

Ingestive behavior, ruminal and blood kinetics in lambs fed cow cheese whey

Comportamento ingestivo, cinéticas ruminal e sanguínea em ovinos alimentados com soro de queijo bovino

GUEDES^{1*}, Luciana Freitas
<https://orcid.org/0000-0002-5059-2011>

NEIVA³, José Neuman Miranda
<https://orcid.org/0000-0001-7817-8210>

PRIMO², Tatiana Santos
<https://orcid.org/0000-0002-9773-7740>

COSTA⁴, Clésio dos Santos
<https://orcid.org/0000-0003-0451-7157>

VASCONCELOS², Ângela Maria de
<https://orcid.org/0000-0002-9690-974X>

OLIVEIRA², Delano de Sousa
<https://orcid.org/0000-0002-1960-335X>

POMPEU¹, Roberto Cláudio Fernandes Franco
<https://orcid.org/0000-0002-4099-3575>

ROGÉRIO¹, Marcos Cláudio Pinheiro
<https://orcid.org/0000-0003-3567-5211>

¹Embrapa Caprinos e Ovinos, Laboratório de Respirometria do Semiárido, Sobral, Ceará, Brasil.

²Universidade Estadual Vale do Acaraú, Departamento de Zootecnia, Sobral, Ceará, Brasil

³Universidade Federal do Tocantins, Departamento de Zootecnia, Araguaína, Tocantins, Brasil

⁴Universidade Federal do Ceará, Departamento de Zootecnia, Fortaleza, Ceará, Brasil

*Corresponding author: lucianafguedes@gmail.com

ABSTRACT

The objective of this study was to evaluate the inclusion of cow cheese whey (CCW) in the diet for lambs on the ingestive behavior, ruminal and blood parameters. Twenty crossbred, male, non-castrated lambs with 25 kg body weight were assigned to four treatments: control diet (composed of corn, soybean meal and native pasture silage) and test diets (control diet + inclusion of CCW in increasing levels of 1.6, 2.7 and 4.0% in DM), in a completely randomized design with four treatments and five replications. Means were compared by Tukey's test, and the strength of association between variables was tested by Pearson's correlation. Regression analysis was also performed for data from the collection of ruminal fluid and blood. Behavioral assessments were made at a 24-hour interval in measurements taken every five minutes. The inclusion of CCW in the diet did not impair ruminal pH, but promoted a decrease in rumination and feeding times, also causing a lower number of cuds and a lower number of cud chews. The lowest concentration of ammonia nitrogen and total protein were found in lambs receiving the diet with 4% CCW. However, these values were within the range recommended for microbial growth. CCW is a potential protein alternative in lambs feed as it does not interfere with nutrient intake, ruminal and blood parameters, and favors a positive nitrogen balance.

Key words: feedlot, diet, rumination, by-product

RESUMO

Objetivou-se avaliar a inclusão de soro de queijo bovino (SQB) na dieta para ovinos sobre o comportamento ingestivo, os parâmetros ruminais e sanguíneos. Vinte ovinos mestiços, machos, não castrados, com 25 kg de peso corporal foram distribuídos em quatro dietas: dieta controle (composta por milho, farelo de soja e silagem de pasto nativo) e as dietas teste (dieta controle + inclusão de SQB em níveis crescentes 1,6; 2,7 e 4,0% na MS), em um delineamento inteiramente ao acaso com quatro tratamentos e cinco repetições. A comparação de médias foi realizada pelo teste Tukey e foram testadas as correlações de Pearson entre as variáveis. Também foi feita a análise de regressão para os dados provenientes da coleta de líquido ruminal e de sangue. As avaliações de comportamento foram feitas em um intervalo de 24 horas em medições realizadas a cada cinco minutos. A inclusão de SQB na dieta não comprometeu o pH ruminal, mas promoveu a queda nos tempos de ruminação e de alimentação, ocasionando também menor número de bolos ruminais e menor número de mastigações meréricas. A menor concentração de nitrogênio amoniacal e proteínas totais foram encontradas nos ovinos que receberam a dieta com 4% de inclusão de SQB. Entretanto, esses valores ficaram dentro do padrão estabelecido para crescimento microbiano. O SQB é uma potencial alternativa proteica na alimentação de ovinos por não interferir no consumo de nutrientes, nos parâmetros ruminais e sanguíneos, e favorecer o balanço positivo de nitrogênio.

Palavras-chave: confinamento, dieta, ruminação, subproduto

INTRODUCTION

The seasonality of rainfall in the Northeast of Brazil reduces the quality and availability of forages and grains, which in turn affect the production indices of herds, which implies an increase in costs related to feeding. Thus, the use of by-products to feed ruminants with nutritional quality can contribute to improve the efficiency of production systems in the northeastern semiarid.

In this condition, cow cheese whey (CCW) is considered a potential food alternative, with wide availability, high acceptability by animals and is made up of proteins of high biological value (DAVID *et al.*, 2010). The annual national production of CCW is estimated at 500 thousand tons, on average, however the use of this by-product reaches only 15% of the total whey produced, being considered a high-grade pollutant (PRAZERES *et al.*, 2012).

Therefore, CCW is a by-product of high nutritional potential, but a large part is disposed in the environment.

When evaluating alternative feeds for animals, dry matter intake and nutrient digestibility important parameters to determine the nutritional value of the food. However, the study of ingestive behavior is essential and complementary, as it allows the adjustment of food management of the animals according to the characteristics of the food and the nictemeral pattern and, thus, seeks the best productive performance according to the diet provided. In general, the periods spent with food intake are interspersed with one or more periods of rumination or idleness, and the time spent on rumination is usually longer at night, being mainly controlled by the nature of the diet. In addition, the assessment of ruminal fluid pH is an important parameter because it is correlated to the

final fermentation products as well as the growth rate of rumen microorganisms, so that it can decline with fiber restriction in the diet or by the addition of rapidly fermentable carbohydrates (VAN SOEST, 1994).

Thus, the objective was to evaluate increasing levels of cow cheese whey in lambs diets on the intake and digestibility of nutrients, ingestive behavior and ruminal and blood parameters.

MATERIAL AND METHODS

The experiment was carried out at the Research Center for Nutrition of Small Ruminants at the Vale do Acarau Experimental Farm, belonging to the Vale do Acarau State University - UVA, in Sobral, state of Ceará, at 3°36' South latitude, 40°18' West longitude, 56 m. The region has a BSh (Köppen classification), megathermal, dry climate, with average rainfall (February to May) of 888.9 mm, corresponding to 92.6% of the average annual total. The average annual temperature is 26.6 °C and the average relative humidity is 67.9%.

Twenty crossbred, non-castrated, male lambs, five months of age, average body weight of 25 kg, were weighed, dewormed and individually housed in metabolic cages. The control diet consisted of corn, soybean meal and siratro silage (*Macroptilium atropurpureum* DC. cv. Siratro) with native Caatinga pasture containing: vassourinha-de-botão (*Borreria verticillata* G.F.W. Mayer), climbing dayflower (*Commelina diffusa* Burnm.F), jetirana (*Merremia aegyptia*), malva-branca (*Sida cordifolia*), capim-milhã (*Brachiaria* spp.; *Panicum* spp.; *Setaria* spp) and pinto peanut (*Arachis pintoi*). Test diets were composed of the same ingredients with inclusion of cow cheese whey (CCW) at increasing levels (1.6; 2.7 and 4.0% in DM) (Tables 1 and 2). Diets were formulated according to the nutritional requirements of finishing lambs, with four months of age, 20 kg body weight and gain of 200 g day⁻¹, based on the NRC (2007), establishing average food leftovers of 10-15% of the total provided in DM. Diets were supplied to the lambs at eight and at 17 hours. Mineral salt and water were available at will.

Table 1. Chemical composition (%) of feed supplied during the experimental period.

| Component (%DM*) | Pasture silage | CCW | Corn | Soybean meal | Limestone e |
|-------------------------|----------------|-------|-------|--------------|-------------|
| Dry matter [#] | 27.58 | 6.78 | 89.36 | 89.65 | 100.00 |
| Crude protein | 8.96 | 18.30 | 10.33 | 53.56 | - |
| Ether extract | 1.26 | 7.37 | 3.48 | 4.30 | - |
| Neutral detergent fiber | 76.17 | - | 29.63 | 43.43 | - |
| Acid detergent fiber | 50.78 | - | 6.09 | 11.27 | - |
| Cellulose | 35.34 | - | 4.04 | 9.87 | - |
| Lignin | 10.40 | - | 0.68 | 1.56 | - |
| Mineral matter | 12.73 | 0.89 | 1.17 | 0.43 | - |
| Calcium | 1.74 | 0.43 | 0.85 | 0.82 | 38.00 |
| Phosphorus | 0.52 | 0.03 | 0.28 | 0.77 | - |
| Total carbohydrates | 77.05 | 73.44 | 85.02 | 35.91 | - |
| Non-fiber carbohydrates | 5.63 | 73.44 | 55.83 | 11.23 | - |
| TDN | 44.15 | 97.60 | 77.41 | 62.61 | - |

*DM = Dry matter; CCW = cow cheese whey; TDN = Total digestible nutrients, according to NRC (2001). [#]in % natural matter.

Table 2. Proximate and chemical composition of diets (%) containing increasing levels of cow cheese whey (CCW) for finishing lambs.

| <i>Proximate composition of the diets (%natural matter)</i> | | | | | |
|---|-----------------------|-------|-------|--------------|-----------|
| CCW inclusion (%MS) | Native pasture silage | CCW | Corn | Soybean meal | Limestone |
| 0 | 73.41 | 0.00 | 17.62 | 8.71 | 0.27 |
| 1.6 | 64.79 | 10.17 | 16.94 | 7.87 | 0.23 |
| 2.7 | 56.83 | 18.25 | 16.76 | 7.91 | 0.26 |
| 4.0 | 49.00 | 26.05 | 17.18 | 7.46 | 0.31 |
| <i>Chemical composition of experimental diets</i> | | | | | |
| Component (% DM) | 0 | 1.6 | 2.7 | 4.0 | |
| Dry matter [#] | 44.07 | 40.98 | 39.24 | 36.63 | |
| Crude protein | 13.06 | 13.63 | 14.40 | 14.93 | |
| Ether extract | 1.91 | 2.49 | 2.98 | 3.46 | |
| Neutral detergent fiber | 64.92 | 57.79 | 51.69 | 45.65 | |
| Acid detergent fiber | 39.33 | 34.82 | 30.77 | 26.77 | |
| Cellulose | 27.51 | 24.36 | 21.54 | 18.75 | |
| Lignin | 7.89 | 6.98 | 6.15 | 5.33 | |
| Mineral matter | 10.09 | 9.03 | 8.09 | 7.14 | |
| Calcium | 1.60 | 1.47 | 1.37 | 1.29 | |
| Phosphorus | 0.50 | 0.45 | 0.41 | 0.37 | |
| Total carbohydrates | 74.67 | 74.61 | 74.28 | 74.17 | |
| Non-fiber carbohydrates | 14.95 | 21.46 | 26.85 | 32.32 | |
| TDN | 61.65 | 63.61 | 67.14 | 68.75 | |

*DM = Dry matter; TDN = Total digestible nutrients, according to NRC (2001).

[#]in % natural matter.

The experiment lasted 30 days with 23 days for adaptation and seven days for data collection (intake and digestibility test, collection of ruminal fluid and blood, and ingestive behavior assessments). Samples of the offered diet, leftovers, urine and feces were collected daily, weighed, packed in plastic bags and refrigerated at -10°C). Subsequently, samples were thawed at room temperature to make a composite sample of the seven days per animal, and taken to a forced ventilation oven at 55 °C for 72 hours. Soon after, they were ground through a 1 mm sieve and reserved for analysis.

Chemical composition was determined at the Animal Nutrition Laboratory of UVA. The methodology proposed by AOAC (2010) was followed for determinations of dry matter (DM), organic matter (MO) and mineral matter (MM), ether extract (EE) and crude protein (CP) of the material. In turn, the methodology of Van Soest *et al.* (1991) was used for quantification of fiber portions, neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin and cellulose.

Total digestible nutrients (TDN) of foods was estimated according to equations of Cappelle *et al.* (2001). For calculation of TDN of experimental diets, the equation of NRC (2001) was used. Non-fiber carbohydrates (NFC) was estimated according to Weiss (1999). Calculations of digestibility coefficients of DM, OM, CP and NDF were determined by the formula [(Nutrient ingested (g) - Amount of nutrients in feces (g)) / Nutrient intake (g)] x 100.

Ruminal fluid was collected by esophageal tube for the measurements of ruminal pH and ammonia nitrogen (NH₃-N) at four pre-established times (0, 3, 6

and 9 h postprandial). The pH was measured in a Micronal B271[®] potentiometer, immediately after the collection of the ruminal fluid, while the samples with 50 mL ruminal fluid were acidified in 1 mL sulfuric acid 1: 1 and stored at -5 °C to determine NH₃-N, by distillation with magnesium oxide, at the Animal Nutrition Laboratory of UVA. Blood was drawn by puncture of the jugular vein to determine serum levels of urea and total proteins, at the times established for the collection of ruminal fluid. Serum analyses were performed using Bioclin[®] kits, at the Animal Nutrition Laboratory of UVA and at the Bromatology Laboratory of the Federal Institute of Education, Science and Technology of Ceara (IFCE - Sobral *Campus*).

Ingestive behavior was evaluated during 24 hours with time spent on feeding (TA), rumination (TR), idle (TO) and other activities, adopting the visual observation of the animals every five minutes according to Johnson and Combs (1991), using a digital stopwatch. In the night observation, the environment was maintained with artificial lighting. Ingestive behavior variables were obtained from the ratios: ERMS = CMS/TRU; ERFDN = CFDN/TRU; TMT = TAL+TRU; tMMb = BOL/MMb, where: ERMS = rumination efficiency in grams of dry matter/hour; CMS = dry matter intake in grams; TRU = rumination time in hours/day; ERFDN = rumination efficiency in grams of neutral detergent fiber/hour; CFDN = neutral detergent fiber intake in grams; TMT = total chewing time in hours/day; TAL = feeding time in hours/day; tMMb = time of cud chews per cud in seconds/cud; BOL = number of cuds/day, considering cud as the portion of food that returns to

the mouth to be chewed for the second time (Polli et al., 1996); MMb = number of cud chews per cud.

Data were initially subjected to normality (Cramer-Von Misses) and homoscedasticity (Levene) tests; when the assumptions were met, an analysis of variance was applied using the F-test. In case of significant differences, means were compared by SNK test at 5% probability level. Statistical analyses were run using the GLM procedure of SAS 9.0 software (2002).

Data on intake, digestibility and behavior were analyzed in a completely randomized design with four treatments (inclusion of CCW) and five replications, according to the statistical model:

$$Y_{ij} = \mu + H_j + e_{ij}$$

Where:

Y_{ij} = value referring to the observation of repetition i of treatment j ;

μ = overall mean;

H_j = effect of treatment j (j = zero; 1.6; 2.7; 4.0);

e_{ij} = random error associated with the observation.

Data on ruminal fluid and blood were analyzed in a completely randomized design, in a split-plot arrangement, with the treatments (inclusion of CCW) in the plots, and the collection times in the subplots, with four replications, according to the statistical model:

$$Y_{ijk} = \mu + F_j + T_k + FT_{jk} + e_{ijk}$$

Where:

Y_{ijk} = value referring to the observation of repetition i in treatment j and in collection time k ;

μ = overall mean;

F_j = effect of treatment j (j = zero, 1.6%, 2.7%, 4.0%);

T_k = effect of the collection time k (k = zero, 3, 6, 9);

FT_{jk} = interaction of the effects of treatment j with collection time k ;

e_{ijk} = random error associated with the observation.

RESULTS AND DISCUSSION

The inclusion of CCW in a lambs diet did not influence the intake and digestibility of nutrients, except ($p < 0.05$) for the intake of the fiber portion of the diet (Table 3).

Table 3. Intake of dry matter, organic matter, crude protein, ether extract and digestible fractions of lambs diets containing increasing levels of cow cheese whey (CCW).

| Component | CCW inclusion (% DM) | | | | CV (%) |
|---|----------------------|----------------------|----------------------|----------------------|--------|
| | 0 | 1.6 | 2.7 | 4.0 | |
| <i>Nutrient intake (grams per unit of metabolic size)</i> | | | | | |
| Dry matter (g/UTM) | 111.75 | 109.14 | 101.26 | 104.36 | 10.63 |
| Dry matter (g/day) | 1345.25 ^b | 1360.70 ^a | 1158.64 ^d | 1309.43 ^c | 18.01 |
| Organic matter (g/UTM) | 95.49 | 91.72 | 83.57 | 84.69 | 10.84 |
| Organic matter (g/day) | 1149.83 ^a | 1143.78 ^a | 956.04 ^b | 1062.23 ^a | 18.09 |
| Crude protein (g/UTM) | 20.05 | 19.59 | 18.78 | 18.79 | 11.70 |
| Crude protein (g/day) | 239.57 | 244.01 | 214.56 | 234.98 | 16.64 |
| Ether extract (g/UTM) | 2.98 | 3.01 | 2.86 | 2.94 | 12.85 |
| Ether extract (g/day) | 35.84 | 37.49 | 32.72 | 36.45 | 16.64 |
| Neutral detergent fiber (g/UTM) | 57.46 ^a | 52.76 ^{ab} | 46.27 ^b | 46.04 ^b | 11.20 |
| Neutral detergent fiber (g/day) | 692.36 ^a | 657.89 ^{ab} | 529.10 ^c | 579.11 ^{bc} | 18.63 |
| Non-fiber carbohydrates (g/day) | 261.72 ^b | 305.25 ^{ab} | 286.01 ^{ab} | 355.10 ^a | 19.55 |
| <i>Nutrient digestibility (%)</i> | | | | | |
| Dry matter | 64.76 | 65.02 | 68.06 | 68.25 | 7.56 |
| Organic matter | 67.11 | 66.97 | 69.25 | 68.91 | 7.25 |
| Crude protein | 66.52 | 66.05 | 66.24 | 65.81 | 8.06 |
| Ether extract | 76.89 | 79.27 | 77.35 | 80.75 | 7.60 |
| Neutral detergent fiber | 46.18 | 43.26 | 41.38 | 39.11 | 18.44 |

Mean values followed by different letters, in the same row, are significantly different by SNK test (P<0.05).

The inclusion of CCW favored the intake of NFC (Table 3), which corresponds to the higher intake of components of fractions A (soluble sugars and rapidly degraded organic acids) and B1 (starch, pectin and glucans). These components are rapidly fermented, providing readily

available energy for ruminal microorganisms (WEISS *et al.*, 1999). In addition, the inclusion provided a positive nitrogen balance due to the effect of protein on digestion and microbial fermentation (Table 4).

Table 4. Nitrogen balance in lambs fed increasing levels of cow cheese whey (CCW) in the diet.

| Parameter | CCW inclusion (% DM) | | | | CV (%) |
|----------------------------------|----------------------|-------|-------|-------|--------|
| | 0 | 1.6 | 2.7 | 4.0 | |
| Ingested nitrogen (g/day) | 38.33 | 39.04 | 34.33 | 37.59 | 16.64 |
| Fecal nitrogen (g/day) | 12.85 | 13.27 | 11.57 | 12.71 | 22.16 |
| Urinary nitrogen (g/day) | 6.17 | 7.17 | 6.56 | 5.24 | 41.28 |
| Nitrogen balance | 19.30 | 18.60 | 16.19 | 19.64 | 30.82 |
| Retained nitrogen (% ingested N) | 49.76 | 47.66 | 47.46 | 50.65 | 22.28 |

Mean values followed by different letters, in the same row, are significantly different by SNK test ($P < 0.05$).

Araujo (2012) tested similar levels of inclusion of CCW in diets for goats and reported that the use of CCW does not affect the use of nitrogen and guarantees sufficient nitrogen for physiological and reserve functions, which reflects this

positive nitrogen balance.

Higher levels of CCW (2.7 and 4.0%) promoted lower TR and TA and higher TO ($P < 0.05$), and the time with other activities was not influenced ($P > 0.05$) by SQB in diet (Table 5).

Table 5. Time spent with rumination, feeding, other activities and idle (hours) of lambs fed increasing levels of cow cheese whey (CCW) in the diet.

| Parameter (hours) | CCW inclusion (% DM) | | | | CV% |
|-------------------|----------------------|--------------------|---------------------|--------------------|-------|
| | 0 | 1.6 | 2.7 | 4.0 | |
| Rumination time | 7.72 ^a | 7.56 ^a | 6.96 ^{ab} | 5.68 ^b | 20.64 |
| Feeding time | 4.39 ^{ab} | 4.60 ^a | 3.50 ^{bc} | 3.42 ^c | 25.00 |
| Idle time | 11.12 ^{bc} | 10.68 ^c | 12.94 ^{ab} | 13.68 ^a | 18.99 |
| Other activities | 0.80 | 1.16 | 0.60 | 1.22 | 66.11 |

Mean values followed by different letters, in the same row, are significantly different by SNK test ($P < 0.05$).

The periods spent with food intake are interspersed with one or more periods of rumination or idleness, and the periods of rumination are controlled by the type of food, rising with a greater forage: concentrate ratio. Fiber requires more time in the digestion and rumination process, and thus greater amounts of NDF in the diet decrease the rate of passage through the digestive tract of the ruminant (CHURCH, 1979). Thus, the shorter feeding and rumination time is justified with the increasing inclusion of CCW as a result of lower NDF intake

and higher NFC intake (Table 3).

As a consequence, the diet with inclusion of CCW by 4% indicated lower BOL ($P < 0.05$) with a high correlation between the intake of NDF (g/UTM) and BOL ($r = 0.777623$; $P < 0.0001$), so that smaller amounts of NDF implies less BOL due to a smaller stimulus to chewing activity. However, the use of CCW did not interfere with the other parameters (FE, ERMS, ERFDN, TMT, MMb and tMMb), which shows the potential of this by-product in lambs feed (Table 6).

Table 6. Feed efficiency (FE), rumination efficiency in grams of DM/hour (ERMS) and in grams of NDF/hour (ERFDN), total chewing time (TMT in hours/day), number of daily cuds (BOL), chews per cud (MMb), time of chews per cud (tMMb in seconds/cud) in lambs fed increasing levels of cow cheese whey (CCW) in the diet.

| Component | CCW inclusion (% DM) | | | | CV (%) |
|-----------|----------------------|--------------------|--------------------|--------------------|--------|
| | 0 | 1.6 | 2.7 | 4.0 | |
| FE | 339.22 | 301.32 | 334.63 | 407.41 | 31.85 |
| ERMS | 179.22 | 179.67 | 174.41 | 234.86 | 21.99 |
| ERFDN | 92.30 | 86.88 | 79.54 | 103.92 | 23.03 |
| TMT | 12.11 | 12.16 | 10.46 | 9.10 | 18.65 |
| BOL | 44.64 ^a | 39.30 ^a | 36.27 ^a | 23.59 ^b | 17.36 |
| MMb | 154.17 | 124.89 | 170.20 | 162.47 | 19.65 |
| tMMb | 98.59 | 106.65 | 112.16 | 106.13 | 15.23 |

Mean values followed by different letters, in the same row, are significantly different by SNK test (P <0.05).

There was no interaction (P > 0.05) between the inclusion of CCW and the collection times for NH₃-N, ruminal pH, serum urea and total proteins (Table 7). However, inclusion with 4% CCW showed lower NH₃-N. The higher intake of NFC with 4% CCW may have led to greater microbial protein synthesis, reducing the concentration of NH₃-N in the rumen fluid. At the same time, the increasing inclusion of CCW may have favored ruminal fermentation, since CCW proteins have high nutritional value, high content of essential amino

acids, especially branched chain (COSTA *et al.*, 2010). The concentrations of NH₃-N were within the range for an adequate microbial fermentation (> 10 mg/100 mL), according to Van Soest (1994).

Values of pH between six and seven allow the presence of all components of the rumen microbial biomass, whether bacteria, fungi or protozoa (VAN SOEST, 1994). Thus, all pH values in this test were within the normal range (Table 7).

Table 7. Concentrations of ammonia nitrogen (mg/100 mL) and ruminal fluid pH, werum urea and total serum protein of lambs fed diets with increasing levels of cow cheese whey (SQB) in postprandial periods.

| Time (hours) | CCW inclusion (% DM) | | | | Means |
|---|----------------------|--------------------|--------------------|--------------------|--------------------|
| | 0 | 1.6 | 2.7 | 4.0 | |
| <i>Ammonia nitrogen (mg/100 mL) (CV = 29.67%)</i> | | | | | |
| 0 | 16.21 | 13.60 | 13.85 | 10.61 | 13.57 ^B |
| 3 | 16.77 | 19.36 | 20.36 | 13.44 | 17.48 ^A |
| 6 | 14.98 | 15.05 | 15.05 | 11.33 | 14.10 ^B |
| 9 | 18.10 | 15.60 | 18.52 | 14.76 | 16.74 ^B |
| Mean | 16.51 ^a | 15.60 ^a | 16.94 ^a | 12.54 ^b | - |
| <i>Ruminal fluid pH (CV = 6.75%)</i> | | | | | |
| 0 | 6.87 | 7.00 | 6.87 | 7.44 | 7.05 ^A |
| 3 | 6.65 | 6.62 | 6.25 | 6.58 | 6.53 ^B |
| 6 | 6.10 | 6.11 | 6.25 | 6.94 | 6.35 ^B |
| 9 | 6.04 | 6.21 | 6.71 | 6.13 | 6.02 ^C |
| Mean | 6.41 ^{ab} | 6.49 ^{ab} | 6.27 ^b | 6.77 ^a | - |
| <i>Serum urea (mg/100 mL) (CV = 28.63%)</i> | | | | | |
| 0 | 50.73 | 46.62 | 46.36 | 41.12 | 46.21 ^B |
| 3 | 30.52 | 34.95 | 54.59 | 41.55 | 40.40 ^B |
| 6 | 29.56 | 26.68 | 38.99 | 35.51 | 32.68 ^C |
| 9 | 47.39 | 52.24 | 73.31 | 43.55 | 54.12 ^A |
| Mean | 39.55 ^b | 40.12 ^b | 53.31 ^a | 40.43 ^b | - |
| <i>Total protein (mg/100 mL) (CV = 17.13%)</i> | | | | | |
| 0 | 8.35 | 8.39 | 8.63 | 6.53 | 7.97 ^{AB} |
| 3 | 8.33 | 8.79 | 7.40 | 6.62 | 7.78 ^B |
| 6 | 7.20 | 8.62 | 9.95 | 8.39 | 8.54 ^A |
| 9 | 4.84 | 4.32 | 4.18 | 4.11 | 4.36 ^C |
| Mean | 7.18 ^a | 7.53 ^a | 7.54 ^a | 6.41 ^b | - |

Mean values followed by different letters, in the same column, are significantly different by SNK test (P <0.05).

Mean values followed by different letters, in the same row, are significantly different by SNK test (P <0.05).

For the diet with 4.0% CCW, the pH was higher than with 2.7% CCW inclusion. Within the times, it can be seen that at time zero there was a higher pH increase, being higher than the other times. The greatest decline in pH occurred at time 9, probably because CCW contains high levels of lactose and soluble proteins with a high fermentation rate, resulting in a greater amount of lactic acid from the fermentation of these nutrients.

There was an increase in urea levels nine hours after supplying the diets and in the diets with 2.7% inclusion of CCW (P <0.05). Concentrations of serum urea can give an indication of ruminal protein availability and adequate protein supply in the diet, and values below normal can indicate deficiency in food or pathological states. Above normal values indicate pathological states, but can also indicate low efficiency in the use of ammonia available in the rumen due to a

possible lack of fermentable energy. Nevertheless, urea levels were above the minimum required (18 mg/100 mL) and in some points higher than the maximum level (31 mg/100 mL), the greater release of urea in plasma may be an indication of smaller use of ammonia nitrogen released in the rumen for microbial protein synthesis (LIMA *et al.*, 2012). Serum levels of total proteins in this study were within the normal range (6.0 and 7.9 g/100 mL), except for time 9, which presented a lower concentration (VAN SOEST, 1994). The increased concentration of amino acids in the blood does not indicate improved protein nutrition of the animal, and the improvement in the balance of amino acids results in its decrease in the blood. After these nutritional, behavioral and blood evaluations, CCW was used in performance tests at Embrapa Goat and Sheep, showing a positive satisfactory effect as an excellent alternative for feeding finishing lambs (Rogério *et al.*, 2017), with its use only dependent on availability in the local region of the production system. Interesting performances have also been proven in studies with CCW for goats (Leite *et al.*, 2015) and in liquid diet in pre-slaughter management of sheep (Silva *et al.*, 2018).

CONCLUSIONS

CCW has the potential to feed lambs because it does not interfere with nutrient intake and digestibility and promotes a positive nitrogen balance. It is indicated the inclusion of up to 4.0% DM of CCW in the lambs diet.

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