0.71 b	1.03 a	0.62 a	1.12 a	0.16	0.25	<0.001
TC	0.19 b	0.25 a	0.22 a	0.23 a	0.03	0.005	<0.001
NFC	0.70 b	0.78 a	0.54 a	0.90 a	0.04	0.18	<0.001

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; DM: Dry matter; OM: Organic matter; CP: Crude protein; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; TC: Total carbohydrates; NFC: Non-fibrous carbohydrates; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

**Table 4.** Dry matter and nutrient intake of sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates.

Items	MGS + 10% DBR		MGS + 30% DBR		SEM	P value
	100% corn	50% corn + 50% rice bran	100% corn	50% corn + 50% rice bran		
Intake (g/day)						
DM	764.36 b	928.32 b	1115.00 a	1076.68 a	79.80	<0.001
OM	737.56 b	875.42 b	1075.92 a	1008.01 a	74.86	<0.001
CP	135.81 b	154.30 b	197.80 a	198.30 a	15.76	<0.001
EE	98.65 b	127.00 b	156.34 a	199.62 a	21.57	<0.001
NDF	291.57 b	434.72 b	451.57 a	514.79 a	47.13	<0.001
ADF	248.70 b	299.01 b	311.38 a	358.67 a	22.59	<0.001
TC	248.70 b	134.98 b	311.38 a	185.17 a	38.31	<0.001

*Continued on the next page*

Items	MGS + 10% DBR		MGS + 30% DBR		SEM	P value
	100% corn	50% corn + 50% rice bran	100% corn	50% corn + 50% rice bran		
Intake (g/day)						
NFC	553.26 b	643.00 b	773.02 a	662.04 a	45.13	<0.001
Water (L/day)	0.330 b	0.327 b	0.440 a	0.365 a	0.03	<0.001
Intake (% body weight)						
DM	2.40 b	3.09 b	3.81 a	3.74 a	0.33	<0.001
OM	2.33b	2.92 b	3.68 a	3.51 a	0.31	<0.001
CP	0.43 b	0.51 b	0.68 a	0.69 a	0.06	<0.001
EE	0.31 b	0.42 b	0.53 a	0.69 a	0.08	<0.001
NDF	0.91 b	1.45 b	1.52 a	1.79 a	0.18	<0.001
ADF	0.42 b	0.99 b	0.82 a	1.24 a	0.17	<0.001
TC	0.17 b	0.20 b	0.26 a	0.24 a	0.02	<0.001
NFC	0.78 b	0.46 b	1.08 a	0.62 a	0.13	<0.001

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; DM: Dry matter; OM: Organic matter; CP: Crude protein; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; TC: Total carbohydrates; NFC: Non-fibrous carbohydrates; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

Similarly, diets containing MGS + 10% DBR resulted in lower DM and nutrient digestibility coefficients and lower urine pH for the animals ( $p < 0.05$ ) (Tables 5 and 6).

**Table 5.** Apparent nutrient digestibility, urine pH, and ruminal fluid pH of sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates.

Items (%)	Silages		Concentrates		SEM Silages	SEM Concentrates	P value
	MGS + 10% DBR	MGS + 30% DBR	100% corn	50% corn + 50% rice bran			
DM	65.71 b	70.63 a	64.39 a	67.96 a	2.46	1.78	<0.001
OM	69.45 b	72.50 a	66.40 a	71.02 a	1.52	2.31	<0.001
CP	78.50 b	83.47 a	78.52 a	83.44 a	2.48	2.46	<0.001
EE	75.96 b	79.45 a	74.26 a	77.18 a	1.745	1.46	<0.001
NDF	62.91 b	67.65 a	64.42 a	66.14 a	2.37	0.86	<0.001
ADF	49.90 b	59.04 a	55.92 a	58.02 a	4.57	1.05	<0.001
TC	67.18 b	71.09 a	66.36 a	70.99 a	1.95	2.31	<0.001
NFC	73.03 b	77.91 a	72.31 a	79.37 a	2.44	3.53	<0.001

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; DM: Dry matter; OM: Organic matter; CP: Crude protein; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; TC: Total carbohydrates; NFC: Non-fibrous carbohydrates; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

There was no effect of the tested diets on rumen fluid pH ( $p > 0.05$ ; Table 6).



**Table 6.** Apparent nutrient digestibility of sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates.

Items (%)	MGS + 10% DBR		MGS + 30% DBR		SEM	P value
	100% corn	50% corn + 50% rice bran	100% corn	50% corn + 50% rice bran		
DM	59.53 b	60.90 b	68.15 a	67.00 a	2.16	<0.001
OM	61.48 b	60.17 b	65.10 a	65.50 a	1.32	<0.001
CP	52.93 b	52.00 b	56.47 a	57.58 a	1.35	<0.001
EE	46.74 b	53.36 b	56.59 a	62.25 a	3.24	<0.001
NDF	65.80 b	65.57 b	70.81 a	70.10 a	1.38	<0.001
ADF	46.82 b	44.83 b	52.72 a	53.29 a	2.11	<0.001
TC	63.60 b	62.33 b	66.31 a	65.21 a	0.88	<0.001
NFC	60.88 b	61.70 b	67.48 a	66.17 a	1.63	<0.001
pH urine	5.59 b	5.50 b	7.08 a	6.82 a	0.41	<0.001
pH ruminal fluid	6.66 a	6.32 a	6.82 a	6.87 a	0.12	>0.05

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; DM: Dry matter; OM: Organic matter; CP: Crude protein; EE: Ether extract; NDF: Neutral detergent fiber; ADF: Acid detergent fiber; TC: Total carbohydrates; NFC: Non-fibrous carbohydrates; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

**Table 7.** Feeding behavior of sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates.

Items	Silages		Concentrates		SEM Silages	SEM Concentrates	P value
	MGS + 10% DBR	MGS + 30% DBR	100% corn	50% corn + 50% rice bran			
Feeding (h)	3.70 a	2.85 b	3.38 a	3.35 a	0.42	0.01	<0.001
Rumination (h)	9.85 a	8.70 b	9.32 a	8.97 a	0.57	0.17	<0.001
Idleness (h)	10.45 b	12.45 a	11.20 a	11.68 a	1.00	0.24	<0.001
FE (g DM/h)	174.12 b	225.34 a	196.22 a	203.23 a	25.61	3.50	<0.001
FE (g NDF/h)	74.40 b	99.60 a	77.59 a	90.41 a	12.6	6.41	<0.001
RUE (g DM/h)	102.17 b	132.98 a	112.69 a	122.46 a	15.40	4.88	<0.001
RUE (g NDF/h)	44.15 b	58.53 a	44.07 b	58.07 a	7.19	7	<0.001
Meritic chews (n°/bolus)	58.04 a	54.40 b	58.18 a	54.32 b	1.82	1.93	<0.001
Meritic chews (n°/day)	4619.82 a	4461.50 b	48,821.64 a	41,986.66 b	3417.5	19,997.6	<0.001
Feeding period (n°/day)	23.06 a	19.50 b	24.31 a	22.25 a	1.78	1.03	<0.001
Rumination period (n°/day)	25.68 a	21.31 b	23.43 a	25.56 a	2.18	1.06	<0.001
Idleness period (n°/day)	26.50 b	29.37 a	28.43 a	29.43 a	1.43	0.5	<0.001
DMI/feeding (kg)	0.042 b	0.062 a	0.043 a	0.051 a	0.01	0.004	<0.001
NDFI/feeding (kg)	0.016 b	0.028 a	0.023 a	0.027 a	0.006	0.002	<0.001
DMI (min/kg)	146.71 b	163.09 a	152.87 a	160.92 a	8.19	4.02	<0.001
NDFI (min/kg)	337.89 b	398.42 a	393.69 a	420.60 a	30.26	13.45	<0.001

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; FE: Feeding efficiency; RUE: Rumination efficiency; DM: Dry matter; NDF: Neutral detergent fiber; DM: Dry matter intake; NDF: Neutral detergent fiber intake; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

The longest feeding and rumination times and the shortest idleness times were observed in sheep fed diets containing MGS + 10% DBR ( $p < 0.05$ ) (Table 7). This diet also resulted in the lowest efficiencies of DM and NDF feeding and NDF rumination, and the lowest DM and NDF intake per feeding, in min/kg ( $p < 0.05$ ) (Table 7).

The lowest NDF rumination efficiency was obtained by animals receiving diets containing MGS + 10% DBR ( $p < 0.05$ ), with the inclusion of a concentrate with 100% corn ( $p < 0.05$ ) (Table 7). Conversely, this diet led to a higher number of mastic chews per bolus and per day for the animals ( $p < 0.05$ ) (Table 7).

The shortest times spent per feeding, rumination, and idleness period, and total chewing time (min/kg/DM), were observed in animals receiving diets containing MGS + 30% DBR ( $p < 0.05$ ) (Table 8). The shortest rumination times (in g DM/bolus, g NDF/bolus, and min/kg/DM) were observed in animals receiving diets containing MGS + 10% DBR ( $p < 0.05$ ) (Table 8). Diets containing MGS + 30% DBR and a concentrate based on 50% corn + 50% rice bran resulted in shorter rumination and total chewing times (in min/kg/NDF) ( $p < 0.05$ ) (Table 8).

**Table 8.** Time spent on feeding, ruminating, idling, and chewing by sheep fed diets containing marandu grass silage associated with dehydrated brewery residue and different concentrates.

Items	Silages		Concentrates		SEM	SEM	P value
	MGS + 10% DBR	MGS + 30% DBR	100% corn	50% corn + 50% rice bran	Silages	Concentrates	
TSFEP (min)	7.97 a	6.39 b	7.83 a	8.13 a	0.79	0.15	<0.001
TSRUP (min)	10.75 a	8.56 b	10.19 a	9.84 a	1.09	0.17	<0.001
TSIP (min)	11.46 a	13.83 b	13.41 a	14.20 a	1.18	0.39	<0.001
Rumination (g DM/bolus)	1.34 b	1.02 a	1.20 a	1.31 a	0.16	0.05	<0.001
Rumination (g NDF/bolus)	0.60 b	0.46 a	0.53 a	0.61 a	0.07	0.04	<0.001
Rumination (min/kg/DM)	303.92 b	221.98 a	261.58 a	270.32 a	40.97	4.37	<0.001
Rumination (min/kg/NDF)	738.87 b	513.44 a	719.61 a	532.70 b	112.71	93.45	<0.001
Total chews (min/kg/DM)	435.11 a	378.16 b	413.63 a	399.65 a	28.47	6.99	<0.001
Total chews (min/kg/NDF)	1049.10 a	808.37 b	1033.44 a	823.18 b	120.36	105.13	<0.001

MGS: Marandu grass silage; DBR: Dehydrated brewery residue; TSFEP: Time spent per feeding period; TSRUP: Time spent per rumination period; TSIP: Time spent per idling period; DM: Dry matter intake; NDF: Neutral detergent fiber intake; SEM: Standard error of the mean. Means followed by different letters differ from each other by Tukey's test at a 5% probability.

## 4. Discussion

### 4.1. Intake and digestibility

The higher digestibility coefficients presented by the animals receiving silages containing 30% dehydrated brewery residue can be explained by the increased dry matter intake shown by the animals receiving this silage. However, all the evaluated diets provided animals with a dry matter intake exceeding the NRC [19] recommended intake of 570 g for sheep with a body weight of 30 kg and expected daily gain of 200 g. Additionally, all diets provided animals with crude protein intake above the requirement of 129 g/day for lambs with an average body weight of 30 kg as per the NRC [19].

The higher dry matter intake of marandu grass silage containing 30% brewery residue compared

to the silage containing 10% brewery residue can be mainly explained by the lower NDF content in the silage with 30% brewery residue (Table 1). This supports the observations described by Mertens [31] and Van Soest [32], who noted that neutral detergent fiber is one of the main factors controlling dry matter intake due to ruminal fill.

The results for NDF and ADF digestibility observed with the provision of marandu grass silage containing 30% brewery residue to the animals may be associated with the increased NFC intake obtained with this diet. Up to a certain point, increasing NFC in the diets can favor fiber digestion; however, beyond a certain point, it can reduce ruminal pH and increase the passage rate, reducing cellulolytic activity and consequently fiber digestibility [32]. However, ruminal pH was not affected by the tested diets. Therefore, the use of marandu silage associated with brewery residue at 30% levels serves as a resource to improve the digestibility of the bulky diet of sheep.

Another favorable factor of brewery residue is due to the presence of sugars (soluble and insoluble) in its composition, which, when combined with nitrogen compounds, form an extract in the rumen for the development of microorganisms, promoting good digestibility of the ingested feed [33, 34]. Consequently, when providing protein supplements associated with different levels of non-fibrous carbohydrates, the diet must be adjusted to the quantity and quality of the silage to ensure better utilization of these nutrients.

The similarity between the digestibility of the tested concentrates is due to the similarity between the ruminal digestion of rice bran and corn, which consequently increases the passage rate, reducing the retention time of digesta in the gastrointestinal tract, promoting greater exposure to digestive processes [35].

Water intake by ruminants is influenced by dry matter and crude protein intake, as it results in a higher water demand due to the caloric increment from the protein digestion process [36, 37]. This was observed in this study, as the increase in dry matter and crude protein intake obtained with the increment of 30% brewery residue in marandu grass silages led to a higher water intake by the animals.

The lower water intake by animals receiving diets containing marandu grass silages + 10% brewery residue possibly increased urinary density, leading to higher mineral excretion in the urine related to the diet composition, potentially causing the decrease in urine pH. Ferreira [35], when evaluating the association of different levels of dehydrated brewery residue (0, 10, 20, 30, and 40%) with marandu grass in silage composition, found that as the levels of brewery residue in the silages offered to sheep increased, the urine pH decreased. However, differing from this study, the author did not provide a concentrate to the animals, which may have influenced the responses obtained in this study. According to Singh et al. [38], urine pH depends on the animal's diet, and urinary acidification (pH lower than 5.5) must be avoided as it leads to a decrease in dry matter intake.

#### *4.2. Feeding behavior*

Sheep fed with marandu grass silage containing 10% brewery residue spent more time feeding and ruminating. This behavior may be associated with the compensatory mechanism of increasing energy increments through adjustments in ingestive behavior. In other words, as the energy supply of the silage decreased by using 10% dehydrated brewery residue in the grass silage, the more time the animals spent feeding and ruminating, and consequently, the less idle time they had.

Animals receiving diets containing marandu grass silage with 30% dehydrated brewery residue showed more idle time due to higher DM and NDF intake (g/day). According to Van Soest [32], the

fiber content and physical form of the diet are the main factors affecting rumination time. Since the diets containing 30% brewery residue had lower NDF content, the efficiency of rumination was affected, as the rumination period increases with the fiber content of the diet, reflecting the need for processing ruminal digesta to enhance digestive efficiency. This resulted in more idle time for the animals, meaning they were more efficient in feeding, spending less time eating or ruminating, and had longer idle periods.

Rumination activity, expressed in min/kg of DM and NDF, may have been influenced by the NDF content in the silages, which varied with the inclusion of dehydrated brewery residue. The difference between the levels of this fraction (2.46 percentage points) in the silages with 10% and 30% addition may have been sufficient to cause changes in rumination activities and chewing times. A factor that may have contributed to the effect on rumination is the small particle size of the dehydrated brewery residue, similar to that of concentrated feeds like ground corn and soybean meal.

## **5. Conclusions**

The use of silages based on marandu grass ensiled with 30% dehydrated brewery residue in sheep diets improves dry matter and nutrient intake as well as nutrient digestibility compared to using diets containing silages based on marandu grass ensiled with 10% dehydrated brewery residue.

### **Use of AI tools declaration**

The authors declare they have not used Artificial Intelligence (AI) tools in the creation of this article.

### **Acknowledgments**

The authors wish to thank the National Council for Scientific and Technological Development (CNPq), Coordination for the Improvement of Higher Education Personnel (CAPES) – Finance Code 001, Foundation for Research Support of the State of Mato Grosso (FAPEMAT), and Foundation to Support Research and Scientific and Technological Development of Maranhão (FAPEMA).

### **Conflict of interest**

The authors declare no conflicts of interest.

### **Authors contribution**

Anderson de Moura Zanine, Daniele de Jesus Ferreira, Michelle de Oliveira Maia Parente, Henrique Nunes Parente, Edson Mauro Santos, and Dilier Olivera Viciado: Conceptualization, Project administration, and Supervision; Breno de Moura Gimenez, Fleming Sena Campos, Luana Milena Pinheiro Rodrigues, Jessica Maria de Sousa Oliveira, Nelquides Braz Viana, Daimy Salas Aguilar, and George de Sousa Lima Paiva: Investigation, Data curation, Formal analysis, Investigation, Methodology; Glayciane Costa Gois: Writing—original draft, Writing—review and editing. All authors have read and agreed to the published version of the manuscript.

## References

1. Pulina G, Francesconi AHD, Stefanon B, et al. (2017) Sustainable ruminant production to help feed the planet. *Italian J Anim Sci* 16: 140–171. <https://doi.org/10.1080/1828051X.2016.1260500>
2. Balehegn M, Duncan A, Tolera A, et al. (2020) Improving adoption of technologies and interventions for increasing supply of quality livestock feed in low- and middle-income countries. *Global Food Sec* 26: e100372. <https://doi.org/10.1016/j.gfs.2020.100372>
3. Balehegn M, Ayantunde A, Amole T, et al. (2022) Forage conservation in sub-Saharan Africa: Review of experiences, challenges, and opportunities. *Agron J* 114: 75–99. <https://doi.org/10.1002/agj2.20954>
4. Ishangulyyev R, Kim S, Lee SH (2019) Understanding food loss and waste—Why are we losing and wasting food? *Foods* 8: e297. <https://doi.org/10.3390/foods8080297>
5. Castro WJR, Zanine AM, Souza AL, et al. (2016) Inclusão de diferentes níveis do resíduo de feijão nas rações de ovinos sobre o consumo e digestibilidade dos nutrientes. *Semina: Ci Agr* 37: 369–380. <https://doi.org/10.5433/1679-0359.2016v37n1p369>
6. Vastolo A, Calabro S, Cutrignelli MI (2022) A review on the use of agro-industrial co-products in animals' diets. *Italian J Anim Sci* 21: 577–594. <https://doi.org/10.1080/1828051X.2022.2039562>
7. Dentinho MTP, Paulos K, Costa C, et al. (2023) Silages of agro-industrial by-products in lamb diets—Effect on growth performance, carcass, meat quality and *in vitro* methane emissions. *Anim Feed Sci Techn* 298: e115603. <https://doi.org/10.1016/j.anifeedsci.2023.115603>
8. Böck MJ, Simões RR, Rici REG, et al. (2023) Morphological aspects of rumen papillae of lambs fed agro-industrial wastes. *Anim Sci J* 94: e13897. <https://doi.org/10.1111/asj.13897>
9. Onofre SB, Bertoldo IC, Abatti D, et al. (2018) Physiochemical Characterization of the brewers' spent grain from a brewery located in the Southwestern region of Paraná—Brazil. *Int J Adv Eng Res Sci* 5: 18–21. <https://dx.doi.org/10.22161/ijaers.5.9.3>
10. Vargas JAC, Mezzomo R, Gomes DI, et al. (2020) Total and partial replacement of corn meal with rice bran in lamb rations: Nutritional effects. *Liv Sci* 234: e103986. <https://doi.org/10.1016/j.livsci.2020.103986>
11. Teixeira WS, Carvalho S, Manzoni VG, et al. (2024) Intake, performance and ingestive behaviour in lambs finished in confinement with wet brewery residue used as roughage. *Ci Rural* 54: e20230089. <http://doi.org/10.1590/0103-8478cr20230089>
12. Yoshihara Y, Yokoyama S (2021) Effects of soybean curd residue and rice bran on lamb performance, health, and meat quality. *Vet Anim Sci* 11: e100166. <https://doi.org/10.1016/j.vas.2021.100166>
13. Terefe G (2022) Preservation techniques and their effect on nutritional values and microbial population of brewer's spent grain: A review. *CABI Agric Biosci* 3: 1–8. <https://doi.org/10.1186/s43170-022-00120-8>
14. Ferro MM, Zanine AM, Castro WJR, et al. (2017) Cinética de fermentação ruminal *in vitro* de silagem de cana-de-açúcar com resíduo de cervejaria desidratado. *Arch Zootec* 66: 237–242.
15. Silva ARP, Dias FJ, Rufino JPF, et al. (2020) Effect of using inoculant on elephant grass silage with aditives. *Acta Scient Animal Sci* 42: e50533. <https://doi.org/10.4025/actascianimsci.v42i1.50533>
16. Dai T, Wang J, Donga D, et al. (2022) Effects of brewers' spent grains on fermentation quality, chemical composition and *in vitro* digestibility of mixed silage prepared with corn stalk, dried apple pomace and sweet potato vine. *Italian J Anim Sci* 21: 198–207. <https://doi.org/10.1080/1828051X.2021.2022994>

17. Ferreira DJ, Zanine AM, Lana RP, et al. (2017) Intake and digestibility in sheep fed on marandu grass silages added with dehydrated barley. *Pesq Vet Bras* 37: 171–178. <https://doi.org/10.1590/S0100-736X2017000200012>
18. Carmo TD, Barbosa PM, Geraseev LC, et al. (2018) Intake and digestibility of lamb fed diets containing banana crop residues. *Pesq Agropec Bras* 53: 197–205. <https://doi.org/10.1590/S0100-204X2018000200008>
19. NRC (2007) National Research Council. Nutrient requirements of small ruminants: sheep, goats, cervids, and new world camelids, 7th Ed. Washington, DC, USA: The National Academy Press.
20. Santarosa BP, Ferreira DOL, Rodrigues MMP, et al. (2016) Avaliação clínica, laboratorial e anatomopatológica do sistema urinário de ovinos confinados com ou sem suplementação de cloreto de amônio. *Pesq Vet Bras* 36: 1–12. <https://doi.org/10.1590/S0100-736X2016000100001>
21. Souza MV, Barcellos AR (1993) Avaliação do fluido rumenal de bovinos e ovinos criados em regime de pastagem. *Ci Rural* 23: 31–36. <https://doi.org/10.1590/S0103-84781993000100007>
22. Martin P, Bateson P (1993) *Measuring Behavior*, 2nd Ed. Cambridge, UK: Cambridge University Press.
23. Silva LV, Vilela GKSM, Rocha KS, et al. (2024) Feeding behavior, water intake, and physiological parameters of feedlot lambs fed with diets containing babassu oil associated with sunflower oil blend. *Vet Med Int* 2024: e8673922. <https://doi.org/10.1155/2024/8673922>
24. Polli VA, Restle J, Senna DB, et al. (1996) Aspectos relativos à ruminação de bovinos e bubalinos em regime de confinamento. *Rev Bras Zootec* 25: 987–993. <https://doi.org/10.1590/S0103-84781995000100025>
25. Bürger PJ, Pereira JC, Queiroz AC, et al. (2000) Ingestive behavior in Holstein calves fed diets with different concentrate levels. *Rev Bras Zootec* 29: 236–242. <https://doi.org/10.1590/S1516-35982000000100031>
26. Silva DJ, Queiroz AC (2002) *Análise de alimentos: métodos químicos e biológicos*, 3rd Ed. Viçosa, BR: UFV.
27. Sniffen CJ, O'Connor JD, Van Soest PJ (1992) A net carbohydrate and protein system for evaluating cattle diets: II. Carbohydrate and protein availability. *J Anim Sci* 70: 3562–3577. <https://doi.org/10.2527/1992.70113562x>.
28. Hall MB (2003) Challenges with non-fiber carbohydrate methods. *J Anim Sci* 81: 3226–3232. <https://doi.org/10.2527/2003.81123226x>.
29. Weiss WP (1993) Energy prediction equations for ruminant feeds. In: *Cornell Nutrition Conference Feed Manufactures*, 61 Ithaca Proceedings, Ithaca: Cornell University, 176–185.
30. Ribeiro Jr JI (2007) *Análises estatísticas no SAEG (Sistema para análises estatísticas)*. Viçosa, BR: UFV.
31. Mertens DR (1994) Regulation of forage intake, In: Fahey GC (Ed.), *Forage Quality, Evaluation and Utilization*, Madison: Asa-Cssasssa, 450–493. <https://doi.org/10.2134/1994.foragequality.c11>
32. Van Soest PJ (1994) *Nutritional ecology of the ruminant*. 2nd ed. Ithaca, New York: Cornell University Press.
33. Rachwał K, Waśko A, Gustaw K, et al. (2020) Utilization of brewery wastes in food industry. *PeerJ* 8: e9427. <http://doi.org/10.7717/peerj.9427>
34. Castro MMD, Cardoso MA, Detmann E, et al. (2021) Marcondes. *In vitro* ruminal fermentation and enteric methane production of tropical forage added nitrogen or nitrogen plus starch. *Anim Feed Sci Techn* 275: e114878. <https://doi.org/10.1016/j.anifeedsci.2021.114878>

35. Ferreira DJ (2013) Resíduo desidratado da agroindústria de cervejaria na 2013 produção de silagem de capim-marandu. 126 f. Tese (Doutorado em Zootecnia) Universidade Federal de Viçosa, Viçosa, MG.
36. Palhares JCP, Novelli TI, Morelli M (2020) Best practice production to reduce the water footprint of dairy milk. *Rev Amb Água* 15: e2454. <https://doi.org/10.4136/1980-993X>
37. Pereira AL, Parente MOM, Zanine AM, et al. (2022) Physiological responses, water consumption, and feeding behaviour of lamb breeds fed diets containing different proportions of concentrate. *J Anim Behav Biometeorol* 10: e2206. <http://dx.doi.org/10.31893/jabb.22006>
38. Singh T, Amarpal A, Kinjavdekar P, et al. (2007) Blood acid-base and electrolyte changes following oral administration of ammonium chloride in goats suffering from obstructive urolithiasis. *Indian J Anim Sci* 77: 745–748.



AIMS Press

© 2024 the Author(s), licensee AIMS Press. This is an open access article distributed under the terms of the Creative Commons Attribution License (<https://creativecommons.org/licenses/by/4.0>)