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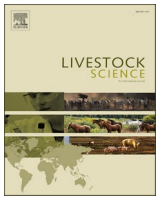

Ruminant Nutrition

Licuri oil improves feedlot performance and modifies ruminal fauna of Santa Inês ewes

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HIGHLIGHTS

- Licuri oil is an alternative to meet the energy demand of the diet provided to ewe.
- Different levels of licuri oil in diets for Santa Inês ewes were tested.
- Diets containing up to 2% licuri oil increase the DM intake and weight gain in ewes.

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ABSTRACT

The aim of this study was to evaluate the effect of inclusion of licuri oil on intake, digestibility, ingestive behavior, rumen protozoa population, and productive performance of Santa Inês ewe. Thirty-two Santa Inês ewe (multiparous, non-lactating, 2–4 years old and 36.7 ± 0.87 kg body weight) were distributed in a randomized block design, receiving diets containing licuri oil (0, 2, 4, and 5% based on total dry matter) in partial replacement of ground corn (n = 8 per treatment). The inclusion of licuri oil promoted a quadratic effect for the intakes of dry matter ($P = 0.008$), and neutral detergent fiber ($P = 0.004$), dry matter digestibility ($P = 0.004$), ether extract ($P < 0.0001$), average daily gain ($P = 0.01$), rumination time (min/day; $P = 0.039$ and min/g DM; $P = 0.041$), chewing time (min/g DM; $P = 0.020$), and for protozoa counts of the genus *Entodinium* ($P < 0.0001$). Ewe fed diets containing licuri oil showed higher consumption of ether extract ($P < 0.0001$), feed conversion ($P = 0.004$), and rumen pH ($P < 0.0001$), in contrast, a reduction in digestibility was observed neutral detergent fiber ($P = 0.002$) and total population of protozoa ($P < 0.0001$) in relation to those fed the control diet. In experimental conditions, it is possible to include licuri oil up to 2% in diets with 50% roughage offered to Santa Inês ewe to provide an increase in the intake and digestibility of dry matter and neutral detergent fiber, greater weight gain, and greater total protozoa count.

1. Introduction

Confinement of less specialized animals like culling ewe requires new strategies to reduce feeding costs. Additionally, the high costs of inputs, such as corn and soybean meal, widely used in animal nutrition, increase production costs (Goes et al., 2019). As a result, it is necessary to use alternative and good quality food that will reduce the costs associated with food and, at the same time, improve the efficiency and competitiveness of production systems (Pinto et al., 2020).

The inclusion of vegetable oils as alternative ingredients in the ewe diet is considered a promising approach in feedlot systems, not only because it provides essential fatty acids and fat-soluble vitamins, but

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also because it increases the energy density of diets and modifies the fatty acid profile of products of animal origin, decreasing the content of short chain fatty acids (SCFA) and increasing the content of vaccenic (18:1 trans11) and rumen (18:2 cis9 trans11) acids (Toral et al., 2018; Meschiatti et al., 2019; Nudda et al., 2020). Thus, vegetable oils have the potential to reduce caloric increase by increasing efficiency in the use of metabolizable energy for animal productive performance (Parente et al., 2020).

However, factors related to lipid sources and concentrations can decrease feed intake and fiber digestibility in ewe, so that the inclusion of levels above 70 g/kg DM can interfere with the ruminal microbiota (Lima et al., 2018; Candyrine et al., 2019) and consequently, modify the protozoan population in the rumen, affecting methane production. This is due to the fact that lipids have toxic effects on ruminal bacteria (Parente et al., 2018), which limits the use of these sources in diets.

The oil extracted from licuri can represent an important alternative to meet the energy demands of the diet supplied to Santa Inês ewe. This oil is extracted from the fruit of a palm tree (*Syagrus coronata* (Martius) Beccari) endemic to the Brazilian semi-arid region, found mainly to the east of the São Francisco River, in the states of Alagoas, Bahia, Pernambuco, Sergipe, and northern Minas Gerais (Noblick, 2017; Araújo et al., 2019) and which has great energy potential. Licuri, whose almond has a high oil content (54% of the almond weight) (Rodrigues et al., 2020), although it is little known as an additive for animal feed (Lima et al., 2015), is quite appreciated in the manufacture of cosmetics and soaps (Daza et al., 2020).

Of the total fatty acids present in licuri oil, more than 40% are made up of lauric acid (C12:0) (Lisboa et al., 2020). This fatty acid has a strong effect on ruminal defaunation, in addition to facilitating digestion and the ability to assist in reducing and controlling blood cholesterol levels. Thus, the inclusion of licuri oil in the diet of ewe may affect ruminal biohydrogenation, favor greater absorption of polyunsaturated fatty acids (Araújo et al., 2020), and promote a greater concentration of conjugated linoleic acid (CLA) (Morais et al., 2017) and α-linolenic fatty acids (Bianchi et al., 2017), in addition to influencing fat deposition in tissues (Miltko et al., 2019; Vargas et al., 2020).

To the best of our knowledge, the potential of licuri oil in diets for ewes, has not been sufficiently studied. Thus, we hypothesize that the inclusion of licuri oil in the diet of ewe increases energy density and reduces the dry matter intake without impairing animal performance. The aim of this study was to evaluate the effect of inclusion of licuri oil in the form of licuri oil on intake, digestibility, ingestive behavior, rumen protozoa population, and productive performance of ewe.

2. Material and methods

2.1. Experiment location and ethical aspects

The experiment was conducted at the premises of the Laboratory of Animal Requirement and Metabolism (LEMA) belonging to the Agricultural Sciences Campus of the Federal University of the São Francisco Valley (CCA/UNIVASF), Petrolina, Pernambuco, Brazil. The climate, according to the classification of Köppen and Geiger (1928), is of the hot semi-arid type, with a rainy season (BSh), with an average annual precipitation of 376 mm. During the experimental period, the maximum and minimum temperatures were 33.83 and 24.56°C, respectively, with relative humidity between 50.50% and 73.56%.

This research was approved and certified by the Ethics Committee on Human and Animal Studies of UNIVASF (protocol n° 0002/241017).

2.2. Animals, treatments, and experimental diets

Thirty-two Santa Inês ewes (multiparous, non-lactating, 2–4 years old, and 36.7 ± 0.87 kg body weight), were distributed in individual pens (2.42 m^2), equipped with drinking fountains and feeders. The experimental design used was in randomized blocks, with four treatments (diets) and eight repetitions per treatment. The initial body weight was used to define the blocks.

The confinement was carried out in a hollow shed (without side walls), with a beaten floor and covered with metal tiles. The experimental period lasted 77 days, preceded by 15 days to adapt the animals to the facilities, experimental diets, and handling. At the beginning of the adaptation period, the animals were identified, weighed, treated against endo and ectoparasites, and randomly allocated to the bays previously identified according to the treatments.

The treatments consisted of increasing levels of licuri oil in the diets (0, 2, 4, and 5%), partially replacing the ground corn, based on the total dry matter. The experimental diets were composed of elephant grass (*Pennisetum purpureum* Schun) *in natura*, ground corn, soybean meal, mineral mixture (Ovinofós, Tortuga, São Paulo, Brazil), dicalcium phosphate, and licuri oil, formulated to be isoproteic, with a 50:50 roughage:concentrate ratio (Table 1), to obtain daily weight gains of 40 g/day, following the recommendations of the NRC (2007), since the same study was evaluated concomitantly to collect test data reproductive system for harvesting oocysts in which high weight gain could impair the responses of the response variables.

The licuri oil used in making the diets was obtained by cold extraction of the almonds from the licuri fruit, preheated, decanted, filtered, and processed at Escola Família Agrícola do Sertão (EFASE), located in the municipality of Monte Santo – BA, Brazil. Samples of licuri oil were collected to determine the fatty acids profile through gas chromatography, adopting the methodology described by Visentainer (2012) (Table 2).

The diets were provided in the form of complete ration twice a day, at 0900 and 1600, and water was provided *ad libitum*. The leftovers of food offered were collected and weighed to determine the intake and to adjust the dry matter intake in order to allow 10% leftovers in the trough. Weekly samples of the food offered and leftovers were collected weekly for chemical analysis.

Table 1
Proportion of ingredients and chemical composition of experimental diets.

Ingredients (% DM)	Inclusion levels of licuri oil (% DM)			
	0	2	4	5
Ground corn	34.3	32.3	29.8	28.5
Soybean meal	13.8	13.8	14.3	14.6
Licuri oil	0	2.0	4.0	5.0
Mineral mixture[1]	1.6	1.6	1.6	1.6
Dicalcium phosphate	0.3	0.3	0.3	0.3
Elephant grass	50.0	50.0	50.0	50.0
Chemical composition (% DM)				
Dry matter[2]	54.9	55.1	55.3	56.1
Ash	8.6	8.6	8.7	8.7
Crude protein	11.6	11.5	11.5	11.6
Ether extract	2.0	3.9	5.8	6.8
Neutral detergent fiber	47.2	46.6	45.9	45.5
Metabolizable energy – SRNS (Mcal/kg)	2.5	2.6	2.7	2.7
Fatty acids (%)				
Caproic (C6:0)	0.045	0.042	0.039	0.039
Capric (C10:0)	0.044	0.169	0.294	0.357
Lauric (C12:0)	1.569	2.409	3.245	3.667
Miristic (C14:0)	0.493	0.779	1.064	1.207
Palmitic (C16:0)	19.539	19.463	19.396	19.364
Palmitoleic (C16:1)	0.144	0.142	0.139	0.138
Heptadecanoic (C17:0)	0.141	0.140	0.140	0.139
Stearic (C18:0)	3.447	3.489	3.543	3.570
Oleic (C18:1n9c/t)	13.515	13.290	13.027	12.892
Linoleic (C18:2n6c)	36.135	35.017	33.883	33.315
α-Linolenic (C18:3)	18.496	18.464	18.474	18.483
Archaic (C20:0)	0.139	0.136	0.133	0.132
Eicosanoic/Gadoleic (C20:1)	0.062	0.060	0.057	0.056
Dihomo-γ-linolenic (C20:3)	0.073	0.071	0.070	0.069

[1] Composition: 7.5% P; 19% Ca; 1% Mg; 7% S; 14.3% Na; 21.8% Cl; 500 ppm Fe; 300 ppm Cu; 4600 ppm Zn; 1100 ppm Mn; 80 ppm I; 405 ppm Co; 30 ppm Se.
[2] in% of natural matter.

Table 2
Fatty acid profile (in %) of ingredients used in experimental diets.

	Licuri oil	Ground corn	Soybean meal	Elephant grass
Fatty acids (%)				
Caproic (C6:0)	-	0.11	0.05	-
Capric (C10:0)	6.27	0.02	0.09	0.04
Lauric (C12:0)	41.97	0.01	-	3.13
Miristic (C14:0)	14.35	0.08	0.14	0.89
Palmitic (C16:0)	7.46	11.25	13.06	27.75
Palmitoleic (C16:1)	0.03	0.14	0.13	0.15
Heptadecanoic (C17:0)	-	0.04	0.13	0.21
Stearic (C18:0)	4.22	2.07	4.08	4.34
Oleic (C18:1n9c/t)	12.47	23.74	16.19	6.27
Linoleic (C18:2n6c)	2.53	58.41	55.20	16.96
α-Linolenic (C18:3)	-	1.62	10.09	33.09
Archaic (C20:0)	0.14	0.29	0.29	-
Eicosanoic/Gadoleic (C20:1)	0.03	0.14	0.10	-
Dihomo-γ-linolenic (C20:3)	-	0.10	0.28	-

*Ground corn and Soybean meal (Lee et al. 2013); Elephant grass (Araújo 2020)

2.3. Intake and apparent digestibility

The daily dry matter intake (DMI) was obtained by the difference between the total DM of feed intake and the total DM present in the leftovers. Nutrient intake was determined as the difference between the total nutrients present in the feed intake and the total nutrients present in the leftovers, on a total DM basis.

A digestibility test was carried out in the final third of the experimental period, lasting five days of collection. Fecal samples were collected directly from the final portion of the rectum of each animal, every two hours (Chizzotti et al., 2007). The feces were weighed and a subsample of 10% of the total amount of feces was collected to form a composite sample for each treatment. The samples were stored at −20°C, for further laboratory analysis.

2.4. Chemical analysis

Samples of the ingredients, diets, leftovers, and feces were pre-dried in a forced ventilation oven at 55°C for 72-h and ground into 1 mm and 2 mm particles (Wiley Mill, Marconi, MA-580, Piracicaba, Brazil). Chemical analyses were performed using the procedures described by the AOAC (2016) for dry matter (DM; method 967.03), ash (method 942.05), crude protein (CP; method 981.10), and ether extract (EE; method 920.29). The neutral detergent fiber (NDF) was determined according to Mertens (2002).

The apparent digestibility coefficient of nutrients was estimated using indigestible neutral detergent fiber (NDFi) as an internal indicator to predict the production of fecal DM (Cochran et al., 1986). Samples of feces, food, and leftovers were incubated *in situ* in the rumen of adult Sindi cattle (± 550 kg), previously adapted for 10 days to a diet containing licuri oil, for a period of 240-h in bags of TNT (tissue non-woven, with 100 g/m^2), in duplicate (Casali et al., 2008). After incubation, the remaining material was subjected to extraction with neutral detergent (Mertens, 2002) and the residue was considered as NDFi. The dry matter digestibility coefficient (DMDC) was calculated according to the formula (Cochran et al., 1986):

$$DMDC = [1 - (\%NDFiDMingested \,/\, \%NDFiDMfeces)] * 100$$

2.5. Ingestive behavior

Ingestive behavior was assessed at the beginning, middle, and end of the experimental period. For this evaluation, all animals were observed visually for 24-h, with observations recorded at intervals of five minutes. A record of water intake (W), food (F), rumination (R), and idleness (I) activities were collected by trained observers using digital timers. Artificial lighting was used to assist in nighttime assessments. The results for the behavioral variables of feeding were obtained using equations adapted from Bürger et al. (2000). The ingestion, rumination, and chewing times per gram of DM ingested and per gram of neutral detergent fiber ingested were calculated considering the dry matter intake and the neutral detergent fiber intake in the respective days assessment of ingestive behavior.

2.6. Productive performance

The animals were weighed at the beginning, weekly, and at the end of the experimental period, after a 16-h period of solid food deprivation (with access to water), to obtain the final body weight and average daily gain. Feed conversion was calculated using the following equation:

$$FC = drymatterintake/averagedailygain$$

2.7. Collection of rumen fluid

After 77 experimental days, the animals were subjected to a fast for 16-h for slaughter. The animals were desensitized by electronarcosis and bled through section of the jugular vein and carotid artery according to Brazilian regulations for industrial and sanitary inspection of products of animal origin (Brazil, 2017). Collection of ruminal fluid was performed immediately after slaughter. The samples of ruminal fluid was obtained at 3 points of the ventral rumen sac using a 25 mL sterile tubes, which was subsequently homogenized and 1 mL was removed from it, being placed in sterile test tubes containing 2 mL of formaldehyde and transported in isothermal boxes.

2.8. Determination of pH, identification and counting of protozoa

The rumen fluid showed mean values of volatile fatty acids (VFA) of 17.64% acetic acid; 12.71% propionic acid; and 1.58% butyric acid. The pH of the ruminal liquid was measured immediately after collection, using a previously calibrated bench pH meter (Simpla PH140, São Leopoldo, Brazil). The total count of the protozoa present in ruminal fluid was performed according to the methodology described by Dehority (2003). The total protozoa count values were transformed into log (x + 1). For identification of these ruminal microorganisms, a drop of 10^{-1} or 10^{-2} dilution was taken and together with a drop of lugol was mounted on microscopy slides. In these slides, coverslips were placed for analysis of the microstructures of the protozoa using a 40x objective and classification according to the key described in Dehority (1977).

2.9. Statistical analysis

The results obtained were analyzed using PROC GLM of the Software Statistical Analysis System (SAS 9.0 statistical package, Cary, NC, USA) and subjected to analysis of variance and regression at 5% probability. Regression equations were estimated using the PROC REG procedure. The following statistical model was adopted:

$$Y = \mu + Bi + Tj + eij$$

where: Y = observed value of the variable; μ = overall mean; Bi = effect of block i; Tj = effect of licuri oil levels j; eij = residual error.

3. Results

The inclusion of licuri oil in the diets promoted a quadratic effect for DM ($P = 0.008$) and NDF ($P = 0.004$) intakes. A quadratic effect was also observed for DM ($P = 0.004$) and EE ($P < 0.0001$) digestibility coefficients (Table 3). Ewe fed diets containing licuri oil showed higher EE intake ($P < 0.0001$) and a reduction in NDF digestibility ($P = 0.002$), compared to those fed the control diet (Table 3).

Table 3
Average values of intake, digestibility coefficient of nutrients, and productive performance of Santa Inês ewes fed diets containing different levels of licuri oil.

Variables	Licuri oil levels (%)				SEM	*P* value	
	0	2	4	5		L	Q
	Intake (g/dia)						
Dry matter	1609.3	1758.9	1355.6	1307.3	50.44	0.002	0.008[1]
Neutral detergent fiber	980	960	780	780	5.27	0.001	0.004[2]
Ether extract	44	64	63	79	0.32	<0.0001[3]	0.257
	Digestibility (%)						
Dry matter	62.78	70.93	62.33	58.49	1.51	0.071	0.004[4]
Neutral detergent fiber	68.64	66.89	57.99	58.73	1.45	0.002[5]	0.677
Ether extract	71.31	84.90	72.19	67.98	1.77	0.007	<0.0001[6]
	Productive performance						
DWG (g/day)	91.31	103.97	63.96	49.58	5.98	0.002	0.01[7]
Feed conversion	18.20	17.07	22.59	27.40	1.33	0.004[8]	0.067

DWG – Daily weight gain; SEM = Standard error of the mean; L – Significant for linear effect; Q – Significant for quadratic effect.
Significant for the 5% probability level.
Equations: [1] $\hat{y}=1631.01+97.07x–34.68x^2$, $R^2=0.60$
[2] $\hat{y}=0.991−0.020x–0.005x^2$, $R^2=0.89$
[3] $\hat{y}=0.04+0.001x$, $R^2=0.82$
[4] $\hat{y}=63.19+6.09x−1.44x^2$, $R^2=0.54$
[5] $\hat{y}=69.46−2.32x$, $R^2=0.63$
[6] $\hat{y}=72.04+9.50x−2.14x^2$, $R^2=0.76$
[7] $\hat{y}=92.84+11.33x−4.16x^2$, $R^2=0.64$
[8] $\hat{y}=16.28+1.83x$, $R^2=0.36$.

The average daily gain showed a quadratic effect ($P = 0.01$), increasing from 0 to 2% of inclusion of licuri oil in the diets and then decreasing, with lower gains observed for diets containing 5% of licuri oil (Table 3). The increase in levels of licuri oil in the diets provided an increase in feed conversion of the ewe ($P = 0.004$), with the worst feed conversion observed for ewe that received diets containing 5% licuri oil in their composition (Table 3).

There was no difference between the levels of licuri oil tested in relation to the time that the ewe spent during feeding, chewing, idleness, and water intake (min/day) ($P > 0.05$). The time spent for rumination (min/day; $P = 0.039$ and min/g DM; $P = 0.041$) and chewing min/g DM; $P = 0.020$) were influenced by the presence of licuri oil in the diets, presenting a quadratic effect (Table 4).

The time spent by the ewe for feeding (min/g DM and min/g NDF), rumination (min/g NDF), and chewing (min/g NDF) were not affected by the inclusion of licuri oil in the diets ($P > 0.05$) (Table 4).

An effect of including licuri oil in the diets of the ewe was observed in the total population of protozoa present in the rumen liquid, with a linear decrease ($P < 0.0001$) being observed in the total population of protozoa with increasing levels of licuri oil in the diets. The reverse behavior was verified for ruminal pH, whose indices increased according to the increase in the levels of licuri oil in the diets, in relation to those receiving the control diet ($P < 0.0001$) (Table 5).

The population of protozoa of the genus *Entodinium* in the rumen fluid of the ewe showed a quadratic effect ($P < 0.0001$), increasing from 0 to 2% of inclusion of licuri oil in the diets (Table 5). There was no effect of the levels of licuri oil tested in relation to the population of protozoa of the genera *Diplodinium, Eodinium, Eudiplodinium*, and *Ostracdinium* ($P > 0.05$) (Table 5).

4. Discussion

Although the levels of EE in the diets were within the maximum limits (between 5 and 7%) established for ruminants (Lima et al., 2015; Lima et al., 2018), the reduction in the dry matter intake observed in ewe fed diets containing licuri oil is probably due to a regulatory response of the intake to the higher energy density of the diets in relation to the control diet. The reduction in the dry matter intake directly

Table 4
Ingestive behavior of Santa Inês ewe fed diets containing different levels of licuri oil.

Variables	Licuri oil levels (%)				SEM	*P* value	
	0	2	4	5		L	Q
Idleness(min/day)	571.25	682.50	727.50	647.50	24.50	0.117	0.087
Water (min/day)	15.00	15.00	11.25	2.50	2.50	0.099	0.302
Feeding							
Min/day	253.75	250,00	205.00	223.75	11.53	0.182	0.954
Min/g DM	0.16	0.13	0.11	0.15	0.01	0.394	0.116
Min/g NDF	0.53	0.57	0.64	0.39	0.03	0.470	0.094
Rumination							
Min/day	600.00	492.50	496.25	566.25	20.34	0.396	0.039[1]
Min/g DM	0.25	0.20	0.22	0.27	0.01	0.717	0.041[2]
Min/g NDF	1.24	1.12	1.55	0.99	0.07	0.868	0.303
Chewing							
Min/day	853.75	742.50	701.25	790.00	23.87	0.148	0.062
Min/g DM	0.56	0.38	0.39	0.54	0.03	0.600	0.020[3]
Min/g NDF	1.77	1.69	2.19	1.39	0.09	0.639	0.120

DM – Dry matter; NDF – neutral detergent fiber; SEM – Standard error of the mean; L – Significant for linear effect; Q – Significant for quadratic effect.
Significant for the 5% probability level.
Equations: [1] $\hat{y}=601.59−90.05x+16.43x^2$, $R^2=0.33$
[2] $\hat{y}=0.26−0.05x+0.01x^2$, $R^2=0.31$
[3] $\hat{y}=0.57−0.16x+0.03x^2$, $R^2=0.38$.

Table 5
pH and protozoa count present in rumen liquid from Santa Inês ewes fed diets containing different levels of licuri oil.

Variables	Licuri oil levels (%)				SEM	*P* value	
	0	2	4	5		L	Q
Entodinium (Log^{10}/mL)	5.3	5.4	5.1	5.0	0.03	<0.0001	<0.0001[1]
Diplodinium (Log^{10}/mL)	4.9	4.8	4.8	4.8	0.05	0.422	0.560
Eodinium (Log^{10}/mL)	4.8	4.7	4.6	4.6	0.05	0.136	0.798
Eudiplodinium (Log^{10}/mL)	1.2	1.8	1.6	0.0	0.35	0.370	0.121
Ostracdinium (Log^{10}/mL)	0.6	1.1	0.6	0.0	0.26	0.404	0.243
Others (Log^{10}/mL)	4.6	4.6	4.8	4.8	0.03	0.999	0.713
Total protozoa (Log^{10}/mL)	5.7	5.6	5.4	5.4	0.03	<0.0001[2]	0.461
pH ruminal	6.6	6.9	7.1	7.2	0.05	<0.0001[3]	0.656

SEM – Standard error of the mean; L – Significant for linear effect; Q – Significant for quadratic effect.
Significant for the 5% probability level.
Equations: [1] $\hat{y}=5.70-0.04x-0.0039x^2$, $R^2=0.74$.
[2] $\hat{y}=5.715-0.066x$, $R^2=0.74$
[3] $\hat{y}=6.624+0.118x$, $R^2=0.56$.

affected the performance of the animals, which reduced daily weight gain and showed worse feed conversion. However, all diets provided a daily weight gain above 40 g/day, established during the formulation of the diets offered to the ewe (Table 3).

The increase in the amount of medium chain fatty acids in diets may be associated with a reduction in the dry matter intake (Hristov et al., 2011). The licuri oil used in our study is largely composed of medium chain fatty acids, with a predominance of lauric (41.97%) and myristic (14.35%) acids (Table 2). These fatty acids, absorbed by the abomasum, reaching the liver through the hepatic portal system, have a high propensity to oxidation, behaving similarly to glucose (Daza et al., 2020). As metabolic fuels that reach the liver can increase satiety (Maher and Clegg, 2019), the presence of licuri oil in diets increased the EE intake, which may have decreased the dry matter intake by a post-absorptive satiety mechanism.

There is interaction between intake, digestibility, and rate of passage. Therefore, it is to be expected that any treatment that changes intake will result in a change in the rate of passage and therefore, the digestibility of nutrients. The increase in the proportion of short and medium chain fatty acids and unsaturated fatty acids in diets containing licuri oil may have caused an increase in cholecystokinin (CCK) production, which caused an increase in its concentration in plasma. CCK can suppress food intake by inhibiting gastric emptying, which consequently reduces motility and the rate of digestion through the gastrointestinal compartments (Bielak et al., 2016; Liu et al., 2020), as a result of the negative effect of the presence of fat in the rumen environment on microbial growth, especially cellulolytic microorganisms (Elghandour et al., 2019), which may have influenced the reduction in neutral detergent fiber digestibility (Table 3).

Considering that there is a negative correlation between short-chain fatty acids and ruminal pH (Shen et al., 2019), it is possible that a higher production of short-chain fatty acids from rumen fermentation may have been generated in ewe that received the control diet and the diet containing 2% licuri oil in its composition, given that the ruminal pH values were lower for these treatments (Table 5). The increase in the pH of the rumen content of ewe that received diets containing higher levels of licuri oil may be related to the partial replacement of corn grain by licuri oil (Table 1). Corn has, in its nutritional composition, rapidly fermentable carbohydrates, which contribute to the production of organic acids, in addition to reducing salivary secretion, which consequently leads to a reduction in ruminal pH (Ma et al., 2015).

The ruminal pH combined with the toxic effects of fatty acids present in licuri oil, mainly caprylic (10.35%), lauric (41.97%), myristic (14.35%), oleic (12.47%), and linoleic (2.53%), on the rumen microorganisms, may also have contributed to the defaunation of the total rumen protozoa population, mainly of the genus *Entodinium* in inclusions above 2% of licuri oil in the diets offered to the ewe. Entodiniomorphic ciliated protozoa have high hemicellulolytic and cellulolytic activity that act directly on the degradation of dietary fiber, colonizing fiber particles, directly ingesting plant tissues, and facilitating the action of specific bacteria (Vargas et al., 2020). The elimination or reduction of protozoa of this genus reduces their digestive activity, due to the lower carboxymethyl cellulase activity in the rumen, directly affecting fiber digestibility (Hristov et al., 2009), a fact observed in the present study.

The direct consequence of defaunation is a drop in the concentration of ammoniacal nitrogen due to a reduction of the proteolytic activity of the protozoa. The decrease in the number of protozoa is generally associated with a reduction in nitrogen recycling in the rumen environment, with an increase in the number of gram negative bacteria and a decrease in the ammonia concentration (Hristov et al., 2019). In this context, the use of licuri oil in diets offered to ewe can be considered a promising strategy to increase the efficiency of feedlots, bringing environmental benefits resulting from the reduction of methane production by 20–30%, and increasing the availability of metabolizable energy for animals by about 12% (Nguyen et al., 2016; Vargas et al., 2020).

5. Conclusion

In experimental conditions, it is possible to include licuri oil up to 2% in diets for ewes provided increase of intake, digestibility of dry matter and neutral detergent fiber, greater weight gain, and a higher total protozoa count.

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Declaration of Competing Interest

The authors declare that they have no competing interests.

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