

# In vitro, in situ, and in vivo evaluation of a sesame paste supplement for grazing calves

González-Jesús, Antonio<sup>1</sup>; Sánchez-Santillán, Paulino<sup>1</sup>; Torres-Salado, Nicolás<sup>1\*</sup>; Herrera-Pérez, Jerónimo<sup>1</sup>; Alaniz-Gutiérrez, Luis<sup>1</sup>; Ayala-Monter, Marco Antonio<sup>1</sup>

- Universidad Autónoma de Guerrero, Facultad de Medicina Veterinaria y Zootecnia No. 2, km 197 Carretera Acapulco-Pinotepa Nacional, Cuajinicuilapa, Guerrero, México, C. P. 41940.
- \* Correspondence: nivigas@yahoo.com.mx

#### ABSTRACT

**Objective**: To carry out an *in vitro*, *in situ*, and *in vivo* evaluation of soybean paste (T1) and sesame paste (T2) supplements for calves under grazing conditions.

**Methodology**: Partial (24, 48, and 72 h) and accumulated biogas and methane production, as well as dry matter degradation (DMD), were measured in the *in vitro* analysis. The digestibility kinetics parameters (a, b, a+b, c, k, ED) were estimated in the in situ test. Feed supplement consumption (SC), daily weight gain (DWG), bacterial and protozoan populations, acetate, propionate, and butyrate were evaluated in the *in vivo* test. A completely randomized experimental design was used.

**Results**: T1 produced a higher amount of partial and accumulated biogas and methane at 24 h, as well as the highest values in the b, a+b, k, and ED ( $p \le 0.05$ ) parameters. Meanwhile the rest of the *in vitro*, *in situ*, and *in vivo* variables did not record any differences between treatments (p > 0.05).

**Study Limitations/Implications**: The production of sesame paste is seasonal; consequently, its harvest limits its year-round availability.

**Conclusions**: Based on the *in vitro*, *in situ*, and *in vivo* evaluations, sesame paste is a feasible option to replace soybean paste as a source of protein, in the supplementation of calves in the tropics.

**Keywords**: grazing, tropics, protein supplementation, alternatives.

# L., & Ayala-Monter, M. S. (2024). *In vitro, in situ*, and *in vivo* evaluation of a sesame paste supplement for grazing calves. *Agro Productividad*. https://doi.

Citation: González-Jesús, A., Sánchez-

Santillán, P., Torres-Salado, N., Herrera-Pérez, J., Alaniz-Gutiérrez,

Academic Editors: Jorge Cadena Iñiguez and Lucero del Mar Ruiz Posadas

org/10.32854/agrop.v17i5.2906

**Guest Editor**: Daniel Alejandro Cadena Zamudio

Received: October 28, 2023. Accepted: March 16, 2024. Published on-line: May XX, 2024.

*Agro Productividad*, 17(5). May. 2024. pp: 87-97.

This work is licensed under a Creative Commons Attribution-Non-Commercial 4.0 International license.

#### INTRODUCTION

Cattle raising is the main agricultural activity worldwide (Gutiérrez et al., 2018). In the tropical regions of Mexico, cattle raising is practiced in extensive systems, in which vast extensions of land are used to grow native and introduced grasses (REDGATRO, 2018). This system accounts for 78.5% of the domestic production (García et al., 2018). Given their high cell wall levels, low digestibility, reduced protein content, and high nitrogen compound degradability, the quality of tropical grasses is associated with animal productivity (Rojo et al., 2000). This situation delays the growth of calves, reducing productivity and fertility (Rodríguez, 2011). Since feeding accounts for 70% of their total production costs, production units frequently seek to improve their feeding efficiency. Therefore, finding new feeding alternatives —which produce a biological and economic efficiency improvement— is fundamental (García-Balbuena et al., 2022).



Oleaginous fruits, oilseeds, or their by-products are a feasible alternative for bovine feeding systems, because they provide proteins to the animals (Aguilera et al., 2018). In most of the producing countries, sesame (Sesamum indicum) is grown by small producers. Few of these producers have technified systems or are aware of the chemical components of the industrial sesame by-products (paste), which leads to the underutilization of this species (Mussi et al., 2016). Consequently, the producers of these regions are limited to subsistence animal production systems (Velázquez et al., 2005). Aguilera (2014) compared the inclusion of sesame paste with soy paste in the diets of pigs, recording a higher ileal digestibility of crude protein (CP) in pigs that were fed with the former diet. Although Cuca and Ávila (1978) evaluated the inclusion of sesame paste in feeding portions for broilers, information about the use of that paste in the diet of ruminants is still scarce. García-Balbuena et al. (2022) evaluated the effect of the addition of sesame paste to the integral diets of Simbrah calves; however, they did not record significant differences between treatments. The hypothesis of this research was that replacing soy paste with sesame paste, as source of protein, did not modify the characteristics of *in vitro* and *in situ* fermentation and the productive response of F1 calves (under grazing conditions and fed a protein supplement). In conclusion, the objective of this study was to carry out an in vitro, in situ, and in vivo evaluation of sesame and soy paste supplements, in F1 calves under grazing conditions.

#### MATERIALS AND METHODS

#### Location

The *in vitro* and *in situ* evaluations were carried out in the facilities of the Facultad de Medicina Veterinaria y Zootecnia No. 2, Universidad Autónoma de Guerrero, located in km 197 of the Acapulco-Pinotepa Nacional highway. The *in vivo* evaluation was carried out in El 29 ranch, located in the community of Cuajinicuilapa, Guerrero. The experiment was conducted from February to April, 2022. Cuajinicuilapa is located at 16° 58' N and –98° 45' W, at 30 m.a.s.l. The climate is sub-humid warm, with summer rains, an annual precipitation of 1,200 mm, and an annual average temperature of 25 °C (INAFED, 2022).

#### **Treatments**

The treatments were based on the soy paste (T1) and sesame paste (T2) content used to prepare feed supplements (Table 1).

#### In vitro evaluation

The culture medium was prepared according to the method proposed by Cobos and Yokoyama (1995) and modified by Cañaveral-Martínez *et al.* (2020). The culture medium was made up of 50.9% distilled water, 30% clarified ruminal fluid, 5% mineral solution I, 5% mineral solution II, 0.1% resazurin (Sigma-Aldrich®), 4% reducing agent, and 5% buffer solution. The clarified ruminal fluid contained fresh ruminal bovine liquid, centrifuged for 10 m at 12,857 × g and was then sterilized at 121 °C, for 15 m, 15 psi (All American® 1941X, USA); the whole process was carried out twice. Mineral solution I contained 6 g of  $K_2HPO_4$  (J. T. Baker®) in 1,000 mL of distilled water. Mineral solution II was made up of 6 g  $KH_2PO_4$  (J. T. Baker®) + 6 g  $(NH_4)_2SO_4$  (J. T. Baker®) + 12 g NaCl (Meyer®)

or protein for grazing curves.			
Ingredient (%)	T1	Т2	
Soybean paste	35	-	
Sesame paste	-	30	
Molasses	10	10	
Salt	20	20	
Mineral mixture	10	10	
Urea	5	5	
Ground maize kernel	20	25	
Chemical composition (%)			
Crude protein	30.63	30.11	
Dry matter	96.37	96.27	
Ash	33.40	34.71	
Neutral detergent fiber	26.01	32.46	
Acid detergent fiber	6.48	10.02	

**Table 1**. Feed supplement composition with different sources of protein for grazing calves.

+ 2.45 g MgSO<sub>4</sub> (Meyer<sup>®</sup>) + 1.6 g CaCl-2H<sub>2</sub>O (Meyer<sup>®</sup>) in 1,000 mL of distilled water. The reducing agent contained 3.125 g L-cysteine (Sigma-Aldrich<sup>®</sup>), its pH was adjusted to 10 using NaOH (Meyer<sup>®</sup>) + 3.125 g Na<sub>2</sub>S-9H<sub>2</sub>O (Meyer<sup>®</sup>), it was gauged with 250 mL of distilled water, and 0.1 mL of resazurin were added, with a low CO<sub>2</sub> flow and heat, in order to obtain anaerobiosis. Finally, the buffer solution consisted of 80 g of Na<sub>2</sub>CO<sub>3</sub> (J. T. Baker<sup>®</sup>) in 1,000 mL of distilled water.

The biodigesters were placed in 120 mL serological pipettes, to which 0.5 g samples of T1 or T2 (particle size: 1 mm), 40 mL of culture medium, and 10 mL of fresh ruminal fluid (inoculum), under a continuous  $\rm CO_2$  flux, were added. There were 10 independent repetitions per treatment. A neoprene cap and an aluminum ring with a removable center were used to seal the pipettes. The biodigesters were incubated for 72 h, at 39 °C, in a bath Marie. In order to precipitate food and protozoan particles, the fresh ruminal fluid was centrifuged at 1,137  $\times$  g for 3 m. The ruminal fluid was obtained from a bovine with a ruminal fistula, previously grazed with Pangola grass. The bovine was handled according to the bioethics and welfare internal regulations of the UAGro and based on the NOM-062- ZOO-1999 Mexican official standard. The biodigesters were incubated for 72 h at 39 °C (Cañaveral-Martínez *et al.*, 2020).

Biogas production was measured displacing the plunger of a 50 mL glass syringe (BD Yale<sup>®</sup>, Brazil). This process was measured at 2, 4, 6, 8, 10, 12, 24, 48, and 72 h of incubation (Hernández-Morales *et al.*, 2018). Partial biogas production was reported at 24, 48, and 72 h; accumulated biogas production was recorded at 72 h.

Methane (CH<sub>4</sub>) production was measured using a Taygon<sup>®</sup> hose (internal- $\emptyset$ : 2.38 mm; length: 45 cm), with 20 G × 32 mm hypodermic needles at each end. The needles were used to couple the biodigesters with measurement vials. These vials were filled with 80 g of NaOH (2N) solution (Merck<sup>®</sup>) in 1,000 mL of distilled water. The procedure was carried

out following the method proposed by Stolaroff *et al.* (2008), as described by Torres-Salado *et al.* (2018). Partial methane production was considered as the mL of the NaOH (2N) solution displaced at 24, 48, and 72 h of incubation; meanwhile, the accumulated production was recorded at 72 h. Dry matter degradation (DMD) was estimated after 72 h, following the methodology proposed by Hernández-Morales *et al.* (2018).

#### In situ evaluation

Two cows with ruminal fistula under stabling conditions were used for the *in situ* test. As part of the adaptation of the microbiota to the rumen, the DM consumption of each cow was estimated at 3% LW, divided as follows: 70% forage and 30% commercial feeding. Cows had *ad libitum* access to water. In order to guarantee that the sample stayed in the ventral part of the rumen, a 1.5 cm wide × 1 m long galvanized iron chain, modified with a security hook at one end, was attached to the cap of the ruminal fistula (Cañaveral-Martínez *et al.*, 2020).

Five-gram samples from each treatment (T1 and T2) were placed in  $10 \times 20$  cm poly-silk bags, with a  $40~\mu m$  average pore, and were kept at constant weight. The bags were sealed with 100~mm long  $\times$  2.5 mm wide plastic belts. The bags were incubated in triplicate for 0, 2, 4, 8, 12, 20, 32, 48, and 64 h, in the rumen of each cow. All the bags were inserted in the rumen in the reverse order of their incubation time (64, 48, 32, 20, 12, 8, 4, 2, and 0 h); therefore, all the samples could be removed simultaneously, after the incubation period. Once the bags were extracted from the rumen, they were immediately washed with cold running water, until the outcoming water was clear. The 0 h bags were not incubated in the rumen: they were just washed, using the same protocol followed for the bags incubated in the rumen.

The bags with residues were dried at 55 °C for 72 h; subsequently, they were weighted to determine DM disappearance. The *in situ* DM disappearance kinetic was estimated with a non-linear regression procedure in SAS (2011), using the equation described by McDonald (1981):

$$P = a + b \left[ 1 - e - c(t - l) \right]$$

Where: P=ruminal digestibility in time (t; %); a=rapidly soluble digestible fraction (%); b=low or potentially digestible fraction (%); a+b=maximum potential digestibility; c= speed at which b is digested  $(\% \text{ h}^{-1})$ ; t=incubation time (h) in the rumen; and l=delay time (h).

DM effective digestibility (ED) was estimated using the equation described by Ørskov and McDonald (1979):

$$ED = a + \lceil b * c / (c + k) \rceil e - (c + k) L$$

Where: a, b, c, and L are used as described in the previous paragraph and k=rumen exit rate (%  $h^{-1}$ ).

#### In vivo evaluation

The experiment lasted 56 days. Ten F1 calves (Bos taurus × Bos indicus) of  $200 \pm 20 \,\mathrm{kg}$  LW were used in the evaluation (five calves per treatment). Based on the number of their SINIGA earring, the calves were assigned to each treatment. A prophylactic treatment consisting of a 5 mL animal<sup>-1</sup> subcutaneous selenium injection was applied to the calves. In addition, they were injected a subcutaneous vaccine with bacterin (Exgon  $10^{\$}$ ; 5 ml animal<sup>-1</sup>). The calves were periodically bathed with tickicides and insecticides (Bovitraz<sup>\&</sup>; 2 mL L<sup>-1</sup>), using a 20 L sprinkler pump (maximum coverage per pump: 5 calves). Afterwards, a cosproparasitoscopic analysis was carried out. The calves were no dewormed, considering the negative result of this analysis.

The calves grazed together in a rotational arrangement, in three paddocks with cross 1 Bermuda grass predominance (Table 2). Before the experiment started, the calves were subjected to the following supplement adaptation procedure for 10 d: the calves were placed into 2 m² individual stables, where they were initially provided 100 g of the supplement at 08:00 am and, subsequently, the supplement amount increased 100 g at a time, until it reached 500 g d⁻¹. Starting from the first day of the experiment, the calves spent 2 h in the stables, where they fed on 500 g of one of the treatments. The dry matter consumption (DMC) of each treatment was measured as the amount of the feeding offered to the calf minus the amount rejected per day by the calf. The calves had ad libitum access to water. In order to determine the daily weight gain (DWG), they were weighted at the beginning and at the end of the experiment, using a digital livestock scale.

Twenty mL of ruminal fluid were extracted at day 56, with an esophageal catheter, 2 h after the supplement consumption. A doble-gauze layer was used to filter the fluid. Immediately after, a SA 210 portable potentiometer (ORION®, USA), with a 7-4 pH calibration, was used to measure pH. A micro-pipette (Corning®, USA) was used to extract a 1 mL sample of the ruminal fluid, in order to carry out the total bacteria counting (TBC). Afterwards, the sample was poured into a test tube (Pirex®, México), with 0.25 ml of 10% formaldehyde (Sigma-Aldrich®). The total bacteria were counted with a Petroff Hausser bacteria counting chamber. A BX31 microscope (Olympus®, USA) with a 1,000 X magnification, was used for the recounting. The number of bacteria was calculated using the following formula:

number of bacteria = 
$$(average)(dilution factor, 2 \times 10^7)$$

Variable (%)	Paddock 1		Paddock 2		Paddock 3	
	Home	End	Home	Final	Home	End
Crude protein	7.07	6.2	9.23	5.19	9.94	5.47
Dry matter	94.9	95.8	95.15	95.46	94.77	95.74
Ash	8.87	8.01	8.85	8.24	8.8	7.89
Neutral detergent fiber	72.42	71.6	70.74	71.29	72.91	74.23
Acid detergent fiber	36.62	36.5	36.87	36.81	39.68	38.83

The same technique used for the total bacteria counting was used for the protozoa counting (PC); however, the recounting was carried out in a Brand<sup>®</sup> bright-line Neubauer chamber with a 400 X magnification. The number of protozoa was calculated using the following formula:

protozoa counting = 
$$(average)(dilution factor, 2 \times 10^4)$$

(Espinoza-Sánchez et al., 2020).

In addition, 1 mL of ruminal fluid was poured into a 2 mL tube, which was then placed inside a Neptune microcentrifuge (Mexico). The content of the tube was mixed with metaphosphoric acid at 25% (4:1 ratio), before the tubes were centrifuged at 18,800  $\times$  g for 10 minutes. The supernatant was placed in 1.5 mL vials (Perkin Elmer<sup>®</sup>, USA) to carry out a chromatography. A Claurus 580 gas chromatograph (Perkin Elmer<sup>®</sup>, USA), equipped with a flame ionization detector and a 30 m  $\times$  0.25 mm capillary column (Elite FFAP, Agilent<sup>®</sup>), was used to determine the VFA concentration. Helium (with a constant pressure of 10 psi) was used as gas carrier for this procedure, along with H<sub>2</sub> and air to generate a flame with a 40- and 400-mL min<sup>-1</sup> flux, respectively. The oven, the injector, and the column temperatures were 80, 240, and 250 °C, respectively; 1  $\mu$ L of the sample were injected. Acetic acid, propionic acid, and butyric acid recorded retention times of 1.22, 1.54, and 2.01, respectively.

## Bromatological analysis

The 967.03, 920.105, and 942.05 methods of the AOAC methodology (2007) were used to determine dry matter (DM), crude protein (CP), and ash (A), respectively. Following the recommendations of Van Soest *et al.* (1991), the ANKOM Technology® methodology was used to determine the neutral detergent fiber (NDF) and acid detergent fiber (ADF) content. Organic matter (OM) was determined subtracting the ash percentage from 100.

#### Statistical analysis

The *in vitro*, DMC, and DWG variables and the ruminal fermentation characteristics were analyzed using a completely randomized design, with the GLM procedure of SAS (2011). The MIXED model of SAS (2011) was used for the *in situ* evaluation.

#### **RESULTS AND DISCUSSION**

The gas production technique is an indirect measurement of the ruminal microbial fermentation and is based on the ratio between the microbial digestion of food and the production of volatile fatty acids and gases ( $CO_2$  and  $CH_4$ ), as the final metabolites of the fermentation (Amanzougarene and Fondevila, 2020). Consequently, no differences were recorded (p>0.05) between treatments, regarding biogas and methane production at 24-48 h and at 48-72 h (Table 3). T1 biogas production obtained higher values from 0 to 24 h (41.82%) and from 0 to 72 h (34.95%) than T2 (p $\leq$ 0.05). Likewise, T1 produced more  $CH_4$  than T2, at 0-24 h (34.32%) and at 0-72 h (22.65%). These results indicate that

the soy paste supplement (T1) had more non-structural carbohydrates (Table 1) —which ferment during the first 24 h (Texta et al., 2019). The biogas accumulated production is a consequence of fermentation. Meanwhile, no differences were recorded in the biogas production after 24 h, which indicates that the fermentation of the structural carbohydrates was similar between treatments (Texta et al., 2019). Additionally, methanogenic archaea use CO<sub>2</sub> and H<sub>2</sub> in their metabolic route, producing CH<sub>4</sub> as a fermentation product. The stoichiometric reactions indicate a higher CO<sub>2</sub> and H<sub>2</sub> production when the final products are acetate and butyrate. Table 3 shows that, if these ratios are analyzed (Blümmel and Ørskov, 1993)two varieties of spring barley and one variety of winter wheat either untreated or treated with anhydrous ammonia, were examined. In vitro gas production was compared with in vivo results and with nylon bag degradabilities; the sources of the gas were determined. Total gas production a+b as described by the exponential equation were correlated with intake (0.88, the higher CH<sub>4</sub> production could be associated with a higher acetate and butyrate production, as a result of the use of the soy paste supplement (T1).

The *in vitro* technique is used to predict digestibility when the animals consume DM (Gosselink *et al.*, 2004). Consequently, the digestibility of the evaluated treatments should record similar results, because DMD did not show differences (p>0.05) between treatments (Table 3). In this study, the biogas production values for T2 were lower than those reported by García-Balbuena *et al.* (2022), who carried out a research about a 10% sesame paste diet and recorded 117.02, 37.14, 18.10, and 172.32 mL g<sup>-1</sup> DM, at 0-24 h, 24-48 h, 48-72 h, and 0-72 h, respectively. Methane production and DMD recorded higher values in T2 than those reported by García-Balbuena *et al.* (2022) who obtained 30.39, 8.14, 4.86, 43.39, and 26.22 mL g<sup>-1</sup> DM, at 0-24 h, 24-48 h, 48-72 h, 0-72 h, and DMD, respectively.

**Table 3**. *In vitro* fermentation characteristics of supplements made from soybean paste or sesame paste for grazing calves.

1 9 9 9			
Variable	T1	Т2	SEM
Production of biogas in vitro (			
0-24 h	141.4 <sup>a</sup>	99.7 <sup>b</sup>	7.7
24-48 h	19.7	17.6	1.1
48-72 h	13.8	12.2	1.1
0-72 h	174.9 <sup>a</sup>	129.6 <sup>b</sup>	6.7
Production of methane (mL g <sup>-1</sup> DM)			
0-24 h	54.0 <sup>a</sup>	40.2 <sup>b</sup>	1.9
24-48 h	11.5	10.3	0.6
48-72 h	6.5	8.2	0.5
0-72 h	72.0 <sup>a</sup>	58.7 <sup>b</sup>	1.7
Dry matter degradation (%)	87.7	83.0	2.1

<sup>&</sup>lt;sup>a,b</sup>: Means in a row with different letter are different ( $p \le 0.05$ ).

T1=protein source with soybean paste; T2=protein source with sesame paste; SEM=Standard error of the mean.

The *in situ* digestibility technique is used to measure digestion and apparent digestibility at a ruminal level (Torres *et al.*, 2009). *In situ* DM digestibility at 2, 4, and 48 h did not record differences between treatments (p>0.05). T1 recorded a higher *in situ* digestibility at 8, 12, 20, 32, and 64 h than T2 (p $\leq$ 0.05) (Table 4). Regarding kinetics, both treatments showed the same content of digestible soluble fraction (a) (p>0.05). However, T1 had a higher potentially digestible fraction (b) (p $\leq$ 0.05), although its fermentation rate was similar to T2 (c) (p>0.05). Consequently, T1 reported higher potentially digestibility rate (a+b), rumen exit rate (k), and effective digestibility (ED) than T2 (p $\leq$ 0.05) (Table 4). These digestibility differences between supplements can be the result of the sesame paste content in T2, which has 5.4% phytic acid, whose chelating ability reduces dry matter digestibility (Cesária *et al.*, 2022). Aguilera *et al.* (2018) reported lower digestibility values at 64 h. These authors recorded a 60.4% value in a sheep diet prepared with 60% sesame paste.

Initial and final weight, DWG, and SC did not record differences (p>0.05) between treatments (Table 5). García-Balbuena *et al.* (2022) reported higher results than T2. They recorded a 238.75 kg initial weight and a 268.75 kg final weight, as well as a 0.77 kg d<sup>-1</sup> DWG, in calves fed on a diet prepared with 10% sesame paste, 48% corn ensilage, 12% star grass hay, 28% grounded corn grain, and 2% mineral mix. In this study, the average supplement consumption was 324.93 g d<sup>-1</sup>—this figure is 55% lower than the findings of Rojo *et al.* (2000), who reported a consumption of 730 g d<sup>-1</sup> in young bulls fed on a feed supplement prepared with 84% corn, 12% molasses, 4% urea, 0.05% mineral salt, and 10 g of *Saccharomyces cerevisiae* animal<sup>-1</sup> day<sup>-1</sup>.

The ruminal variables —such as pH and total bacteria and protozoa counting— did not record differences between treatments (p>0.05); in average, the following results were recorded: pH, 6.74; total bacteria,  $1.82 \times 10^9$  bacteria mL<sup>-1</sup>; and total protozoa, 2.41×10<sup>5</sup> protozoa mL<sup>-1</sup> (Table 5). Consequently, according to Cardona-Iglesias et al. (2016), these variables fall within the optimal intervals for the induction of a fast growth of cellulolytic bacteria (Borroto, 2015): 6.2-7.0 pH and 1010 cells mL<sup>-1</sup> ruminal bacteria, and 10<sup>5</sup> cell mL<sup>-1</sup> protozoa concentrations. García-Balbuena et al. (2022) reported a higher pH (7.47), bacteria (4.67×10<sup>9</sup> cell mL<sup>-1</sup>) and protozoa (11.02×10<sup>5</sup> cell mL<sup>-1</sup>) than this study. In addition, Carbajal-Márquez et al. (2021) reported similar pH (6.6), bacteria (4.27×10<sup>9</sup> cell mL<sup>-1</sup>), and protozoa (2.5×10<sup>5</sup> cell mL<sup>-1</sup>) values for Suiz-Bu calves that fed on Mulato II grass and were supplemented (1% of LW) with increasing levels of Earpod tree (Enterolobium cyclocarpum) pods. The concentration of acetate, propionate, and butyrate in the rumen depends on the diet composition, the microbial activity, the pH, the frequency with which the diet is consumed, the texture of the ingredients, and the prevailing substrate of the portion (Carbajal-Márquez et al., 2021). Consequently, the evaluated treatments did not have an impact on the production of these volatile fatty acids (p≤0.05) (Table 5). For their part, Carbajal-Márquez et al. (2021) reported higher acetate values (24.63 Mmol L<sup>-1</sup>) and lower propionate and butyrate values (9.00 Mmol L<sup>-1</sup> and 5.60 Mmol L<sup>-1</sup>. respectively).

**Table 4**. *In situ* dry matter digestibility of supplements made from soybean paste or sesame paste for grazing calves.

Variable	T1	T2		
Dry matter digestibility (%	)			
0 h	57.75	59.21		
2 h	65.98	63.54		
4 h	73.22	65.25		
8 h	79.20 <sup>a</sup>	75.44 <sup>b</sup>		
12 h	86.05 <sup>a</sup>	82.07 <sup>b</sup>		
20 h	91.94ª	87.00 <sup>b</sup>		
32 h	91.01 <sup>a</sup>	87.26 <sup>b</sup>		
48 h	90.43	84.93		
64 h	91.74 <sup>a</sup>	85.29 <sup>b</sup>		
Kinetics of digestibility				
a	57.69	58.25		
<i>b</i>	34.10 <sup>a</sup>	29.33 <sup>b</sup>		
a+b	91.8ª	87.79 <sup>b</sup>		
С	0.12	0.11		
k	0.11 <sup>a</sup>	0.14 <sup>b</sup>		
ED	75.86 <sup>a</sup>	71.47 <sup>b</sup>		

<sup>&</sup>lt;sup>a,b</sup>: Means in a row with different letter are different ( $p \le 0.05$ ).

**Table 5.** Body weight, dry matter intake and rumen fermentation characteristics of grazing calves supplemented with soybean meal paste (T1) or sesame paste (T2).

Variable	T1	T2	SEM
Initial weight (kg)	220.04	221.80	5.67
Final weight (kg)	257.52	254.24	6.57
Daily weight gain $(kg d^{-1})$	0.67	0.58	0.03
Supplement consumption $(g d^{-1})$	326.47	323.40	5.56
рН	6.66	6.82	0.11
Total bacteria $(10^9 \text{ Cells mL}^{-1})$	2.07	1.58	0.76
Total protozoa ( $10^5 \text{ Cells mL}^{-1}$ )	2.63	2.19	0.82
Acetate (Mmol $L^{-1}$ )	28.31	23.63	2.36
Propionate (Mmol L <sup>-1</sup> )	9.69	7.24	1.01
Butirate (Mmol L <sup>-1</sup> )	6.45	4.27	0.71

## **CONCLUSIONS**

Based on the *in vitro* and *in situ* evaluations, as well as on the productive behavior of the calves, sesame paste is a feasible option to replace soy paste as a source of proteins, in feed supplements for grazing calves, in the tropical region of Mexico.

T1=protein source with soybean paste; T2=protein source with sesame paste; SEM=Standard error of the mean; a=fast digestible soluble fraction; b=slow or potentially digestible fraction; a+b=maximum potential digestibility; c=rate at which b is digested; k=ruminal output rate; ED=effective digestibility.

#### **ACKNOWLEDGEMENTS**

The authors would like to thank the academic body of UAGro-CA-183 "Producción Sustentable de Rumiantes en el Trópico" for funding this research.

#### REFERENCES

- Aguilera BA. 2014. Pasta de ajonjolí (*Sesamum indicum*) un alimento proteico para cerdos y lechones recién destetados con base a sus características químicas y a la respuesta fisiológica del aparato digestivo. Universidad Autónoma de Querétaro.
- Aguilera CGC, Valiente VOL, Stanley SWE, Corrales MMP, Branda de PLN, Peralta JR, Castellani RPG. 2018. Nutritional Valuation of the sesame cake by *in vivo* and *in vitro* digestibility studies in sheep. *Investigación Agraria* 20, 118–126. https://doi.org/10.18004/investig.agrar.2018.diciembre.118-126
- Amanzougarene Z, Fondevila M. 2020. Fitting of the *in vitro* gas production technique to the study of high concentrate diets. *Animals* 10, 1935. https://doi.org/10.3390/ani10101935
- AOAC. 2007. OMA [WWW Document]. URL https://www.eoma.aoac.org/ (accessed 5.18.22).
- Blümmel M, Ørskov ER. 1993. Comparison of in vitro gas production and nylon bag degradability of roughages in predicting feed intake in cattle. Animal Feed Science and Technology 40, 109-119. https://doi. org/10.1016/0377-8401(93)90150-I
- Borroto GO. 2015. La fisiología digestiva del rumiante, objeto de investigación en el Instituto de Ciencia Animal durante cincuenta años. *Revista Cubana de Ciencia Agrícola* 49, 179-188.
- Cañaveral-Martínez UR, Sánchez-Santillán P, Torres-Salado N, Sánchez-Hernández D, Herrera-Pérez J, Rojas-García AR. 2020. Características de calidad, bromatológicas y fermentativas *in vitro* de ensilado de mango maduro. *Revista Mexicana de Agroecosistemas* 8, 82-90.
- Carbajal-Márquez U, Sánchez-Santillán P, Rojas-García AR, Ayala-Monter MA, Mendoza-Núñez MA, Hernández-Valenzuela D. 2021. Effect of parota (Enterolobium cyclocarpum) pod protein supplement on feed intake and digestibility and calf ruminal characteristics. Tropical Animal Health Production 53, 323. https://doi.org/10.1007/s11250-021-02772-6
- Cardona-Iglesias JL, Mahecha-Ledesma L, Angulo-Arizala J. 2016. Arbustivas forrajeras y ácidos grasos: estrategias para disminuir la producción de metano entérico en bovinos. *Agronomia Mesoamericana*. 28, 273. https://doi.org/10.15517/am.v28i1.21466
- Cesária RST, Aguilera BA, Mariscal LG. 2022. Digestibilidad ileal estandarizada de la proteína y aminoácidos de la pasta de ajonjolí en cerdos en crecimiento. *Revista Mexicana Ciencias Pecuarias*. 12, 1292–1304. https://doi.org/10.22319/rmcp.v12i4.5714
- Cobos MA, Yokoyama MT. 1995. *Clostridium paratrifcum* var. Ruminanitum: colonisation and degradation of shrimp carapaces *in vitro* observed by scanning electron microscopy, in: Wallace, R.J., Lahlou-Kassi (Eds.), Rumen Ecology Research Planning. Proceedings of a Workshop held at the International Livestock Research Institute, Addis Ababa, Ethiopia, pp. 151–161.
- Cuca MG, Ávila EG. 1978. Fuentes de energía y proteínas para la alimentación de las aves. *Ciencia Veterinaria* 2, 34.
- Espinoza-Sánchez J, Sánchez-Santillán P, Torres-Salado N, Ayala-Monter MA, Herrera-Pérez J, Magadan-Olmedo F. 2020. Inclusion of ripe mango as a source of energy in diets for Creole lambs in the dry tropics. *Tropical Animal Health Production* 52, 3519-3526. https://doi.org/10.1007/s11250-020-02386-4
- García MA, Albarrán PB, Rebollar RS. 2018. La ganadería en condiciones de trópico seco: el caso del sur del Estado de México, condiciones actuales y perspectivas de desarrollo, 1a edición. ed, Investigación universitaria. Universidad Autónoma del Estado de México, Toluca, Estado de Mexico.
- García-Balbuena A, Torres-Salado N, Herrera-Pérez J, Maldonado-Peralta MD los Ángeles, Mayren-Mendoza FDJ, Mendoza-Medel G. 2022. Producción de gas in vitro y respuesta productiva de becerras alimentadas con una dieta integral que contiene pasta de ajonjolí (Sesamun indicum) como fuente proteica. Tropical and Subtropical Agroecosystems 25. https://doi.org/10.56369/tsaes.4155
- Gosselink JMJ, Dulphy JP, Poncet C, Jailler M, Tamminga S, Cone JW. 2004. Prediction of forage digestibility in ruminants using in situ and in vitro techniques. Animal Feed Science and Technology 115, 227-246. https://doi.org/10.1016/j.anifeedsci.2004.01.008
- Gutiérrez FA, Estrella A, Irazábal E, Quimiz V, Portilla A, Bonifaz N. 2018. Mejoramiento de la eficiencia de la proteína de los pastos en bovinos de leche utilizando cuatro formulaciones de balanceados. LA GRANJA: *Revista de Ciencias de la Vida. 28*(2): 116-123. https://doi.org/10.17163/lgr.n28.2018.09
- Hernández-Morales J, Sánchez-Santillán P, Torres-Salado N, Herrera-Pérez J, Rojas-García AR, Reyes-Vázquez I, Mendoza-Núñez MA. 2018. Composición química y degradaciones *in vitro* de vainas y

- hojas de leguminosas arbóreas del trópico seco de México. Revista Mexicana de Ciencias Pecuarias 9, 105-120. https://doi.org/10.22319/rmcp.v9i1.4332
- INAFED. Instituto Nacional para el Federalismo y el Desarrollo Municipal. 2022. Guerrero Cuajinicuilapa [WWW Document]. URL http://www.inafed.gob.mx/work/enciclopedia/EMM12guerrero/municipios/12023a.html (accessed 5.18.22).
- Mussi C, Nakayama H, Cristaldo ROD. 2016. Phenotypic variability in M 1 populations of sesame (Sesamum indicum L.) with gamma ray irradiated. Cultivos Tropicales. 37: 74-80. https://doi.org/10.13140/RG.2.1.3749.3362
- Ørskov ER, McDonald I. 1979. The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. *The Journal of Agricultural Science*. 92, 499–503. https://doi.org/10.1017/S0021859600063048
- REDGATRO. Red de investigación e innovación tecnológica para la ganadería bovina tropical. 2018. Estado del arte sobre investigación e innovación tecnológica en ganadería bovina tropical, 2°. ed.
- Rodríguez I. 2011. Estrategias de alimentación para bovinos en el trópico. *Mundo pecuario. 7*(3): 167-170. http://www.saber.ula.ve/bitstream/handle/123456789/33777/articulo6.pdf?sequence=1&isAllowed=y
- Rojo RR, Mendoza MD, García BM, Bárcena GJR, Aranda IEM. 2000. Consumo y digestibilidad de pastos tropicales en toretes con suplementación nitrogenada y *Saccharomyces cerevisiae*. *Revista de la facultad de agronomía (LUZ)* 17: 358–370. file:///C:/Users/mvzje/Downloads/Rojo2000Consumodepastostropical es.pdf
- Texta JN, Sánchez-Santillán P, Sánchez DH, Salado NT, Galvan MC, Pérez JH, Rojas-García RA. 2019. Use of disaccharides and activated carbon to preserve cellulolytic ruminal bacterial consortiums lyophilized. *Revista MVZ Córdoba* 24, 7305-7313. https://doi.org/10.21897/rmvz.1412
- Torres GG, Arbaiza TF, Carcelén CF, Lucas OA. 2009. Comparación de las técnicas in situ, in vitro y enzimática (celulasa) para estimar la digestibilidad de forrajes en ovinos. Revista de Investigaciones Veterinarias del Perú 20, 5-9. https://doi.org/10.15381/rivep.v20i1.348
- Torres-Salado N, Sánchez-Santillán P, Rojas-García AR, Herrera-Pérez J, Hernández-Morales J. 2018. Producción de gases efecto invernadero *in vitro* de leguminosas arbóreas del trópico seco mexicano. *Archivos de Zootecnia* 67, 55–59. https://doi.org/10.21071/az.v67i257.3491
- Van Soest PJ, Robertson JB, Lewis BA. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* 74, 3583-3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2
- Velázquez AJ, Garza RP, Zebadúa MEV, Martínez LZ. 2005. Evaluación de vainas de quebracho (*Acacia farnesiana*) en alimentación de ganado lanar. *Archivos de Zootecnia* 54, 535-540.

