

**Animal feeding and nutrition**

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# Effect of partial replacement of *Dichanthium* spp. hay with *Guazuma ulmifolia* foliage on hair lambs' intake, digestibility, and blood metabolites

Efecto del reemplazo parcial del heno *Dichanthium* spp. por follaje de *Guazuma ulmifolia* sobre la ingesta, la digestibilidad y los metabolitos sanguíneos de corderos de pelo

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**Abstract:** Extensive livestock management is a common practice in the dry tropics. However, summer conditions can cause high fiber and low protein concentrations, even in grasses adapted to tropical soils, such as Angleton (*Dichanthium* spp.), widely used in producing small ruminants. On the other hand, Guacimo (*Guazuma ulmifolia*) is a tree that overcomes high temperatures and hydric stress. This research evaluated the effect of replacing Angleton with four foliage levels of Guacimo on hair lambs' intake, digestibility, and blood metabolites. Twelve male hair lambs with an average of  $22.0 \pm 1.3$  kg of body weight were used in an experimental  $4 \times 4$  square design. Treatments were as follows: 100 % Angleton hay; 85 % Angleton hay + 15 % Guacimo foliage; 70 % Angleton hay + 30 % Guacimo foliage, and 55 % Angleton hay + 45 % Guacimo foliage. Dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) intake, as well as CP digestibility and glucose concentrations, increased linearly as the level of Guacimo rose ( $P > 0.05$ ). Guacimo foliage improves intake, blood metabolites, and *in vivo* digestibility of DM and can be used to replace part of the hay in lamb rations.

**Keywords:** animal nutrition, foliage, silvopastoral systems, small ruminants, tropical forests.

**Resumen:** La ganadería extensiva es una práctica común en los trópicos secos. Sin embargo, las condiciones de verano pueden provocar una alta concentración de fibra y una baja concentración de proteína en gramíneas adaptadas a suelos tropicales como el Angleton (*Dichanthium* spp.), especie utilizada en la producción de pequeños rumiantes. Por otro lado, el Guácimo (*Guazuma ulmifolia*) es un árbol que supera las altas temperaturas y el estrés hídrico. El objetivo de esta investigación fue evaluar la sustitución de Angleton por cuatro niveles foliares de Guácimo sobre la ingesta, la digestibilidad y los metabolitos sanguíneos en corderos de pelo. Se utilizaron doce corderos de pelo machos con  $22,0 \pm 1,3$  kg de peso corporal en un diseño experimental de  $4 \times 4$ . Los tratamientos fueron: 100 % heno de Angleton; 85 % heno de Angleton + 15 % follaje de Guácimo; 70 % heno de Angleton + 30 % follaje de Guácimo; y 55 % heno de Angleton + 45 % follaje de Guácimo. La ingesta de materia seca (MS), materia orgánica (MO), proteína cruda (PC) y fibra de detergente neutro (FDN), así como la digestibilidad de proteína bruta y las concentraciones de glucosa, aumentó linealmente a medida que incrementó el nivel de Guácimo ( $P > 0,05$ ). El follaje de Guácimo mejoró la ingesta, los metabolitos sanguíneos y la digestibilidad *in vivo* de la MS, por lo que puede usarse para reemplazar parte del heno en dietas para corderos.

**Palabras clave:** bosque tropical, follaje, nutrición animal, pequeños ruminantes, sistemas silvopastoriles.



## Introduction

In tropical dry forests, small ruminant productions are limited by low rainfall and high temperatures, which considerably reduce the nutritional properties of forages (Ulukan, 2011). Therefore, it is necessary to look for alternative feedings, such as silvopastoral systems or the inclusion of tree leaves and fruits, that improve animal performance and mitigate the impact of the dry season (Castrejón-Pineda et al., 2016). In addition, *Dichanthium* spp. is one of the most used forages by small farmers in the Colombian tropical dry forest, and it is often used as a primary source for small ruminant feeding. However, its offer and nutritional quality are reduced during the dry season, limiting its performance.

*Guaçuma ulmifolia* is an adaptable tree of the American tropic that can withstand adverse climatic conditions such as high temperatures or hydric stress (Calzavara et al., 2017; Villa et al., 2009). It is considered a multipurpose tree due to the wide variety of products and services that it provides to agroforestry, agriculture, livestock farming, and alternative medicine (Manríquez-Mendoza et al., 2011). Studies have shown the *Guaçuma ulmifolia* as an alternative for ruminant feeding, either partially replacing soybean meal (*Glycine max*) in sheep production (Castrejón-Pineda et al., 2016) or being used as an alternative source of forage biomass in silvopastoral systems (Nicodemo et al., 2016; Villanueva et al., 2016). Nonetheless, there is a lack of information about the nutritional value, digestibility, and ruminal kinetic degradability of *Guaçuma ulmifolia* leaves in hair lambs. Therefore, this research aimed to evaluate the effect of replacement *Dichanthium* spp. hay by *G. ulmifolia* leaves on *in vivo* and *in vitro* digestibility, ruminal kinetics, and blood metabolites in hair lambs.

## Materials and methods

### Location

This research was carried out in the experimental farm “Las Brisas” located in Ibagué, Tolima province, Colombia (4°25' 35.5" N 75°13'47.8" W), at 1,285 m a.s.l., with an average temperature of 22 °C, relative humidity of 94 %, and annual rainfall of 1,620 mm. It is classified as tropical dry forest according to Holdridge (1978).

*G. ulmifolia* leaves were obtained from the farm “El Recreo” located in Guamo, Tolima, Colombia (4°00'18.0" N 74°58' 47.2" W), at 321 m a.s.l., with an average temperature of 28 °C, relative humidity of 74.9 %, and annual rainfall between 1,000 and 1,400 mm. Samples of adult trees were taken manually; the leaves were dried in the shade and then stockpiled in sealed containers for later use in the experimental diets.

### Animals and experimental design

Twelve non-castrated hair lambs  $22.0 \pm 1.3$  kg were assigned to a Latin square design (4 × 4) of four treatments, four periods, and three animals in each experimental group. The treatments consisted of increasing levels of Guacimo leaves (*G. ulmifolia*) by replacing *Dichanthium* spp. hay (aged 90 days) as follows: T1 = 100 % *Dichanthium* spp. hay, T2 = 85 % *Dichanthium* spp. hay and

15 % *G. ulmifolia* leaves; T3 = 70 % *Dichanthium* spp. hay and 30 % *G. ulmifolia* leaves, and T4 = 55 % *Dichanthium* spp. hay and 45 % *G. ulmifolia* leaves. Each experimental period lasted 21 days: 17 days for adaptation to the experimental diet and four days for the sample collection. The animals were confined in individual metabolic cages (1 m in height, 0.8 m in width, and 1.5 m in length) equipped with collectors, feces and urine separators, individual feeders, and troughs.

### Chemical properties of the diets

The assigned diets, feces, and orts were determined by proximal chemical analysis according to the methods established by the Association of Analytical Communities (AOAC) (2012) for dry matter (DM), organic matter (OM), crude protein (CP), and mineral matter (MM) content. The neutral detergent fiber (NDF) and acid detergent fiber (ADF) were found based on the protocol of Van Soest et al. (1991; Table 1).

**Table 1.** Nutrient composition of ingredients and experimental diets as DM %

%	<i>Dichanthium</i>	<i>G. ulmifolia</i>	Treatments			
	spp. hay	leaves	1	2	3	4
DM	90.41	30	90.41	90.35	90.29	90.23
OM	89.75	89.90	89.75	89.77	89.80	89.82
CP	3.56	12.80	3.56	4.95	6.33	7.72
NDF	75.49	61.44	75.49	73.38	71.28	69.17
ADF	39.27	39.22	39.27	39.26	39.26	39.25
MM	10.25	10.1	10.25	10.23	10.21	10.18

*Note.* Diets formulated for the study based on the DM intake. Proportions were as follows: T1 = 100 % *Dichanthium* spp. hay; T2 = 85 % *Dichanthium* spp. hay, 15 % *G. ulmifolia* leaves; T3 = 70 % *Dichanthium* spp. hay, 30 % *G. ulmifolia* leaves; T4 = 55 % *Dichanthium* spp. hay, 45 % *G. ulmifolia* leaves.

Source: Prepared by the authors

### Dry matter intake

The treatments were calculated based on 4 % of the live weight of the animals. The feed was provided in two daily meals at 7:00 A.M. and 3:00 P.M. in a sufficient quantity to obtain 5–10 % orts. The DM intake of each treatment was calculated during collection days, and orts were removed daily for each animal before the morning meal. This intake was expressed in animal

grams per day (ING g/d<sup>-1</sup>) concerning live weight (ING % LW) and metabolic weight (kg W 0.75) (ING % MW).

### ***In vivo* digestibility**

The animals had 17 days for adaptation to the experimental diets in individual pens per treatment, and they were offered 4 % of the live weight in DM, daily adjusted to obtain between 5 % and 10 % of orts. After this period, each animal was housed in a metabolic cage for four days for sample collection. Samples of diets, orts, and feces were dried for chemical analysis of DM, OM, NDF, and CP digestibility according to the previous bromatologically described methods.

### **Degradation kinetics and *in vitro* digestibility**

The *in vitro* DM digestibility (IVDMD) of the experimental diets was determined based on the Tilley and Terry (1963) technique adapted to the artificial rumen DAISY II<sup>®</sup>-ANKOM<sup>®</sup>. A Girolando breed was used to obtain the ruminal fluid (provided with a ruminal cannula) maintained in a *Cynodon* spp. pasture with 500 g/day of *G. ulmifolia* leaves. Next, 0.5 g samples of each diet (previously ground to 1 mm) were weighed and deposited in F57 ANKOM<sup>®</sup> filter bags distributed in four glass jars to which A and B buffer solutions and a ruminal inoculum were added. Then they were introduced in the DAISY II<sup>®</sup> incubator for 48 hours, guaranteeing a temperature of 39 °C. At the end of this period, 40 ml of 6N HCl and 8 g of pepsin (EC 3.4.23.1 Sigma<sup>®</sup>) were added, leaving the samples in the incubator for other 24 hours. Afterward, the bags were dried at 105 °C for eight hours. IVDMD was calculated by the difference between the incubated feed and the residue after incubation. The degradation kinetics were determined parallel to the IVDMD using DAISY II<sup>®</sup>, incubating each diet at 3, 6, 12, 24, 36, 48, 72, and 108 hours. The *in vitro* ruminal degradation parameters of the DM were calculated using Equation 1, as described by Ørskov and McDonald (1979):

$$p = \alpha + b(1 - \exp^{-ct}) \quad (1)$$

where p = rate of degradation at time t;  $\alpha$  = water-soluble fraction; b = water-insoluble, potentially degradable fraction; c = rate of degradation of fraction b; t = incubation time.

The DM's effective degradability (ED) was calculated using Equation 2:

$$ED = \alpha + (b \cdot c / c + k) \quad (2)$$

where k is the speed of the passage of particles in the rumen.

The ED of the DM *in vitro* was estimated for each diet, considering the passage rates of 2.5 and 8 %/hour (values that can be attributed to low, medium, and high intake levels), respectively. Potential degradability was calculated using Equation 3:

$$PD = a + b \quad (3)$$

Protein degradation kinetics were obtained by quantifying the nitrogen in *Dichanthium* spp. hay samples and *G. ulmifolia* leaves; 5-gram samples (previously ground) were weighed and deposited in F57 ANKOM® filter bags. These samples were incubated in the DAISY II® at 3, 6, 12, 24, 36, 48, 72, and 108 hours.

### Blood metabolites

The glucose, blood urea nitrogen (BUN), and beta-hydroxybutyric acid ( $\beta$ HBA) were determined for each animal on the last day of the experimental period utilizing a venipuncture of the jugular vein with Vacutainer™ tubes. The samples were refrigerated and analyzed using commercial Biosystems® kits (glucose and BUN and Randox® ( $\beta$ HBA) with the automated system of blood chemistry Biosystems A15® at the Veterinary Diagnostic Laboratory (LADIVE, for its acronym in Spanish) of the Universidad del Tolima.

### Secondary metabolites in *G. ulmifolia* leaves

The *G. ulmifolia* leaf samples were analyzed in the LASEREX laboratory of the Chemistry Department of the Universidad del Tolima, where the total phenols were quantified using the Folin-Ciocalteu method (Singleton et al., 1999). Tannins, flavonoids, and terpenes content were determined using the guide techniques described by Terrill et al. (1992) and modified by Del Pino et al. (2005; Table 2).

**Table 2.** Secondary metabolites of *G. ulmifolia* leaves used in the experiment

Metabolite	Test	Units	Sample
			<i>G. ulmifolia</i> leaves
Saponins	Foam	NA	+
	Rosentaler	NA	+
Total phenols	Spectrophotometry	mEq-tannic acid/g	6.47
	FeCl <sub>3</sub>	NA	++
Tannins	Gelatin-salt	NA	++
	Spectrophotometry	mEq-tannic acid/g	4.12
Flavonoids	Spectrophotometry	mEq-tannic acid/g	2.75
Terpenes	Lieberman-Buchard	NA	+
	Tanred	NA	++
Alkaloids	Dragendorff	NA	++
	Mayer	NA	++

Source: Prepared by the authors

### Statistical analysis

The results were interpreted by regression study using the PROC REG package of the statistical program Statistical Analysis System, version 9.1 (SAS). The *in vitro* ruminal degradation

parameters were estimated using the Gauss-Newton iterative process applying the SAS PROC NLIN package. Significance was set at  $P < 0.05$ .

## Results

### Intake and *in vivo* digestibility

There was a linear effect of DM, OM, NDF, and CP intake ( $P < 0.05$ ; Table 3) with the increase of replacing *Dichanthium* spp. hay with *G. ulmifolia* leaf levels in the diets. The DM intake in g/day rose from 609.47 (T1) to 762.66 (T4), respectively. Live weight and metabolic weight varied from 2.17 % to 2.99 % and 4.87 % to 6.57 %, respectively. The fecal production of DM, OM, NDF, and CP increased ( $P < 0.05$ ) with the *G. ulmifolia* leaf volume. The CP digestibility linearly increased ( $P < 0.05$ ) with the *G. ulmifolia* leaf batch. Meanwhile, the DM and OM digestibility did not vary between treatments ( $P > 0.05$ ); however, the NDF digestibility linearly decreased with the *G. ulmifolia* size replacing the hay in the diets, going from 58.6 (T1) to 55.4 (T4).

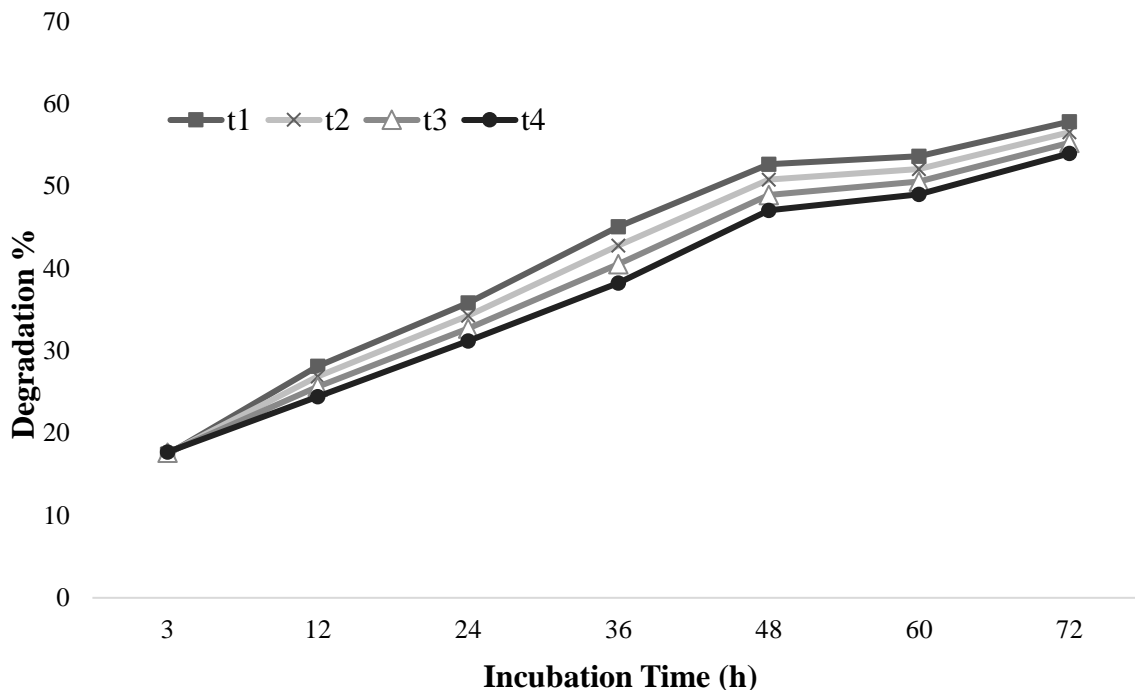
**Table 3.** Intake (INT), fecal excretion (FE), and digestibility (Dig) of dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), and crude protein (CP) in lambs fed with *G. ulmifolia* leaves

Parameters	Treatments				SEM	Significance level		
	1	2	3	4		L	Q	
DM	INT g.d <sup>-1</sup>	609.47	676.34	735.73	762.66	12.09	0.016	0.047
	INT % BW	2.17	2.54	2.98	2.99	0.11	0.045	0.161
	ING <sup>0.75</sup> % BW	4.87	5.63	6.52	6.57	0.21	0.035	0.081
	FE g.d <sup>-1</sup>	271.87	311.50	338.15	353.39	11.41	0.020	0.006
	Dig, %	55.08	55.04	54.10	53.79	1.53	0.055	0.303
OM	INT g.d <sup>-1</sup>	544.60	605.67	659.69	685.44	10.99	0.014	0.044
	FE g.d <sup>-1</sup>	233.08	264.74	286.29	299.72	9.64	0.017	0.009
	Dig, %	59.31	58.87	57.56	56.98	1.39	0.083	0.190
NDF	INT g.d <sup>-1</sup>	464.05	496.00	524.01	529.58	8.75	0.036	0.079
	FE g.d <sup>-1</sup>	191.14	207.84	214.98	220.34	7.60	0.037	0.079
	Dig, %	58.62	57.54	55.73	55.40	1.51	0.028	0.188
CP	INT g.d <sup>-1</sup>	25.27	39.77	55.42	68.27	0.44	0.001	0.027
	FE g.d <sup>-1</sup>	15.36	20.64	25.12	31.66	0.83	0.003	0.053
	Dig, %	44.05	51.53	54.26	54.94	1.45	0.019	0.070

Note. 1: *Dichanthium* spp. hay; 2: 85 % *Dichanthium* spp. hay, 15 % *G. ulmifolia* leaves; 3: 70 % *Dichanthium* spp. hay, 30 % *G. ulmifolia* leaves; 4: 55 % *Dichanthium* spp. hay, 45 % *G. ulmifolia* leaves. SEM: standard error mean; L: linear effect; Q: quadratic effect. Significance level < 0.05. Source: Prepared by the authors

### Degradation kinetics and *in vitro* digestibility

The CP degradation increased as the *G. ulmifolia* leaf supplementation increased (Figure 1).



**Figure 1.** *In vitro* degradation kinetics of protein in diets with different levels of *G. ulmifolia* leaves.

Source: Prepared by the authors

The *in vitro* dry matter degradability decreased linearly in each incubation period. This trend remained for all incubation schedules. From 36 hours of incubation, the control treatment had a degradability of 10 % above T4 (45 % *G. ulmifolia*). Additionally, we observed a linear decrease ( $P < 0.05$ ) of the IVDMD, while the levels of *G. ulmifolia* leaves in the diets increased, obtaining values of 51.51 % (T1), 48.8 % (T2), 46.1 % (T3), and 43.4 % (T4), respectively (Table 4).

The ruminal degradation parameters are presented in Table 5. The water-soluble fraction (a) decreased ( $P < 0.01$ ) as the level of inclusion of *G. ulmifolia* leaves increased, going from 13.31 % (T1) to 9.83 % (T4). However, the potentially degradable fraction (b) did not have significant differences ( $P > 0.05$ ). In the meantime, the inclusion levels of *G. ulmifolia* leaves increased in the diets while the effective and potential degradability increased linearly ( $P < 0.05$ ).

**Table 4.** *In vitro* degradability and digestibility of dry matter in diets with levels of *G. ulmifolia* leaves

Degradability%. Hour <sup>-1</sup>	Treatments				SEM	Significance level <i>L</i>
	T1	T2	T3	T4		
3	17.43	16.09	14.75	13.40	0.35	< 0.01
6	20.28	18.46	16.63	14.80	0.47	< 0.01
12	28.11	25.64	23.16	20.68	1.41	< 0.01
24	35.80	33.50	31.19	28.88	0.41	< 0.01
36	45.05	41.74	38.44	35.13	1.12	< 0.01
48	52.65	49.45	46.26	43.06	1.11	< 0.01
72	57.81	54.40	51.00	47.59	1.09	< 0.01
108	63.82	60.51	57.20	53.89	0.86	< 0.01
IVDMD %	51.51	48.81	46.11	43.4	1.26	0.01

Note. 1: *Dichanthium* spp. hay; 2: 85 % *Dichanthium* spp. hay, 15 % *G. ulmifolia* leaves; 3: 70 % *Dichanthium* spp. hay, 30 % *G. ulmifolia* leaves; 4: 55 % *Dichanthium* spp. hay, 45 % *G. ulmifolia* leaves; IVDMD %: *in vitro* dry matter digestibility; SEM: standard error mean.

Source: Prepared by the authors

**Table 5.** *In vitro* ruminal degradation parameters in diets with different levels of *G. ulmifolia* leaves

Parameters	Treatments				SEM	<i>P/L</i> >
	1	2	3	4		
a (%)	13.31	11.89	10.49	9.83	0.39	0.012
b (%)	52.60	50.17	47.78	46.65	0.92	0.094
c (%).h <sup>-1</sup>	0.025	0.024	0.023	0.023	0.0009	0.800
PD	65.91	62.06	58.27	56.49	1.10	0.003
ED (K = 0.02)	40.41	39.94	36.04	35.06	0.76	0.011
ED (K = 0.05)	29.36	28.79	25.55	24.79	0.63	0.008
ED (K = 0.08)	24.74	23.99	21.68	20.47	0.55	0.004

Note. 1: *Dichanthium* spp. hay; 2: 85 % *Dichanthium* spp. hay, 15 % *G. ulmifolia* leaves; 3: 70 % *Dichanthium* spp. hay, 30 % *G. ulmifolia* leaves; 4: 55 % *Dichanthium* spp. hay, *G. ulmifolia* leaves 45 %; a (%): water soluble fraction; b (%): water insoluble, potentially degradable fraction; c (%).h<sup>-1</sup>: degradation rate of an “a” fraction; PD: potential degradation; ED: effective degradation; SEM: standard error mean.

Source: Prepared by the authors

### Blood metabolites

There was an increasing linear effect on blood glucose values with the replacement levels of *G. ulmifolia* ( $P < 0.05$ ; Table 6). Although the BUN was greater in T4, the other treatments did not

differ. Regarding the blood  $\beta$ HBA in all the treatments with *G. ulmifolia*, the blood  $\beta$ HBA values were higher than the control treatment, but no linear effect was observed.

**Table 6.** Blood metabolites in lambs fed with different levels of *G. ulmifolia* leaves

Parameters	Treatments				SEM	Significance level	
	1	2	3	4		L	Q
Glucose (mg/dL)	54.25	56.58	57.67	60.17	1.53	0.010	0.909
BUN (mg/dL)	3.98	4.57	5.14	8.48	0.52	0.098	0.271
$\beta$ HBA (mmol/L)	0.22	0.35	0.36	0.36	0.04	0.190	0.230

Note. 1: *Dichanthium* spp. hay; 2: 85 % *Dichanthium* spp. hay, 15 % *G. ulmifolia* leaves; 3: 70 % *Dichanthium* spp. hay, 30 % *G. ulmifolia* leaves; 4: 55 % *Dichanthium* spp. hay, 45 % *G. ulmifolia* leaves; BUN: blood ureic nitrogen;  $\beta$ HBA: beta-hydroxybutyric acid; SEM: standard error mean. Source: Prepared by the authors

## Discussion

The increase in DM intake in diets replacing *Dichanthium* spp. hay with *G. ulmifolia* leaves is probably favored by its high palatability (Baumont, 1996). It is also reported by Villa et al. (2009), who observed a high *G. ulmifolia* intake in cattle. Likewise, a relationship exists with the lower NDF content in the *G. ulmifolia* leaves compared with the hay used in this study (Table 1), which would decrease the physical effect of the ruminal filling (Gebremariam et al., 2006). On the other hand, Nkosi et al. (2016) describe a direct relationship between digestibility and intake, also affecting growth; therefore, it can be deduced that the CP digestibility rose, favoring the intake of OM, NDF, and CP (Table 3). The hay used in the present study had an IVDMD % of 51.51 % compared to 59.61 % reported by Giraldo et al. (2007) for the same species, which is evidence of the loss of quality related to age. The NDF amount in *G. ulmifolia* leaves against the NDF of the 90-day *Dichanthium* spp. hay can be linked to improved CP digestibility by replacing the hay over the *G. ulmifolia* leaf percentage. In that sense, Slanac et al. (2011) reported that fiber lignification decreases the ruminal degradation level. Similarly, increasing age accentuates lignification. García-Castillo et al. (2008) showed DM degradability reductions between 7 % and 13 % in *Parmentiera edulis* fruit, as its maturity stage increases.

In the *in vitro* experiment, different tendencies than those *in vivo* were noted. The *in vitro* CP degradability was lower in *G. ulmifolia* leaves compared to *Dichanthium* spp. hay. Also, less IVDMD occurs with the *G. ulmifolia* leaf level compared to *in vivo* digestibility. This behavior could be explained by the tannin and phenol contents in the *G. ulmifolia* leaves, 4.12 mEq-tannic acid/g and 6.47 mEq-tannic acid/g, respectively (Table 2). Castro et al. (2006) reported tannin and phenol concentrations of 4.7 % and 2.8 %, respectively, in the *G. ulmifolia* leaves, and Rojas et al. (2015) showed a failure in the digestibility of *G. ulmifolia* leaves and Ayale (*Crescentia alata*) fruit related to high concentrations of secondary metabolites. Recent studies defend the

importance of tannins in feeding lambs because they favor the animals' health (García-Hernández et al., 2017; Peng et al., 2016). Nevertheless, they had been previously classified as anti-nutritional factors for decreasing protein bioavailability at the ruminal level (Huisman et al., 1990). This has also been reported by Otero and Hidalgo (2004), who explain that condensed tannins decrease the proteolysis the ruminal microflora performs due to its ability to form complexes with the proteins, preventing bacterial action. According to Min et al. (2003), when a pH of less than 3.5 is reached, the nutrients can be released from these complexes, allowing their absorption at the intestinal level.

Moreover, some proteins present in saliva as proline-rich proteins and histidine-rich proteins have a high affinity for condensed and hydrolyzable tannins, interacting with them and preventing the formation of tannin diet protein complexes (Shimada, 2006). According to Alonso-Díaz et al. (2012), lambs and goats have a high salivary protein concentration and are physiologically adapted to consume plants with high tannin levels. Also, the constant rumination action can increase the salivation and release of these proteins, which could explain the *in vivo* protein digestibility increase contrasted with the *in vitro* protein degradability by increasing the replacement with *G. ulmifolia* leaves.

The DM degradation kinetics showed a reduction in the water-soluble fraction (a) and a potentially degradable fraction (b) when the *G. ulmifolia* leaf content was increased in the diets. Probably for the *G. ulmifolia* leaves, the bacterial ruminal colonization phase described by Mertens (1993) would depend on external factors previously described (such as salivary proteins and pH intervention), which would delay the *in vitro* degradation kinetics.

The positive effect on blood glucose did not overcome the typical values that Galván Doria et al. (2014) reported in Colombian-bred lambs, where the glucose was 75.57 mg/dL in females and 83.70 mg/dL in males. However, the animals were fed low nutritional value diets in the present study, so these results were expected. The glucose increment can be correlated with DM and metabolizable energy intake, while the *G. Ulmifolia* levels increased in the diets (Przemysław et al., 2015). On the other hand, Knaus et al. (1998) affirm that the protein with surpassing properties (such as that found in *G. ulmifolia*) can increase glucose as an indirect effect of the increase in glucogenic precursors.

The high protein diets tend to raise the BUN concentration (Rufino et al., 2016), and it can be inferred that the BUN increase resulted from the more significant protein contribution in *G. ulmifolia* leaves, adding an insufficient DM intake. Nevertheless, the values in all treatments are below the reported standard by Singleton et al. (1999).  $\beta$ HBA has an inverse relationship to the blood glucose level, which has been reported in both cows (Erfle et al., 1974) and sheep (Knaus et al., 1998) in negative energy balance. Nonetheless, the  $\beta$ HBA values of the present study tended to increase with the replacement of hay with *G. ulmifolia* leaves in the diet (together with the blood glucose level), but this increase was below the standard (Galván Doria et al., 2014) (which reflects that all animals had low energy values); so  $\beta$ HBA was not stored in the adipose tissue of the animals, being free in the blood plasma. A similar phenomenon was observed by Penner et al. (2009) by increasing the amount of concentrate in the diet of cows in a positive energy balance. Steele et al. (2012) explain that the increase of  $\beta$ HBA in high levels of non-structural carbohydrate diets results from greater ketogenesis by the ruminal epithelium by having an alternative source of energy, which would increase hepatic gluconeogenesis and lower

use of  $\beta$ HBA. Despite the increase described, the  $\beta$ HBA body values follow the standard for lambs reported by Russel et al. (1967), which was less than 0.71 mmol/L for nutrition without energy deficit.

## Conclusions

The Guácimo (*G. ulmifolia*) leaf supplementation in hair lambs fed with low-quality hay can improve the *in vivo* DM, OM, NDF, and CP intake and their digestibility. Furthermore, some blood metabolites can increase. However, the *in vitro* digestibility and degradability are negatively affected by replacing hay with *G. ulmifolia* leaves. It is suggested to carry out more research that contributes to the understanding and analysis of the vegetal species of the tropical dry forest and the role of the secondary metabolites of plants in ruminal kinetics in small ruminants. These findings demonstrate that *G. ulmifolia* leaves can be used safely to replace part of hay in the rations of hair lambs.

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## Authors' contributions

Edwin Sandoval-Lozano: sampling, laboratory analysis, construction of databases, analysis of information and preparation of manuscript. Diana Cediél-Devia: laboratory analysis, preparation of the manuscript. Román Castañeda-Serrano: Design of methodologies, supervision of activities

## Ethical implications

The experiment was conducted based on Universidad del Tolima's bioethical regulations for animal experimentation (Academic Council Agreement Number 0171 dated October 29, 2008) and Committee of Bioethics Minutes 02/2017 and following the National Institutes of Health guide for the care and use of laboratory animals (NIH Publications No. 8023, revised 1978).

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