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Carcass characteristics and visceral histomorphometric parameters of lambs fed diets with reallocated sorghum silage and increasing levels of concentrate. Características de carcaça e parâmetros histomorfométricos viscerais de cordeiros alimentados com dietas contendo silagem de sorgo reconstituída e níveis crescentes de concentrado.

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Abstract
The objective of this study was to evaluate diets containing reconstituted sorghum silages with increasing levels of concentrate on carcass characteristics and cellular morphometry of the small intestine, kidney, and liver at lamb finishing. Twenty-eight uncastrated, crossbred lambs with an average body weight of 18.46 ± 2.3 kg were used. The experimental treatments consisted of increasing concentrate levels: 430, 660, 810, and 910 g/kg. The experimental design was completely randomized. A quadratic effect was observed. ($p < 0.05$) on dry matter intake [g/100g body weight (BW)], BW at slaughter, and consequently, cold carcass weight and weight of the following commercial cuts: leg, loin, and shoulder. However, dressing percentage increased linearly ($p < 0.05$), while yields of commercial carcass cuts and non-carcass components did not change ($p > 0.05$) with experimental diets. Increasing levels of concentrate reduced ($p < 0.05$) liver glycogen scores but increased hyaline cylinders, necrosis, and inflammation in kidneys. The increasing levels of concentrate also increased ($p < 0.05$) epithelium thickness, keratinized portion, height of papillae, and papillae width of the rumen, but reduced goblet cells (GC) in the rumen. In the small intestine, a quadratic effect ($p > 0.05$) was observed on morphological characteristics such as submucosal thickness, mucosal thickness, and GC. Reallocated sorghum silage is an excellent fiber source for finishing lambs when included in diets with concentrate proportions up to 810 g/kg to maximize dressing percentage without damaging kidney histological parameters.
Keywords: Dressing percentage. Fat deposition. Glycogen store. Histological characteristics.

Resumo
O objetivo deste estudo foi avaliar dietas contendo silagens de sorgo reconstituídas com níveis crescentes de concentrado sobre as características de carcaça e a morfometria celular do intestino delgado, rins e fígado durante a terminação de cordeiros. Foram utilizados vinte e oito cordeiros mestiços, não castrados, com peso corporal médio de 18,46 ± 2,3 kg. Os tratamentos experimentais consistiram em níveis crescentes de concentrado: 430, 660, 810 e 910 g/kg. O delineamento experimental foi inteiramente casualizado. Observou-se efeito quadrático ($p < 0,05$) sobre o consumo de matéria seca [g/100 g de peso corporal (PC)], o PC ao abate e, consequentemente, o peso da carcaça fria e o peso dos seguintes cortes comerciais: pernil, lombo e paleta. Entretanto, o rendimento de carcaça aumentou linearmente ($p < 0,05$), enquanto os rendimentos dos cortes comerciais da carcaça e dos componentes não carcaça não se alteraram ($p > 0,05$) com as dietas experimentais. O aumento dos níveis de concentrado reduziu ($p < 0,05$) os escores de glicogênio hepático, mas aumentou a ocorrência de cilindros hialinos, necrose e inflamação nos rins. Os níveis crescentes de concentrado também aumentaram ($p < 0,05$) a espessura do epitélio, a porção queratinizada, a altura das papilas e a largura das papilas do rúmen, porém reduziram as células caliciformes (CC) no rúmen. No intestino delgado, observou-se efeito quadrático ($p > 0,05$) sobre características morfológicas como espessura da submucosa, espessura da mucosa e CC. A silagem de sorgo reconstituída é uma excelente fonte de fibra para a terminação de cordeiros quando incluída em dietas com proporções de concentrado de até 810 g/kg, maximizando o rendimento de carcaça sem causar danos aos parâmetros histológicos renais.
Palavras-chave: Rendimento de carcaça. Deposição de gordura. Reserva de glicogênio. Características histológicas.

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Introduction

Sheep farmers around the world have recently been looking to sell larger, higher-quality lambs. Consequently, diets with high levels of concentrate have been used to increase dietary energy density and reduce the age at which lambs are slaughtered (KARACA et al., 2016). However, one of the challenges is balancing heavy slaughter weights with a desirable amount of fat thickness (JABOREK et al., 2017), as greater concentrate levels result in fatter lambs compared to those fed more forage (BORTON et al., 2005; KARACA et al., 2016; LADEIRA et al., 2016).

The challenge in raising small ruminants is to determine the ideal forage-to-concentrate ratio that reduces the slaughter age while maintaining a balanced rumen bacterial population. For ruminant species, the presence of fibrous carbohydrates, as contained in sorghum silage, is essential to maintain the balance of the rumen bacterial population. However, in small ruminant production, there is little information about fiber requirements, unlike what is observed with dairy cattle (NRC, 2021) and beef cattle (NRC, 2016).

Recently, there has been a growing interest in cultivating sorghum for silage, especially in areas with rainfall irregularities, due to its greater efficiency in water usage and biomass yield compared to corn (FERNANDES et al., 2020). Sorghum silage has great potential and is an excellent source of fibrous carbohydrates, replacing corn silage without causing undesirable effects on animal performance and carcass characteristics when compared to corn silage (WU et al., 2021).

In animal production systems, one of the biggest challenges is silage conservation logistics, regardless of the crop used, which requires meeting increased marketing demands without high losses. Currently, there is a growing practice of selling silage to animal production farms. This involves producing forage in more suitable areas and transporting it to livestock properties, allowing for the sale of roughage between properties to meet demand. Thus, the concept of reallocated silage involves a practice that includes unloading, transport, recompression, and sealing by producers, facilitating herd feeding planning in the face of climatic variations and forage production seasonality (ANJOS et al., 2018).

The reallocation of silage is a widely adopted practice on Brazilian farms, aiming to meet the high demand for forage in livestock production (ANJOS et al., 2018). However, there is limited information in the literature regarding reallocated silage that take into account the characteristics of tropical climates (ANJOS et al., 2018; LIMA et al., 2020; SILVA et al., 2025). Therefore, the aim of this research was to evaluate diets containing relocated sorghum silages with increasing levels of concentrate on the carcass characteristics and cellular morphometry of the small intestine, kidney, and liver in finishing lambs.

Materials and methods

Location, animals and experimental facilities

All procedures involving animals were approved by the Animal Ethics Committee of the Federal University of Paraíba, under protocol no. 5391070619. The experiment was conducted at the Animal Requirement and Metabolism Laboratory, Center of Agrarian Sciences, Federal University of Vale do São Francisco, located in Petrolina, PE, Brazil. The city is located in the Mesoregion of São Francisco in Pernambuco and the microregion of Petrolina, at latitude 9º 23' 34" South, longitude 40º 30' 28" West, and an altitude of 376 m. The climate is classified as BSh-type hot semi-arid according to the Köppen-Geiger classification, with average annual rainfall of 443 mm, characterized by rainfall variability due to the seasonal precipitation regime (ALVARES et al., 2013). During the experimental period, the average temperature and relative humidity were 26.14 ºC and 58.10%, respectively, with an average evapotranspiration of 4.06 mm (EMBRAPA Semiárido, 2021).

Twenty-eight non-castrated, undefined breed lambs (18.46 ± 2.3 kg) were individually housed in covered pens with hard soil for 73 days. The first 13 days were to adapt the lambs to the experimental diets and housing, and the remaining 60 days were for the finishing period. Prior to the experiment, the lambs were weighed, tagged with ear identification, dewormed, and vaccinated against clostridiosis.

Experimental design, treatments and dietary management

Lambs were distributed in a randomized complete design, with four treatments and eight replicates, totaling 32 experimental units. The experimental diets were defined by reallocated sorghum silage and increased levels of concentrate (430, 660, 810, and 910 g/kg) (Table 1) on a dry matter (DM) basis and formulated to meet the requirements of growing lambs (18 kg BW and 200 g/day BW gain), according to the National Research Council (NRC, 2007).

Table 1 – Proportion of ingredients and chemical composition of experimental diets, in dry matter (DM) basis.

	Concentrate levels, g/kg			
Ingredients, g/kg	**430**	**660**	**810**	**910**
Sorghum silage	570.7	344.7	193.9	86.2
Corn	108.8	320.3	484.7	600.5
Cottonseed cake	282.6	300.0	286.7	279.0
Urea	7.0	4.4	4.1	3.8
Mineral supplement[1]	12.9	12.9	12.9	12.9
Sodium bicarbonate	9,7	9.7	9.7	9.7
Ammonium chlorate	7.5	7.5	7.5	7.5
Ammonium sulfate	0.8	0.5	0.5	0.4
Chemical composition, g/kg				
Dry matter	417.0	531.2	649.5	772.5
Crude protein	151.1	151.2	151.0	150.9
Ether extract	53.1	61.1	65.6	68.8
Neutral detergente fiber	483.8	402.4	339.5	295.3
Non-fiber carbohydrate	283.4	330.8	402.3	452.2
ME, Mcal/kg DM[2]	2.3	2.6	2.7	2.8

[1] Mineral supplement: Calcium (Ca) - 140 g; Phosphorus (P) - 70 g; Magnesium (Mg) - 1,320 mg; Iron (Fe) - 2,200 mg; Cobalt (Co) - 140 mg; Manganese (Mn) - 3,690 mg; Zinc (Zn) - 4,700 mg; Iodine (I) - 61 mg; Selenium (Se) - 45 mg; Sulfur (S) - 12 g; Sodium (Na) - 148 g; Fluorine (F) - 700 mg.

[2] Metabolizable energy: According Cqbal 4.0 (https://www.cqbal.com.br/).

Sorghum [*Sorghum bicolor* (L.) Moench] silage (BRS Ponta Negra) was grown in the experimental area of EMBRAPA Semiarid, located in Petrolina-PE, under an irrigated cropping system using central pivot irrigation. Agronomic management followed the technical recommendations of Embrapa Semiarid (REGITANO NETO et al., 2016). The harvest was mechanized for the ensiling process. After the initial opening of the silo, the silage was relocated to 200-liter plastic drums. Following this relocation, the material was stored for an additional period of 20 months before being used for experimental purposes.

The animals were fed diets in the form of a complete ration, in two daily meals (8:00 am and 3:00 pm). The amount of feed offered and refused was recorded daily to adjust feed offered to have

10% refusal. Both feed and refusals were sampled weekly and frozen at –20 ºC for later analysis in the Feed Analysis Laboratory of EMBRAPA Semiarid. Water was provided ad libitum.

Chemical analysis and calculations

At the end of the trial, samples of dietary ingredients and refusals were thawed and pooled by animal, ground through a 1 mm Wiley Mill screen (Marconi, Piracicaba, SP, Brazil) and the DM (Method 934.01), ash (Method 942.05), ether extract (Method 954.01) and total nitrogen (N; Method 968.06) were determined (in triplicate) according to the AOAC (1990). Crude protein was calculated by multiplying the total nitrogen by 6.25. Neutral detergent fiber was determined according to Van Soest et al. (1991), using the fiber analyzer of ANKOM (ANKOM200 Fibre Analyzer – ANKOM Technology Corporation, Fairport, NY, EUA).

Animal slaughter and carcass characteristics after feedlot

After 60 days in feedlot, animals were subjected to fasting with access to water for 16 h. The slaughter proceeded according to the rules of the Regulation of Brazilian Industrial and Sanitary Inspection of Animal Products (BRASIL, 2000).

Lambs were weighed before slaughter to obtain the body weight at slaughter (BWS), and after slaughter, carcasses were weighed to determine the hot carcass weight (HCW) (Welmy, W 300, Santa Bárbara d'Oeste, SP, Brazil). The total gastrointestinal tract (TGI), defined as the sum of the rumen, reticulum, omasum, abomasum, and intestines weights, was removed from each carcass and immediately weighed within 60 minutes of slaughter on an electric scale (Welmy, BCW 6/15/30, Santa Bárbara d'Oeste, SP, Brazil). These data were used to calculate the empty body weight (EBW).

After 24 hours of cooling at 4 °C, the carcasses were weighed again to obtain the cold carcass weight (CCW). Dressing percentage (DP) and true carcass dressing yield (TCD) were calculated using Equations (1) and (2), respectively:

$$DP = (HCW / BWS) \times 100 \quad (1)$$

$$TCD = (HCW / EBW) \times 100 \quad (2)$$

Where:
HCW: hot carcass weight;
BWS: body weight at slaughter;
EBW: empty weight at slaughter.

The pH of the carcasses was measured using a penetration electrode (One-hand pH/temperature measuring instrument: testo 205, Campinas, SP, Brazil) inserted into a cut 2 to 4 cm deep in the *Longissimus lumborum* muscle, between the 4th and 5th lumbar vertebrae, avoiding contact with fatty and connective tissue. Measurements were taken immediately after slaughter (pH0 and T0) and 24 hours post-slaughter (pH24 and T24), following the methodology of Cezar and Sousa (2007).

The *Longissimus lumborum* (LL) muscle was transversely cut between the 11th and 12th ribs, and the subcutaneous fat thickness (SFT) was measured using an outside digital caliper (DIGIMESS, São Paulo, SP, Brazil). Grade rule is a measurement of the abdominal wall, observing the depth of the soft tissue (muscle and fat) deposited on the 12th rib at a point 11 cm away from the midline of the loin, with values below 7 mm considered poor and above 12 mm excessively finished.

The exposed side of the LL was measured using a permanent marker, with a 2.0 mm medium tip, on a transparent plastic film, which determined the rib eye area (REA) in cm². The values obtained from the right and left sides of the carcass were used to calculate the arithmetic meaning of the SFT, GR, and REA per carcass.

The maximum distance of the LL muscle in the mediolateral direction (Measure A) and another perpendicular to it, measuring the maximum distance of the *Longissimus lumborum* muscle in the dorsoventral direction (Measure B), were used to calculate REA using Equation (3):

$$\text{REA} = [(A/\ 2) \times (B/2)] \times \pi \quad (3)$$

Where:

REA: rib eye area in cm^2;

A: maximum distance of the *Longissimus lumborum* muscle in the mediolateral direction;

B: maximum distance of the *Longissimus lumborum* muscle in the dorsoventral direction;

π: mathematical constant Pi, approximately equal to 3.14159.

Qualitative analysis of carcass

For the assessment of conformation and finishing, carcasses were graded from 1 (poor) to 5 (excellent), with emphasis on the following anatomical regions: hind limbs, rump, loin, shoulder, and their muscle plains (CEZAR; SOUSA, 2007).

A score of 1 to 3 was assigned for perirenal fat assessment based on the amount of fat in the abdominal cavity around the kidneys: 1 = left kidney without fat cover; 2 = fully coated left kidney and partially coated right kidney; and 3 = both kidneys covered by a thick layer of fat, according to Cezar and Sousa (2007).

Carcass characteristics, morphometric parameters and commercial cuts after cooling

After the cooling period, external morphometric measurements were taken on whole carcasses: carcass external length (CEL), rump width (RW), thorax width (TW), rump perimeter (RP), and chest width (CW). The carcasses were then halved and weighed.

Measurements were taken on the left half-carcass suspended by the Achilles tendon: carcass internal length (CIL), leg length (LegL), chest depth (CD), and leg perimeter (LP), according to the methodology proposed by Cezar and Sousa (2007).

The carcass compactness index (CCI) was calculated using Equation (4):

$$\text{CCI (kg/cm)} = \text{Weight of cold carcass} / \text{internal length of the carcass} \quad (4)$$

Where:

CCI: carcass compactness index.

The commercial meat cuts of the half carcass were divided into six commercial cuts: leg, shoulder, rib, breast, neck, and loin. Cut yields were estimated in relation to reconstituted cold carcass weight.

Histological and morphological characteristics of viscera's

Fragments (1 cm^2) were collected from the stomach compartments (rumen, reticulum, omasum, and abomasum) and small intestine (duodenum) from the dorsoventral regions for morphological analysis, shortly after the removal of these organs. To avoid postmortem changes, all samples were washed with saline solution to remove excess impurities before fixation. The fragments were quickly immersed in 10% buffered formaldehyde, contained in identified plastic pots.

Histological and histomorphometric analyses were carried out at the Animal Histology Laboratory of the Agricultural Sciences Center of the Federal University of Paraíba (CCA/UFPB).

Fragments smaller than 0.5 cm^3 from organs like the liver (left lateral lobe) and kidney (cortical and medullary areas), as well as fragments no larger than one centimeter from the rumen (wall of the dorsal sac) and small intestine (middle portion of the duodenum), were used and fixed in 10% formaldehyde and packed in identified containers. These fragments were taken from the same topographical portion in all animals. Histological processing included dehydration, clarification, and paraffin embedding, following the methodology of Matos et al. (2022). The microtomy of the blocks was performed with a thickness of 5 µm.

The stains used were hematoxylin and eosin (HE) for morphological, histopathological, and histomorphometric characterization, and periodic acid-Schiff (PAS) to quantify liver glycogen and duodenal goblet cells. Samples were visualized using an Olympus BX53F microscope (Tokyo, Japan) coupled to a digital camera (Olympus DP73) with the aid of cellSens Dimension® software, using the 40x objective for the liver and kidneys. For the liver, six photomicrographs per animal were scanned, totaling 42 per treatment (seven animals x six photomicrographs).

For the analysis of the hepatic glycogen storage index, PAS staining was used, which stains glycoproteins, including hepatic glycogen, the same histologist observer analyzed it by optical microscopy, without prior knowledge of the group belonging to each animal and verified the degree of positivity to the PAS staining (proportional to the amount of hepatic glycogen stock), being: Grade +: little deposit of hepatic glycogen; Grade ++: moderate hepatic glycogen deposition; and Grade +++: heavy deposit of hepatic glycogen. In the analysis of the hepatic glycogen deposit index, the crosses were transformed into corresponding numbers (+ = 1, ++ = 2, +++ = 3) to perform the statistics, according to the Semi Quantitative Score by Ishak et al. (1995) modified. Each of the photomicrographs was given a score between 0 and 3, with 3 being the highest degree of glycogen deposition, according to the modified methodology of Ishak et al. (1995).

For the kidney, six photomicrographs per animal were scanned, totaling 42 per treatment (seven animals x six photomicrographs). The observer looked for histopathological alterations in the components of the nephron (renal corpuscle, proximal convoluted tubules, loop of Henle, and distal convoluted tubules) to identify possible renal damage caused by dietary anti-nutritional factors.

For rumen and small intestine morphometry, the 10x objective was used. In these organs, several photomicrographs per animal were digitized, and five measurements were performed on each (eight animals x five measurements). The variables for the rumen were: papilla height (from the base to the apex), papilla width (in the middle region of the papilla), muscle layer thickness, epithelium thickness, and keratinized portion. The variables for the duodenum were submucosal thickness and mucosal thickness. To measure the amount of goblet cells in the duodenum, several digitized images were used for histomorphometry under PAS staining. For each animal, the number of goblet cells in 2000µm of linear intestinal epithelium was counted. Measurements of papilla height, papilla width, keratinized portion, muscle, epithelium, submucosa, mucosa, and goblet cells were performed using imageJ® software and cellSens Dimension® software using µm as the unit of length.

Statistical analysis

Statistical analysis for all variables was performed using analysis of variance and regression based on concentrate levels in the diet, except for the hepatic glycogen storage index, renal histopathological changes, and histological analyses of the rumen and small intestine. The model was chosen based on the significance of the regression coefficients through the F test using a 5% probability. Data were analyzed using the statistical package R (version 4.2) (R Core Team, 2021).

The data were analyzed using the following mathematical model:

$$Y_i = \beta_0 + \beta_1 X_i + \varepsilon_i$$

Where:

Y_i = observed value for the dependent variable Y at the level of the independent variable X;

β_0 = regression constant, representing the intercept of the curve with the Y-axis;

β_1 = regression coefficient, representing the variation in Y as a function of a one-unit change in X;

X_i= level of the independent variable X (for i = 1,2,…,n);

ε_i = error associated with the distance between the observed value Y_i and the corresponding point on the fitted curve of the proposed model for the same level i of X.

Data for the hepatic glycogen storage index, renal histopathological alterations, and histological analyses of the rumen and small intestine were analyzed using analysis of variance and Tukey's test at 5% probability using the R software.

Results

The increasing levels of concentrate exhibited a quadratic effect on dry matter intake (DMI, $p = 0.008$), metabolizable energy intake (MEI, $p < 0.001$), empty body weight (EBW, $p = 0.045$), and hot carcass weight (HCW, $p = 0.029$), with maximum values observed when animals consumed 560, 608, 727, and 716 g/kg of concentrate, respectively (Figure 1, Table 2).

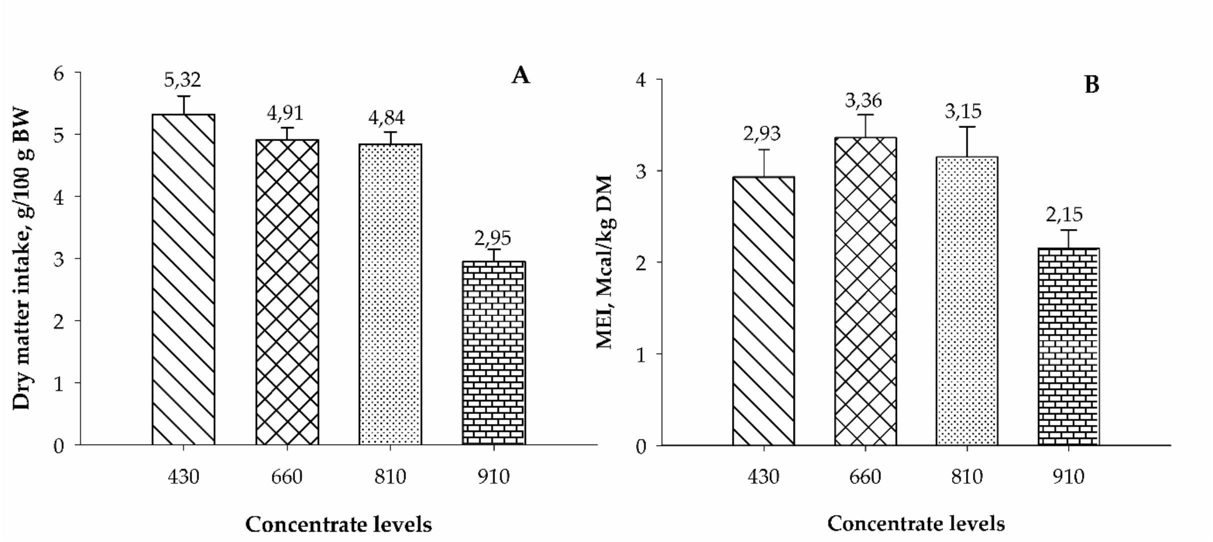

Figure 1 – Dry matter intake (A; DMI; $\hat{Y} = -0.0017x^2 + 0.1907x + 0.2618$; $R^2 = 0.8511$), in g/100 g of body weight, and metabolizable energy intake (B; $\hat{Y} = -0.0016x^2 + 0.1948x - 2.5905$, $R^2 = 0.9333$), in Mcal/kg DM, of lambs fed increasing levels of concentrate (430 g/kg, 660 g/kg, 810 g/kg and 910 g/kg) and reallocated sorghum silage.

Increasing concentrate levels in diets with reallocated sorghum silage did not significantly affect carcass finishing ($p = 0.183$) and ribeye area (REA, $p = 0.124$), averaging 3.76 and 12.81 cm^3, respectively (Table 2), but tended to have a quadratic effect on conformation ($p = 0.056$). Subcutaneous fat thickness (SFT) also had a quadratic effect ($p = 0.011$), with the maximum value found in lambs fed 650 g/kg of concentrate (Table 2). Kidney fat weight increased linearly ($p = 0.006$) with the addition of concentrate. No effect ($p > 0.05$) of experimental diets was observed on kidney fat score, omental fat, and heart fat, averaging 2.2, 1.28, and 0.10 kg, respectively. Dress percentage increased linearly ($p < 0.01$) with increasing levels of concentrate.

Table 2 – Carcass characteristics of lambs fed diets with realocated sorghum silage and increasing levels of concentrate.

Item[1]	**Concentrate levels** [2]**, g/kg**				**CV%**	***P*-value** [4]	
	430	**660**	**810**	**910**		**L**	**Q**
BWS, kg	29.25	36.14	30.64	32.21	10.10	0.320	0.011[5]
Empty body weight, kg	23.60	29.70	26.30	28.00	3.10	0.088	0.045[6]
Hot Carcass weight, kg	13.02	16.65	14.68	15.27	11.01	0.053	0.014[7]
Shrink after chilling, g/100g	3.36	3.15	3.41	3.18	19.11	0.790	0.968
Dress percentage	44.43	46.13	47.91	47.33	4.53	<0.001[8]	0.079
REA (cm^2)	11.80	13.10	13.00	13.50	14.32	0.124	0.601
SFT, cm	0.02	0.03	0.03	0.02	29.32	0.273	0.011[9]
Kidney fat (1-5)	2.31	2.36	2.36	1.79	23.21	0.093	0.146
Conformation (1-5)	2.75	3.86	3.14	3.08	22.91	0.835	0.056
Finishing (1-5)	3.56	4.29	3.64	3.57	19.30	0.634	0.183
Kidney fat, kg	0.28	0.44	0.43	0.53	32.40	0.006[10]	0.565
Heart fat, kg	0.10	0.09	0.10	0.12	35.90	0.372	0.281
Omental fat, kg	0.97	1.34	1.38	1.44	38.26	0.116	0.856

[1] BWS, Body weight at slaughter; REA, Rib eye area; SFT, Subcutaneous fat thickness.

[2] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, on natural basis.

[3] CV, coefficient of variation.

[4] L, linear effect; Q, quadratic effect ($p < 0.05$)

[5] $\ddot{Y} = -0.071x^2+0.9829x+0.5441$, $R^2 = 0.5327$

[6] $\hat{Y} = -0.0052x^2+0.7569x+0.8664$, $R^2 = 0.7133$

[7] $\hat{Y} = -0.0035x^2 + 0.5016x - 1.9381$, $R^2 = 0.7007$

[8] $\hat{Y} = 0.0691x^2 + 41.592$, $R^2 = 0.8814$

[9] $\hat{Y} = -0.00002x^2 + 0.0026x – 0.0564$, $R^2 = 0.9316$

[10] $\hat{Y} = 0.0047x + 0.091$, $R^2 = 0.8901$

However, the weight of half carcass ($p = 0.041$), shoulder ($p = 0.015$), loin ($p = 0.002$), and leg ($p = 0.014$) had a quadratic effect, while the yield of cuts did not change ($p > 0.05$) with experimental diets (Table 3).

Increasing levels of concentrate did not affect ($p < 0.05$) the evaluated non-carcass yield components (Table 3).

Table 3 – Commercial cuts and non-carcass components of lambs fed diets with realocated sorghum silage and increasing levels of concentrate.

Item	**Concentrate levels**[1]**, g/kg**				**CV%**[2]	**P-value**[3]	
	430	**660**	**810**	**910**		**L**	**Q**
Half carcass weight, kg	6.28	7.98	6.87	7.14	11.81	0.326	0.041[4]
Cuts weight, kg							
Neck	0.54	0.59	0.54	0.58	19.62	0.725	0.866
Ribs	1.82	2.40	1.96	2.06	16.80	0.658	0.091
Shoulder	1.13	1.38	1.31	1.28	10.91	0.144	0.015[5]
Loin	0.41	0.58	0.46	0.46	13.30	0.626	0.002[6]
Leg	1.97	2.42	2.15	2.14	9.90	0.512	0.014[7]
Breast	0.40	0.53	0.43	0.51	15.71	0.076	0.301
Cuts yield, g/100 g							
Neck	8.63	7.40	7.95	8.07	15.93	0.625	0.197
Ribs	28.70	29.96	28.44	28.70	7.33	0.699	0.546
Shoulder	18.05	17.38	19.15	17.85	6.41	0.656	0.568

Loin	6.52	7.28	6.73	6.53	10.72	0.684	0.104
Leg	31.60	30.37	31.35	30.21	5.31	0.305	0.971
Breast	6.33	6.77	6.34	7.23	12.90	0.147	0.523
Non-carcass components yield, g/100 g							
Rumen	1.97	1.90	1.89	1.92	4.20	0.696	0.606
Reticulum	0.36	0.36	0.31	0.33	3.01	0.218	0.599
Omasum	0.35	0.32	0.27	0.32	3.02	0.128	0.106
Abomasum	0.54	0.47	0.52	0.49	4.50	0.535	0.483
Small intestine	2.32	2.26	2.33	2.17	9.20	0.689	0.822

[1] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[2] CV, coefficient of variation.

[3] L, linear effect; Q, quadratic effect ($p < 0.05$)

[4] $\hat{Y} = -0.0017x^2 + 0.2451x - 0.9499$, $R^2 = 0.6474$

[5] $\hat{Y} = -0.0003x^2 + 0.0403x - 0.0784$, $R^2 = 0.9430$

[6] $\hat{Y} = -0.0002x^2 + 0.0284x - 0.4179$, $R = 0.7186$

[7] $\hat{Y} = -0.0005x^2 + 0.0741x - 0.2054$, $R^2= 0.7750$

The mean pH values of the leg (0 h) and ribs (0 and 24 h) of lambs did not change ($p > 0.05$) with experimental diets. However, the pH value of the leg measured at 24 h post-slaughter had a quadratic effect ($p = 0.033$), with the maximum value when lambs were fed 660 g/kg of concentrate in the diet (Table 4).

Table 4 – Meat pH measured at 0 h and 24 h after slaughter in leg and rib of lambs fed diets with realocated sorghum silage and increasing levels of concentrate.

Item[1]	**Concentrate levels**[2]**, g/kg**				**CV%**[3]	**P-value**[4]	
	430	**660**	**810**	**910**		**L**	**Q**
Leg							
0 h	7.14	7.03	7.23	7.12	1.20	0.571	0.959
24 h	6.20	6.23	6.43	6.07	0.81	0.622	0.034[5]
Ribs							
0 h	7.57	7.34	7.50	7.28	1.10	0.089	0.888
24 h	6.29	6.28	6.47	6.29	1.30	0.624	0.344

[1] 0h, meat pH measured immediately after slaughter; 24h, p meat pH measured 24 h after slaughter.

[2] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[3] CV, coefficient of variation.

[4] L, linear effect; Q, quadratic effect ($p < 0.05$)

[5] $\hat{Y} = - 0.003x^2 + 0.0399x + 5.023$; $R^2 = 0.3382$

Increasing levels of concentrate did not ($p > 0.05$) change the following morphometric measures: carcass internal length, carcass external length, leg length, rump perimeter, rump width, and chest width. Chest depth increased linearly ($p = 0.048$), while leg perimeter ($p = 0.005$), thorax width ($p = 0.048$), and carcass compactness index (CCI, $p = 0.003$) had a quadratic effect, with maximum values when lambs were fed 697, 622, and 755 g/kg of concentrate, respectively (Table 5).

Table 5 – Morphometric measures on carcasses of lambs fed lambs fed diets with realocated sorghum silage and increasing levels of concentrate.

Item, cm[1]	**Concentrate levels**[2]**, g/kg**	**CV%**[3]	**P-value**[4]

	430	660	810	910		L	Q
CIL	52.10	48.10	48.3	53.00	11.20	0.769	0.067
CEL	57.10	56.70	55.60	56.70	4.71	0.645	0.490
Chest depth	29.20	32.00	31.70	31.90	6.60	0.048[5]	0.124
Leg length	41.00	40.40	40.70	40.80	6.30	0.937	0.756
Leg perimeter	38.25	44.85	42.14	41.28	7.31	0.244	0.005[6]
Rump perimeter	48.50	50.42	48.71	50.57	5.22	0.331	0.972
Rump width	14.12	14.42	14.00	14.78	10.01	0.545	0.678
Thorax width	14.12	13.71	13.71	15.14	6.80	0.083	0.024[7]
Chest width	11.43	12.14	12.14	12.13	17.30	0.494	0.738
CCI, kg/cm	0.24	0.33	0.29	0.28	13.41	0.301	0.003[8]

[1] CIL, carcass internal length; CEL, carcass external length; CCI, Carcass compactness index.

[2] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[3] CV, coefficient of variation.

[4] L, linear effect; Q, quadratic effect ($p < 0.05$)

[5] $\hat{Y} = 0.0543x + 27.383$, $R^2 = 0.7170$

[6] $\hat{Y} = -0.008x^2 + 1.1148x + 5.1779$, $R^2 = 0.9020$

[7] $\hat{Y} = 0.0019x^2 - 0.2365x + 20.848$. $R^2 = 0.8307$

[8] $\hat{Y} = -0.0001x^2 + 0.0151x - 0.2082$, $R^2 = 0.823$

The increasing levels of concentrate altered the deposition of hepatic glycogen levels, with higher liver glycogen scores observed for the 430 g/kg concentrate diet and lower scores for the 810 g/kg concentrate level (Table 6).

Table 6 – Liver glycogen stores score of finishing lambs and frequency of scores per experimental diets.

Concentrate levels [1], g/kg	**Adapted score of Ishak ***				**Average/Standard deviation [2]**
	0	**1**	**2**	**3**	
	Frequency of score by diet **				
430	2	14	23	9	1.81 ± 0.79 a
660	8	20	20	0	1.25 ± 0.73 b
810	20	18	6	0	0.68 ± 0.71 c
910	8	21	10	1	1.10 ± 0.74 b

*0, absence of positivity, 1, little positivity; 2, moderate positivity; 3 intense positivity.

** Score frequency of each photomicrograph analyzed by treatment.

[1] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[2] Means followed by different lowercase letters in the columns and different capital letters in the rows differ from each other by the Tukey test, at 5% probability.

Renal histopathological alterations were also observed, with lambs fed 810 g/kg of concentrate showing deleterious effects on the kidneys, including higher incidences of hyaline cylinders in the lumen, necrosis, and inflammation (Table 7; Figure 2).

Table 7 – Histological changes observed in kidneys of finishing lambs fed increasing levels of concentrate and realocated sorghum silages.

Item	**Concentrate levels [1], g/kg**			
	430	**660**	**810**	**910**
Hyaline cylinders	+	+	++	+

Necrosis	-	+	+	-
Inflammation	-	-	+	-

- Absent; + Mild; ++ Moderate; +++ Acute.

[1] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

Lambs fed 910 g/kg of concentrate had higher epithelium thickness ($p < 0.05$), papilla height ($p < 0.05$), and rumen papillae width ($p < 0.05$) (Table 8; Figure 2). The keratinized portion of the rumen was also greater for animals consuming 910 g/kg compared to those consuming 430 and 660 g/kg of concentrate. The muscle layer thickness did not differ between animals ($p = 0.099$), averaging 930.90 µm.

Table 8 – Rumen morphology characteristics of finishing lambs fed increasing levels of concentrate and realocated sorghum silages.

Item [1](µm)	**Concentrate levels [2,3], g/kg**			
	430	**660**	**810**	**910**
MLT	855.00 ± 181.95	906.71 ± 226.55	852.71 ± 189.33	1057.65 ± 341.46
ET	92.58 ± 20.84 b	104.20 ± 36.08 a	117.64 ± 42.58 a	117.77 ± 41.84 a
KP	17.56 ± 5.24 b	17.32 ± 5.15 b	24.19 ± 8.18 a	26.91 ± 8.59 a
HP	2476.47 ± 745.78 b	2525.47 ± 1939.09 b	2843.77 ± 1461.12 ba	3500.07 ± 1017.9 a
PW	230.29 ± 47.78 b	224.53 ± 144.68 a	278.45 ± 104.90 a	306.24 ± 185.87 a
GC	24.50 ± 9.00 bc	25.42 ± 6.58 b	32.79 ± 5.64 a	18.61 ± 7.88 c

[1] MLT, Muscle layer thickness; ET, Epithelium thickness; KP, Keratinized portion; HP, Height of papillae; PW, Papillae width; GC, goblet cells.

[2] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[3] Averages followed by different lowercase letters in the rows differ from each other by the Dunn test ($p < 0.05$).

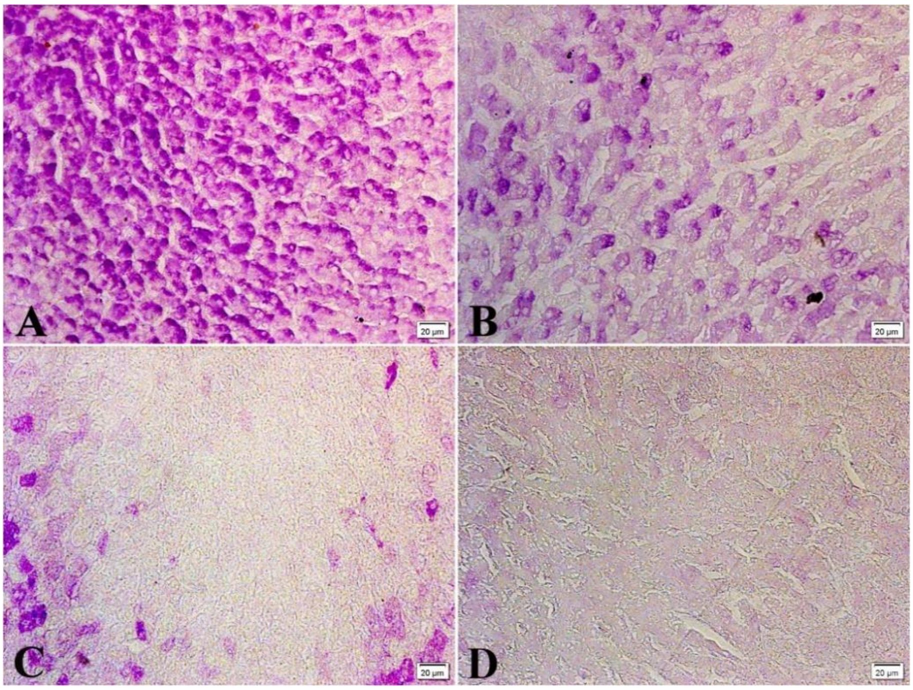

Figure 2 – Photomicrographs of liver from feedlot lambs fed different levels of concentrates in diets with reallocated sorghum silage: A) 430 g/kg of supplement concentrate, B) 660 g/kg of supplement concentrate, C) 810 g/kg of supplement concentrate and, D) 910 g/kg of supplement concentrate. Staining: Periodic acid-Schiff. Scale bar: 20µm.

A quadratic effect was observed on the submucosal ($p = 0.001$) and mucosal ($p = 0.033$) thickness of the small intestine (Table 9, Figure 3). Additionally, lambs fed 810 g/kg of concentrate had higher goblet cell (GC) counts compared to other experimental diets ($p < 0.001$). The diet with 910 g/kg of concentrate resulted in lower GC counts in the animals, not differing significantly from the animals consuming 430 g/kg of concentrate.

Table 9 – Small intestine morphology of lambs fed increasing levels of concentrate and realocated sorghum silages.

Item[1] (µm)	**Concentrate levels[2], g/kg**				**CV%[3]**	***P*-value[4]**	
	430	**660**	**810**	**910**		**L**	**Q**
ST	113	174	153	132	7.20	0.344	< 0.001[5]
MT	735	697	872	693	4.50	0.736	0.034[6]
GC	22.20 bc	25.41 b	32.82 a	18.60 c	21.90	0.735	< 0.001[7]

[1] ST, Sub1; ST, Submucosal thickness; MT, mucosal thickness; GC, goblet cells.

[2] 430, adding of 430 g/kg concentrate and 570 g/kg of sorghum silage; 660, adding of 660 g/kg concentrate and 340 g/kg of sorghum silage; 810, adding of 810 g/kg concentrate and 190 g/kg sorghum silage; 910, adding of 910 g/kg concentrate and 90 g/kg sorghum silage, in natural basis.

[3] CV = coefficient of variation.

[4] L, linear effect; Q, quadratic effect ($p < 0.05$)

[5] $\hat{Y} = -0.0854x^2 + 11.767x - 234.33$. $R^2 = 0.9751$

[6] $\hat{Y} = -0.0639x^2 + 9.0609x + 448.74$. $R^2 = 0.0682$

[7] $\hat{Y} = -0.0139x^2 + 1.8624x - 33.039$. $R^2 = 0.4355$

Averages followed by different lowercase letters in the rows differ from each other by the Dunn test ($p < 0.05$).

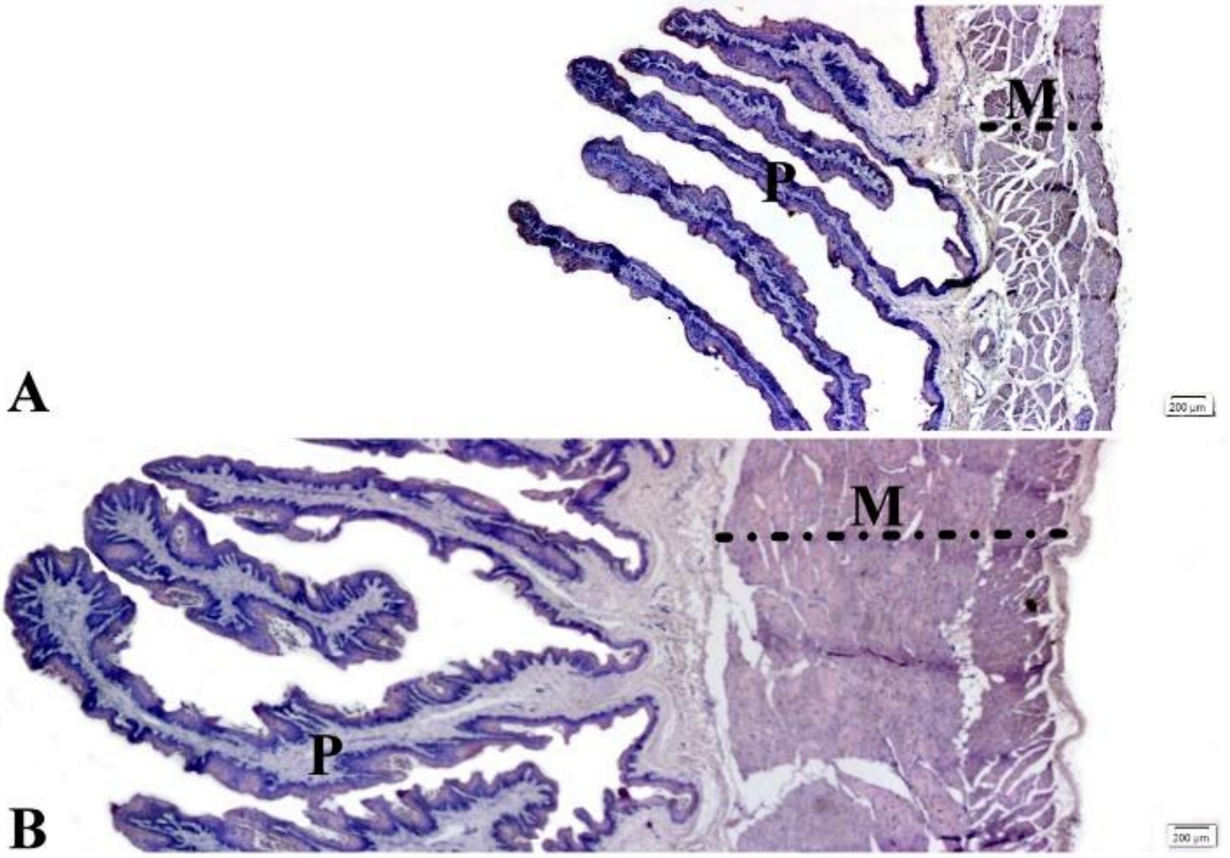

Figure 3 – Photomicrographs of small intestine from feedlot lambs fed different levels of concentrates in diets with reallocated sorghum silage: A) 430 g/kg of supplement concentrate, B) 660 g/kg of supplement concentrate. M) Muscle layer; P) Papilla. Scale bar: 200µm.

Discussion

Diet composition is one of the main factors influencing animal performance (WU et al., 2021). Increasing concentrate levels in the diet may lead to a reduction in DMI, an effect generally associated with higher energy density and a lower fiber proportion in the diet (OLIVEIRA et al., 2024; WU et al., 2021), as observed in the composition of the high-concentrate diets (Table 1). This reduction in fiber content can result in decreased total chewing time and increased ruminal passage rate (OLIVEIRA et al., 2024).

The increasing levels of concentrate in diets containing reallocated sorghum silage are closely related to carcass characteristics, including the DP, which is an important index for measuring carcass traits (WANG et al., 2020).

Nutrition plays a critical role, leading to variations in gut-fill that can significantly influence visceral weight and body fat, and consequently, the dressing percentage (ASSAN, 2020; GARDNER et al., 2015). In this study, although BWS and, consequently, HCW had a quadratic response, DP increased linearly as concentrate was added to the diet, indicating that higher levels of concentrate promote fat deposition in the carcass (JABOREK et al., 2017) or visceral fat, given that DP is a proportion of HCW relative to BWS, expressed as a percentage (GARDNER et al., 2015).

A greater visceral fat depot was also reported in a previous study comparing concentrate-fed lambs with those fed roughage, as the supply of larger quantities of starch and glucose to the small intestine in high-starch diets makes the energy substrate readily available for use and storage (JABOREK et al., 2017), as likely occurred in this research when lambs were fed increasing levels of concentrate. Although omental fat did not change in this study, lambs fed higher levels of concentrate had up to 400 g more omental fat than animals fed diets with a higher proportion of reallocated silage.

In ruminants, internal fat is the first depot formed, followed by intermuscular, subcutaneous, and intramuscular fat or marbling (PETHICK et al., 2004). This explains the linear increase in kidney fat and the quadratic effects on carcass subcutaneous fat thickness.

Thus, although DP increased linearly with concentrate addition, the proportion of 741 g/kg of concentrate in diets with sorghum silage promoted the maximum leg weight (2.54 kg), an important carcass cut, affecting the carcasses of feedlot lambs fed reallocated sorghum silage. In this study, diets with concentrate levels above 810 g/kg negatively affected carcass characteristics, likely due to nutritional disorders directly impacting kidney activity.

Increasing levels of concentrate in diets containing reallocated sorghum silage promoted histopathological alterations in the kidneys, with rupture and leakage of hyaline substance into the tubular lumen, resulting in higher amounts of hyaline cylinders in the lumen and severe lesions, including necrosis and inflammation due to toxic injury. Regulated necrosis is associated with damage and loss of the plasma membrane, with the release of proteolytic enzymes and organelles due to extracellular causes. When apoptotic cells are not engulfed by phagocytes in a timely manner, their content increases in high numbers in the extracellular space, triggering inflammation and consequently causing kidney failure (PRIANTE et al., 2019).

Grain challenge experiments, where a large portion of the diet is suddenly replaced by highly fermentable grains, result in systemic inflammation in dairy cattle (KROGSTAD; BRADFORD, 2023) and beef cattle (GOUVÊA et al., 2022). An extensive review demonstrated that more than 350 g/kg concentrate in the diet of cattle resulted in a linear rise in the concentration of rumen endotoxin, leading to the presence of inflammation biomarkers in the plasma (ZEBELI et al., 2012).

It is possible that the histological changes observed in the kidneys of finishing lambs fed increasing levels of concentrate and reallocated sorghum silage resulted in a reduction in DMI, as suggested by Bertoni et al. (2015), because the lambs also decreased their MEI. Consequently, a reduction in liver glycogen storage was observed, along with a quadratic response in BWS, carcass characteristics, and important morphometric measures such as leg perimeter and thorax width. This effect also explains the tendency for a quadratic effect on carcass conformation, but this was not reflected in the proportion of commercial cuts of the carcass and non-carcass components.

A reduction in rumen proportion was expected, as diets with high fermentable starch increase propionate flux to the liver, stimulating hepatic oxidation and ATP production, which reduces meal size (NRC, 2021). Starch escaping the rumen is digested to glucose, absorbed, and partially metabolized to lactate (NRC, 2021). Consequently, glycogen storage was reduced with increasing dietary concentrate levels because glucose is also used as fuel by portal-drained visceral tissue (HUNTINGTON et al., 2006).

Regarding meat pH, after slaughter, biochemical reactions in the muscle continue, but since blood no longer circulates, glucose and oxygen are not delivered to the muscle. As a result, glycogen stored locally in the muscle is used as an energy source and catabolized anaerobically (TERLOUW et al., 2021), leading to a reduction in pH. Evidence suggests that muscle glycogen levels increase with metabolizable energy intake (PETHICK et al., 2004). In agreement with this, the quadratic effect observed on pH at 24 hours post-slaughter of the leg aligns with MEI. However, this effect was not observed for the pH of the ribs at 24 hours post-slaughter, likely due to site-specific variations and fiber-type-specific preferential use of muscle glycogen during exercise, as reported by Schweitzer et al. (2017).

The use of starch-rich diets enhances the absorption of short-chain fatty acids and, consequently, elongates the ruminal papillae (DANTAS JÚNIOR et al., 2022). The fermentation of non-fiber carbohydrates increases the propionate proportion in the rumen, but its association with roughage is essential for promoting the growth and width of ruminal papillae, thereby increasing the ruminal absorption area compared to diets containing exclusively concentrate (AL-GALBI et al., 2022). Similar results were found with young goat (SHEN et al., 2004) and calves (KIM et al., 2012) when tested with high energy density diets due to high concentrate levels. Additionally, according to Dantas Júnior et al. (2022), a high number of goblet cells indicates better intestinal health, demonstrating that both excess and deficiency of concentrate in the diet negatively affect the cells of the small intestine and rumen.

Conclusion

Reallocated sorghum silage is an excellent fiber source for finishing lambs when included in diets with concentrate proportions up to 810 g/kg to maximize dressing percentage without damaging kidney histological parameters.

Conflicts of interest

There was no conflict of interest between the authors.

Author contributions

Antoniel Cruz – designed the experiment, sampling, data collection and wrote the manuscript; Nelson Santos – conducted sampling, data collection, wrote the first draft of the manuscript, designed the experiment and analyzed the data; Fleming Campos – conducted sampling, data collection and revised the final version of the manuscript; Michele Parente – wrote the manuscript and revised the final version of the manuscript; Gherman Araújo – revised the final version of the manuscript; Paulo Júnior – conducted sampling, data collection and analyzed the data; Alberto Macêdo – conducted sampling, data collection and revised the manuscript; Carla Saraiva – revised the final version of the manuscript; Ricardo Guerra – revised the final version of the manuscript; Anderson Pereira – review and editing of the manuscript, data curation and visualization; Edson Santos – designed the experiment and revised the manuscript; Juliana Oliveira – designed the experiment, wrote the manuscript and revised the manuscript. All authors have revised the final version of the manuscript.

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Declaration of Generative AI and AI-Assisted Technologies in the Writing Process

The authors used ChatGPT 4.0 to improve the writing and check the consistency of the text. The content was subsequently reviewed and edited by the authors, who take full responsibility for the publication.

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