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The effect of avocado flour, sunflower oil and different forage : concentrate ratios in the final diet on feed intake, digestibility and productive performance of male sheep

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ABSTRACT

The objective of this study was to evaluate the inclusion of avocado flour and sunflower oil as energy sources in different proportions, as well as different forage:concentrate ratios on the productive performance and feed intake of male rams, and the *in vitro* dry matter digestibility, organic matter digestibility and gas production. The intake was measured in diets containing avocado flour or sunflower oil with 60% forage: 40% concentrate, and in diets with different forage:concentrate ratios and 10% inclusion of avocado flour or sunflower oil. The daily weight gain was registered for 84 days, the *in vitro* dry matter digestibility was determined at 48 h, and the *in vitro* gas production was evaluated at 72 h. The partial production of gas at 24 hours, raw protein, raw fat, and ash were used to estimate the metabolizable energy and organic matter digestibility. Low levels of avocado flour and sunflower oil in the diet showed the highest intake, *in vitro* dry matter digestibility and *in vitro* gas production. Daily weight gains were 0.28 kg in avocado flour and 0.30 kg in sunflower oil diets, both at 10% inclusion and 40% forage: 60% concentrate. The inclusion of high levels of avocado flour or forage in the diet decreased the feed intake of male sheep. The best avocado flour level was 10% with 40% forage: 60% concentrate.

Key words: acceptability; digestibility; gas production; avocado; rams

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Introduction

Sheep production in Mexico is an important activity with great potential for development due to the unsatisfied demand for sheep meat and the rising meat prices (ARTEAGA, 2012). Many factors limit sheep production, preventing it from becoming a good source of income for developing countries. One of the main problems in sheep production is the seasonality in the production of tropical pastures, animal health problems, poor agricultural practices and the marketing of sheep (WU et al., 2016).

In the tropical regions, the feeding of ruminants is mainly based on forages that animals graze in the areas near to the site where they are kept, and hence, it is necessary to know the nutritional quality of the forages used in different edaphoclimatic conditions. Energy and protein sources are of great importance for lambs since they are required to stimulate microorganisms in the rumen and improve animal production (COMBATT et al., 2015). In this regard, the lack of food sources imposes a challenge for the production of small ruminants, leading to the use of unconventional food sources, which are available and cheaper than conventional food sources (OKORUWA et al., 2015).

Avocado fruit has been widely used as a source of food and for the treatment of diseases. This fruit has several biological properties, such as analgesic, anti-inflammatory and total cholesterol-reducing activity (DINIZ et al., 2017), as well as high levels of carbohydrates, vitamins, and minerals. It is well known that avocado is a nutritious fruit, with high levels of unsaturated fatty acids, vitamins, antioxidants, minerals, and the fat content (10-30%) its main nutritional characteristic. The avocado fats contain mainly oleic and linoleic fatty acids that counteract the harmful effects of saturated fats found in animal fats (HERNÁNDEZ et al., 2016).

Mexico is the world leader in avocado fruit production, contributing 30% of the worldwide production, and the state of Nayarit is one of the main avocado producers (SIAP, 2017). On the other hand, the high production and export requirements lead to the rejection of large quantities of fruit during packing, due to small size or physical damage. However, since the nutritional value of the discarded avocado is not affected, this fruit could used as food for pigs (GRAGEOLA et al., 2010) and small ruminants (OKORUWA et al. 2015).

Although it is not easy to separate the pulp from the rest of the fruit to feed animals, it is possible to mix all the dried and ground fruit with the rest of the concentrate feed ingredients to feed sheep. Information about the amount of avocado paste or flour included in sheep diets could help us to determine its use and impact on the composition and quality of feed rations. Nevertheless, it is not known if the acceptability of the food will vary with that mixture. Therefore, this information is necessary to include avocado flour in the diets of sheep. The objective of this study was to evaluate the effect of different amounts of avocado flour (AF), and compare it with the inclusion of sunflower oil using different forage: concentrate ratios on feed intake, the *in vitro* dry matter digestibility, organic dry matter digestibility, gas production, and the productive performance of rams in the finishing stage.

Materials and methods

Location. The study was carried out in the Laboratory of Nutritional Physiology and Experimental Surgery of the “Unidad Academica de Agricultura” of the “Universidad Autónoma de Nayarit” located at km 9 of the Tepic-Puerto Vallarta highway, in Nayarit, Mexico. *In vitro* digestibility and gas production assays were carried out in the Animal Nutrition Laboratory of the experimental center "El Verdineño" of the “Instituto Nacional de Investigaciones Forestales y Agropecuarias”, located at km 6.5 of the Navarrete-Sauta road, in Nayarit, Mexico.

Preparation of flour of avocado full fruit. Hass avocados discarded for human consumption due to their small size and/or physical damage were used in this experiment. Fruits were collected in packing plants located in Xalisco, Nayarit, Mexico. The producers cut the avocados at physiological maturity and then deliver them to the packing plant. The fruits were stored at room temperature until they reached maturity (when the avocado skin changed from green to black, where the color was determined by a Minolta R-400 colorimeter with

average values and standard deviation of L * = 27.47 ± 2.06, a * = 2.27 ± 2.23 and b * = 4.28 ± 2.74). The firmness of the avocado was determined using a digital penetrometer (GY-4 Stable Micro Systems Model TA.XT2, Texture Technologies Corp., Scarsdale, NT), with force values of 2.41 ± 0.34 kg (HERNÁNDEZ et al., 2016).

To obtain a homogeneous mixture of ripe and whole avocados (pulp, seed, and shell), the fruit was ground in a mobile hammer mill without a sieve, driven by a 5 HP gasoline engine. The fresh paste was stored at room temperature without additives, in plastic containers. Then, the paste was left at room temperature for four days until a dry paste was obtained (flour of avocado full fruit), which was ground again to incorporate it into the lamb diets.

The proximal chemical characteristics and fatty acid profile of whole avocado paste have been previously published (LEMUS et al., 2017).

Evaluation of dietary intake of diets with avocado flour and sunflower oil in sheep. To measure the intake of the experimental diets, three crossbred male sheep were used for each of the four trials. The Pelibuey x Dorper rams had an initial live weight of 40 ± 2 kg, and they were included in the diets for 7 experimental days and 5 days for adaptation. In the first trial, 0, 10, 20 and 30% avocado flour was included in the diets, with 60% forage 40% concentrate (Table 1). In the second trial, 0, 10 and 20% of sunflower oil were used, with 60% forage: 40% concentrate (Table 1).

The third trial considered diets with 10% avocado flour, with different forage: concentrate ratios (60:40, 50:50 and 40:60; Table 2). In the last

Table 1. Diets with different concentrations of avocado flour (AF) and sunflower oil (SO) for the evaluation of acceptability, *in vitro* dry matter digestibility and *in vitro* gas production in sheep

	Avocado flour %			Control	Sunflower oil %	
Ingredients	30	20	10	0	20	10
AF or SO	30.00	20.00	10.00	0.00	20.00	10.00
Alfalfa	30.00	40.00	50.00	60.00	60.00	60.00
Sorghum	24.90	28.66	30.11	31.40	10.41	20.91
Soybean meal	4.10	1.80	0.75	0.10	0.29	0.22
Canola meal	1.90	0.54	0.44	0.10	0.34	0.21
Molasses	7.00	7.00	7.00	7.00	7.00	7.00
Minerals with monensin (0.3%)	1.00	1.00	1.00	1.00	1.00	1.00
Urea	0.70	0.70	0.40	0.10	0.66	0.36
Magnesium oxide (0.3 %)	0.30	0.30	0.30	0.30	0.30	0.30
Calcium carbonate	0.10	0.00	0.00	0.00	0.00	0.00
Nutritional value dry basis						
Digestible energy Mcal/kg	3.60	3.20	2.80	2.40	3.48	2.99
Crude protein	14.00	14.00	14.00	14.09	14.00	14.00
Crude fat	15.65	11.18	6.71	2.24	21.58	11.91
Crude fiber	15.30	16.22	17.30	18.36	17.77	18.07
Calcium	0.80	0.86	0.98	1.09	1.09	1.09
Phosphorus	0.27	0.25	0.25	0.25	0.18	0.22
Cost/kg (MXN$)	4.32	4.51	4.77	5.04	7.09	6.07

MXN$: Mexican pesos

Table 2. Diets with different concentrate: forage ratios and 10% avocado flour (AF) or sunflower oil (SO) for the evaluation of acceptability, *in vitro* dry matter digestibility and *in vitro* gas production

	Avocado flour			Sunflower oil		Control	
Forage : Concentrate	60:40	50:50	40:60	60:40	50:50	40:60	60:40
Ingredients							
AF or SO	10.00	10.00	10.00	10.00	10.00	10.00	0.00
Alfalfa	50.00	40.00	30.00	60.00	50.00	40.00	60.00
Sorghum	30.11	37.35	44.47	20.91	28.09	35.27	31.40
Soybean meal	0.75	2.30	3.56	0.22	1.65	3.05	0.10
Canola meal	0.44	1.65	3.27	0.21	1.60	3.02	0.10
Molasses	7.00	7.00	7.00	7.00	7.00	7.00	7.00
Minerals (Monensin 0.3%)	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Urea	0.40	0.40	0.40	0.36	0.36	0.36	0.10
Magnesium oxide (0.3%)	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Calcium carbonate	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Nutrimental value dry basis							
Digestible energy Mcal	2.80	2.88	2.97	2.99	3.07	3.16	2.40
Crude protein	14.00	14.00	14.00	14.00	14.00	14.00	14.09
Crude fat	6.71	6.81	6.92	11.91	12.00	12.11	2.24
Crude fiber	17.30	14.85	12.43	18.07	15.63	13.20	18.36
Calcium	0.98	0.86	0.75	1.09	0.98	0.87	1.09
Phosphorus	0.25	0.27	0.30	0.22	0.24	0.27	0.25
Cost/Kg (MXN$)	4.77	4.87	4.96	5.04	6.16	6.26	6.07

MXN$: Mexican pesos

trial, the diets included 10% of SO, with different forage: concentrate ratios (60:40, 50:50 and 40:60; Table 2). All diets were isoproteic (14% crude protein) but varied in energy levels.

The animals were kept in 2 m × 2 m individual sheds with a cement floor, and separated from each other for the 7 days of the experimental period. Each shed was equipped with a steel sheet feeder, divided into compartments. The diets were simultaneously placed in the different compartments, and 1.4 kg of DM/animal/day was administered. An intake of 3.5% of the live weight of a sheep was considered. The feeding of the sheep started at 08:00 a.m. and finished at 4:00 p.m. The daily intake was determined as the difference between the weight of the feed offered and the amount rejected at the end of each experimental day.

To carry out the cafeteria test, each diet was randomly distributed to each feeder for the 7 days of evaluation to have the greatest possible number of combinations in the feeder, and to block the habituation reflex of the animal to the first position, distance and feed when confronted with the different diets in the feeder compartment. Animals were handled according to national guidelines for the use and care of animals (NOM-062-ZOO-1999).

Experimental procedures for digestibility. In vitro digestibility of dry matter (IVDMD) and *in vitro* gas production were evaluated using the same diets as previously used to estimate the consumption of avocado flour and sunflower oil (Tables 1 and 2).

In vitro dry matter digestibility (IVDMD) assessment. The IVDMD was determined using three samples per diet at 48 h and expressed as a

percentage of dry matter. The IVDMD was obtained using the TILLEY and TERRY protocol (1963) and modified according to the filter bag method (F-57 ANKOM technology), using a DaisyII incubator (ANKOM, 2010). The ruminal inoculum was obtained from *Bos taurus x Bos indicus* heifers, provided with a permanent ruminal fistula.

In vitro gas production. In vitro gas production was calculated using the technique described by MENKE and STEINGASS (1988) modified by THEODOROU et al. (1994). The procedure uses an automated and wireless gas production system and software (version 9.5), described by ANKOM (2009). Three samples per diet at 24, 48 and 72 h were evaluated and expressed as gas mL g^{-1} dry matter. The metabolizable energy (Mcal kg^{-1} dry matter) and the organic matter digestibility (OMD) were estimated from the partial gas production at 24 h and CP, crude fat (CF) and ash; using the appropriate equations for the type of forage according to MENKE and STEINGASS (1988):

ME (Mcal kg^{-1} DM) = 2.43 + 0.1206 * PG24h + 0.0069 * CP + 0.0187 * CF / 4.184 (12c)

OMD (%) = 16.49 + 0.9042 * PG24h + 0.0492 * CP + 0.0387 ASH (43c)

Where:

ME is the metabolizable energy
PG24h is the gas production at 24 h, mL gas g^{-1} DM.
CP is the crude protein (%)
CF is raw fat
OMD is the organic matter digestibility.
ASH is the percentage of ash.

Evaluation of the productive performance of sheep fed avocado flour and sunflower oil. In the final trial, three diets were compared using 16 Pelibuey-Dorper crossed male sheep per diet. The diets included 10% avocado flour, 10% sunflower oil and a control diet, all with 40% forage: 60% concentrate (Table 2). Live weight, weight gain, feed intake and feed efficiency, carcass yield and meat yield were measured. The trial lasted 84 days, in two blocks.

Fifteen days before the start of the experiment, the male sheep were dewormed and their bodyweight recorded. After this, the animals were housed in individual cages of 3.5 m^2, equipped with a galvanized sheet feeder and an automatic drinking trough. One and a half kg of feed, dry base, was offered every day at 08:00 a.m. per experimental unit.

Experimental design and statistical analysis. Feed intake data of avocado flour or sunflower oil were analyzed using a randomized block design. IVDMD data and *in vitro* gas production were analyzed by time (24, 48 and 72 h) using a one-way analysis of variance. For productive performance data, a randomized block design was used, in which besides diet, the initial weight was included as a covariate. The comparison of means was performed using the Tukey test with a 5% signifance level using the Statistical Analysis System (SAS, 2004).

Results and discussion

Feed intake of avocado flour and sunflower oil and different forage: concentrate ratios. Intake of diets with whole avocado fruit flour and sunflower oil with different forage: concentrate ratios are shown in Tables 3 and 4. Statistical differences ($P<0.05$) were found between treatments. The diet with 10% inclusion of avocado flour or 10% sunflower oil was similar to the control diet; however, the feed intake decreased as the level of avocado flour increased to 20 or 30% and sunflower oil to 20%. The animals preferred diets with 10% avocado flour and with a ratio of 40% forage: 60% concentrate. In addition, we observed that as the forage increased, the feed intake decreased. The regression coefficient for dry matter intake was -0.014 kg ± 0.002 with increasing levels of avocado flour, which means that for every 1% inclusion of avocado flour, the intake decreased by 14 g ($P<0.05$). When the forage was increased by 1% in the diet, the intake decreased, -0.027 kg ± 0.004 ($P<0.05$). The results of this study indicate that the inclusion of avocado flour in the diet had a negative effect on intake, particularly when the inclusion level was greater than 10%.

This result agrees with that reported by PATRA and SAXENA (2010) and YANG et al. (2009), who observed that the addition of vegetable oil in diets for ruminants was associated with a decrease in the digestibility of the plant cell wall due to the negative effect of oils on the protozoa and fibrolytic

bacteria in the stomach. On the other hand, MARTÍNEZ-MARÍN et al. (2012) investigated the use of vegetable oils in sheep diets, and concluded that it is possible to include moderate amounts of vegetable oils rich in unsaturated fatty acids in the diet of small ruminants without causing negative effects on nutrient digestibility. Similar results were obtained in this study (Table 4), showing that high levels of avocado flour reduced feed intake, but adequate intake was observed at low levels of avocado, which was influenced by the forage: concentrate ratio used.

Table 3. Means of the level of avocado flour and forage: concentrate ratio for daily intake of male sheep

Level (%)	Intake[1] (kg)	Forage : Concentrate	Intake[2] (kg)
0	0.587[a]	60:40	0.205[b]
10	0.448[a]	50:50	0.331[b]
20	0.295[b]	40:60	0.745[a]
30	0.173[b]		
SEM	0.011		0.043

Different letters in the columns indicate significant differences (P<0.05). SEM - Standard error of the means.

Table 4. Means of sunflower oil level and forage: concentrate ratio for daily intake in sheep

Level (%)	Intake (kg)	Forage : Concentrate	Intake (kg)
0	0.689[a]	60:40	0.107[c]
10	0.753[a]	50:50	0.379[b]
20	0.158[b]	40:60	0.706[a]
SEM	0.071		0.022

Different letters in the columns indicate a statistically significant difference (P<0.05). SEM - Standard error of the mean.

According to VASTA and LUCIANO (2011), several studies can be found related to the regulation of intake in ruminants and all of them agree that many factors exist that affect it. In this sense, the forage: concentrate ratio is an important factor of intake, considering the size of the animals' organs. GONZÁLEZ et al. (2011) mentioned that the nutritional characteristics of some food resources could depress intake, digestibility, fermentation rate and the supply of microbial nitrogen.

The inclusion of sunflower oil and different forage: concentrate ratios in the diet allowed us to identify male sheep preferences, according to the characteristics of the digestive tract and physiological state. TARAZONA et al. (2012) reported that when only one type of food is available, the animal cannot minimize metabolic stress by changing its intake ratio to achieve nutrient balance. Instead, the animal increases intake in an attempt to meet its nutritional needs, and this leads to problems associated with gastric overload and metabolic disorders. Therefore, the best option is to offer various diets, which will allow the animal to select a feed and thus regulate intake. This was observed in this study by allowing animals to select food from the different diet combinations analyzed, in order to obtain a real estimation.

IVDMD for diets with different levels of avocado flour and sunflower oil. The IVDMD for diets with increasing levels of avocado flour and sunflower oil showed significant differences (P<0.05). The diets with 10, 20 and 30% avocado flour were similar, but with lower IVDMD than the control and sunflower oil diets (Table 5). The IVDMD of the diets with 10 and 20% sunflower oil was statistically similar to the control diet.

Table 5. Means of levels of avocado flour and sunflower oil with 60% forage: 40% concentrate on *in vitro* dry matter digestibility of sheep

Level (%)	Treatment	Mean (%)
0	Control	89.57[a]
10	Avocado flour	82.51[b]
20	Avocado flour	82.82[b]
30	Avocado flour	81.94[b]
10	Sunflower oil	89.69[a]
20	Sunflower oil	88.84[a]
SEM		0.87

Different letters indicate significant differences (P<0.05). SEM - Standard error of the means.

According to the literature, avocado flour has high tannin content (LEMUS et al., 2017), which could explain the preservation of the pulp. It has been shown that high concentrations of tannins influence the digestibility of food (HUANG et

al., 2018; ROSSI et al., 2008; MLAMBOA et al., 2007). These researchers suggest that anti-nutritional factors compromise the degradation of fodder in the stomach of domestic animals, and this is the possible explanation of the results of this study. These results agree with those obtained in this investigation. Besides, the fact that digestibility is lower in the rumen could promote the derivation of proteins from the stomach to the small intestine where it can be used, which would allow greater weight gains, regardless of the low dry matter intake. This is consistent with the review by ORTIZ-LÓPEZ et al. (2016), where they discussed the effect of tannins on meat quality.

GONZÁLEZ et al. (2011) assessed the inclusion of peas (*Pisum sativum l.*) and *in vitro* gas production in diets for growing sheep, using levels of inclusion from 0 to 75%. For digestibility, the best inclusion levels were the lowest, and 75% of peas decreased digestibility, a situation similar to what happened here.

WANAPAT et al. (2013) mentioned that the addition of coconut oil and mangosteen in association or alone, had an effect on the digestibility and production of methane at the ruminal level in buffalo. These same authors mentioned that the use of secondary vegetable compounds, such as condensed tannins, saponins, and diets rich in minerals, as well as garlic and vegetable oils, resulted in a decrease in ruminal protozoa, methanogens, and methane mitigation. Therefore, they affected the digestibility and use of the forage resources that are supplied to ruminants. In this study, sunflower oil and avocado flour were added in different proportions, hence, their effect was manifested by modifying digestibility, mainly at levels of 20% onwards.

The aforementioned results could be due to the inclusion of avocado seed in the preparation of the avocado paste, which caused a low intake when combined with the base food. According to UCHENNA et al. (2017) in a study conducted with avocado seed as part of the diet, the acceptance of the food changed according to the amount of avocado seed added.

IVDMD for diets with 10% avocado flour, 10% sunflower oil and different forage: concentrate ratios. The IVDMD at 48 h for diets with 10% avocado flour or 10% sunflower oil, and with different forage: concentrate ratios were statistically different ($P<0.05$; Table 6). The control diet had the highest digestibility value followed by diets with the lowest forage: concentrate ratio. MARTÍNEZ-MARÍN et al. (2012) indicated that the addition of vegetable oil has different effects on ruminant production, such as a decrease in the organic matter digestibility. Those authors also suggested that the inclusion of oil could have an effect on the daily intake of dry matter. According to some authors, the increase in oil in ruminant diets reduces the availability of fermentable energy, and can generate negative effects on the activity of ruminants (YANG et al., 2009), but this depends on the level of inclusion; low levels (4.5%) of vegetable oil showed no effect on digestibility or feed intake (SANTANA and CORREA, 2016).

Table 6. Means of different forage: concentrate ratios fixed at 10% whole avocado fruit flour and 10% sunflower oil for *in vitro* dry matter digestibility.

Forage: Concentration ratio	Treatments	Mean
60:40	Control	89.92^{a}
60:40	Avocado flour	83.62^{bc}
50:50	Avocado flour	83.33^{c}
40:60	Avocado flour	84.94^{bc}
60:40	Sunflower oil	83.71^{c}
50:50	Sunflower oil	87.18^{ab}
40:60	Sunflower oil	85.09^{bc}
SEM		0.73

Different letters indicate statistically significant differences ($P<0.05$). SEM - Standard error of the mean.

In vitro gas production, metabolizable energy (ME) and organic matter digestibility (OMD) in diets with different levels of avocado flour or sunflower oil. The mean gas production and ME (Table 7) of treatments with avocado flour or sunflower oil differed from the control diet ($P<0.05$). The inclusion of avocado flour or sunflower oil affected the production of gas and ME, which decreased as the level of avocado flour or sunflower oil increased.

Table 7. Mean of treatment and time for gas production (mL gas g^{-1} DM), metabolizable energy and organic matter digestibility

Treatment (%)	pg24	pg48	pg72	ME	OMD
0 Control	157.50[a]	177.90[a]	182.00[a]	5.39[a]	82.87[e]
10 Avocado flour	126.55[b]	150.75[b]	150.75[b]	4.53[bc]	84.98[ab]
20 Avocado flour	126.43[b]	144.93[b]	142.97[b]	4.58[b]	84.09[d]
30 Avocado flour	116.10[c]	132.90[c]	130.07[c]	4.32[c]	84.12[cd]
10 Sunflower oil	119.87[bc]	130.30[c]	128.07[c]	4.39[bc]	84.66[bc]
20 Sunflower oil	101.37[d]	106.50[d]	102.20[d]	3.96[d]	84.38[cd]
SEM	1.56	1.39	1.65	0.05	0.17

Pg24 - Gas production at 24 h; Pg48 - Gas production at 48 h; and Pg72 - Gas production at 72 h; ME - Metabolizable energy; OMD - Organic matter digestibility; SEM - Standard error of the mean. Different letters in the columns indicate statistically significant differences (P<0.05).

In the case of the OMD, low levels of avocado flour and sunflower oil (10%) had the highest digestibility compared to the control treatment that presented the lowest values. In a study of the inclusion of coconut oil and mangosteen, in rations for river buffalo, WANAPAT et al. (2013) mentioned that the use of this type of product has repercussions on the production of methane at the ruminal level. They evaluated the population dynamics in the rumen, and evaluated the amounts of bacteria, fungi, and protozoans present and their modifications, according to the different levels of inclusion. It was noted that high levels of inclusion caused a loss in gas production, a situation similar to what occurred in this study.

WU et al. (2016) investigated the *in vitro* influence of oleic acid on ruminal fermentation and the production of fatty acids, finding that the inclusion of oleic acid caused a decrease in the *in vitro* dry matter digestibility. These results are similar to those obtained here, where the addition of vegetable oils in levels greater than 20% caused a decrease in digestibility.

Significant differences were observed between levels of avocado flour or sunflower oil. The diet with 10% avocado flour and 40% forage: 60% concentrate was similar to the control diet for gas production, ME and OMD (P<0.05; Table 8). Diets with 10% sunflower oil had lower gas and ME production.

In vitro gas production, energy metabolizable (ME) and organic matter digestibility (OMD) of the diets with 10% avocado flour or 10% sunflower oil and different forage: concentrate ratios. Significant differences were observed between the diets with different avocado flour or sunflower oil levels. The diet with 10% avocado flour and 40% forage: 60% concentrate was similar to the control diet for gas production, ME and OMD (P<0.05; Table 8). Diets with 10% sunflower oil had lower gas and ME production.

As mentioned before, and according to MARTÍNEZ-MARÍN et al. (2012), the addition of vegetable oil has different effects on ruminant production, one of them being a decrease in organic matter digestibility. The increase in oil in ruminant diets reduces the availability of fermentable energy, and can generate negative effects on the activity of ruminants (YANG et al., 2009) depending on the level of inclusion (SANTANA and CORREA, 2016).

Productive performance of sheep fed avocado flour and sunflower oil. Table 9 shows the productive performance of lambs fed with 10% avocado flour or 10% sunflower oil, where the highest values for final weight and daily weight gain were greater in the treatments that included 10% avocado flour or 10% sunflower oil compared to the control treatment. The highest daily intake was obtained in the treatment with sunflower oil. In the case of the treatments with 10% avocado flour, significant

Table 8. Means of different forage: concentrate ratio with 10% avocado flour or 10 sunflower oil for gas production (mL gas g-1 DM) at different times and for metabolizable energy and organic matter digestibility

Treatments	F : C	Pg24	Pg48	Pg72	ME	OMD
Control	40 : 60	152.35^{a}	164.85^{a}	167.15^{a}	5.23^{a}	83.39^{bc}
Avocado flour	60 : 40	128.20^{bc}	140.93^{cd}	135.17^{bc}	4.94^{bc}	84.82^{a}
Avocado flour	50 : 50	137.35^{b}	148.60^{bc}	143.25^{bc}	5.17^{ab}	83.91^{ab}
Avocado flour	40 : 60	146.90^{a}	153.53^{ab}	146.00^{b}	5.31^{a}	82.95^{c}
Sunflower oil	60 : 40	126.37^{c}	134.13^{de}	130.00^{cd}	4.80^{cd}	84.05^{ab}
Sunflower oil	50 : 50	130.47^{bc}	136.17^{de}	130.37^{cd}	4.86^{c}	83.54^{bc}
Sunflower oil	40 : 60	124.47^{c}	127.63^{e}	121.33^{e}	4.62^{d}	83.98^{ab}
SEM		1.83	2.30	2.69	0.47	0.18

F:C - Forage - concentrate; Pg24 - Gas production at 24 h; Pg48 - Gas production at 48 h; Pg72 - Gas production at 72 h; ME - Metabolizable energy; OMD - Organic matter digestibility; SEM - Standard error of the means. Different letters in the columns indicate statistically significant differences (P<0.05).

Table 9. Productive performance of sheep fed 10% avocado flour or 10% sunflower oil with 60% forage: 40% concentrate

Variables	Treatment			SEM	P<
	Avocado flour	Control	Sunflower oil		
Initial weight (kg)	24.18	23.39	23.68	0.65	0.6900
Final weight (kg)	48.25^{a}	46.29^{b}	48.70^{a}	0.45	0.0001
Final weight gain (kg)	24.42^{a}	22.57^{b}	24.98^{a}	0.44	0.0001
Daily weight gain (kg)	0.28^{a}	0.27^{b}	0.30^{a}	0.01	0.0020
Daily feed intake (kg)	1.16^{b}	1.16^{b}	1.21^{a}	0.01	0.0001
Feed conversion (kg MS/kg weight)	4.17	4.35	4.11	0.09	0.1500
Feed efficiency (kg weight /kg MS)	0.24	0.23	0.25	0.01	0.1700
Daily protein intake (kg)	0.16^{b}	0.16^{b}	0.17^{a}	0.001	0.0001
Protein conversion (kg PC/kg weight)	0.60	0.61	0.57	0.16	0.2600
Protein efficiency (kg peso/ kg PC)	1.69	1.65	1.76	0.04	0.1800
Daily energy intake (Mcal)	3.44^{b}	2.99^{c}	3.82^{a}	0.02	0.0001
Energy conversion (Mcal/kg weight)	12.78^{a}	11.23^{b}	12.97^{a}	0.33	0.0001
Energy efficiency (kg weight/Mcal))	0.08^{b}	0.09^{a}	0.08^{b}	0.001	0.0020
Slaughter weight at 24 ht (kg)	45.63	42.56	47.35	1.61	0.0900
Carcass weight (kg)	24.22^{ab}	24.12^{b}	25.22^{a}	0.31	0.0200
Carcass yield (%)	52.89	53.20	53.41	0.50	0.7700
Leg weight (kg)	3.35	3.42	3.42	0.07	0.7100
Shoulder weight (kg)	4.78^{b}	5.01^{a}	5.03^{a}	0.07	0.0200
Spine weight (kg)	6.12^{b}	6.38^{ab}	6.76^{a}	0.10	0.0080
Rib weight (kg)	5.10^{b}	5.96^{a}	5.81^{a}	0.17	0.0010

Adjusted means with covariate in model (LSMeans); Different literals indicate significant statistical differences (P<0.05). SEM - Standard error of the mean.

differences were not found in comparison with the control. Efficiency and feed conversion were not affected by the inclusion of avocado flour or sunflower oil in the diet.

Regarding carcass traits, statistical analysis showed no significant differences in carcass yield and leg weight. Conversely, significant differences were recorded in carcass weight, being higher in the diets that included avocado flour or sunflower oil. We reported that the shoulder, spine, and ribs improved with the level of sunflower oil in the diet.

MANEERAT et al. (2015) evaluated the inclusion of agro-industrial by-products, such as bagasse, vinegar, and pineapple hull. The treatments that contained these by-products had the highest values with daily gains of up to 550 g/day in fattening steers. These values could be compared with those reported here since the addition of agro-industrial by-products improved the daily weight gain.

HUANG et al. (2018) mentioned that plant compounds of natural origin that contain tannins, have been used in the nutrition of ruminants to reduce the degradation of proteins at the ruminal level to improve the use of proteins, and make the animal production system more efficient. Traditionally, tannins have been considered as an anti-nutritional factor. Nonetheless, it has been shown that, used correctly, they improve the intestinal microbial ecosystem, intestinal health and increase the flock production. This is a possible explanation of the results obtained in this study: the compounds of plant origin with tannins present in avocado flour may have improved sheep production, as compared to the control treatment.

Conclusion

Here we showed that low levels of inclusion in the diets had a positive effect on *in vitro* dry matter digestibility, production of *in vitro* gas, and the acceptability of sunflower oil and avocado paste meal. The inclusion of 10% avocado or sunflower oil with 40% forage: 60% concentrate was the best diet. Therefore, the use of 10% avocado flour in sheep diets is recommended to increase daily weight gain, without affecting carcass traits, digestibility, and feed intake.

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References

ANKOM (2010): Operator´s Manual, DaisyII Incubator. ANKOM Technology Method 3 In Vitro True Digestibility using the DAISYP II P Incubator ANKOM Technology. Ankom Technology. Macedon, N.Y, pp. 1-2.

ANKOM (2009): Gas Production System. Operator´s Manual. Ankom Technology. Macedon N. Y, pp. 1-48.

ARTEAGA, C. J. D. (2012): Institutional message at the Opening ceremony of VII. Sheep Forum of the State of Mexico. Jilotepec, State of Mexico. (CD-ROM) INIFAP. ICAMEX (in Spanish).

COMBATT, C. E., A. O. JARMA, E. D. PATERNINA (2015): Bromatology of *Brachiaria decumbens Stapf* and *Cynodon nlemfuensis Vanderyst* in acid sulphate soils in Cordoba, Colombia Rev. Mex. Cienc. Agríc 6, 1035-1049.

DOI: 10.29312/remexca.v6i5.597

DINIZ, N. H., F. R. NETO, M. B. CORRÊA, D. H. LOPES, E. N. RONDON, L. F. R. SANTOS, P. F. OLIVEIRA, J. D. LOPES, N. A. OLIVEIRA, I. C. T. CASANOVA, M.T. GOMIDE, W. C. ROBERTO, R. F. ANDRADE, D. T. CRISPIM (2017): Toxicogenetic study of *Persea americana* fruit pulp oil and its effect on genomic instability. Food Chem. Toxicol. 101, 114-120.

DOI: 10.1016/j.fct.2017.01.009.

GONZÁLEZ, G. U. A., R. M. GONZÁLEZ, F. J. G. ESTRADA, G. J. L. BASTIDA, S. N. PECADOR, Z. ABDEL-FATTAH, M. SALEM (2011): Inclusion of pea hay (*Pisum sativum* L.) and *in vitro* gas production in diets for growing lambs. Trop. Subtrop. Agroecosyst. 14, 989-997 (in Spanish).

GRAGEOLA, F., L. SANGINÉS, C. DÍAZ, A. GÓMEZ, M. CERVANTES, C. LEMUS, J. LY (2010): The effect of breed and dietary level of avocado fat on the N and energy balance in young pigs. J. Anim. Feed Sci. 19, 37-49.

DOI: 10.22358/jafs/66268/2010

HERNÁNDEZ-LÓPEZ, S. H., J. G. RODRÍGUEZ-CARPENA, C. LEMUS-FLORES, F. GRAGEOLA-NUÑEZ, M. ESTÉVEZ (2016): Avocado waste for finishing pigs: Impact on muscle composition andoxidative stability during chilled storage. Meat Sci. 116, 186-192.

DOI: 10.1016/j.meatsci.2016.02.018

HUANG, Q., X. LIU, G. ZHAO, T. HU, Y. WANG (2018): Potential and challenges of tannins as an alternative to in-feed antibiotics for farm animal production. Anim. Nutr. 4, 137-150.

DOI: 10.1016/j.aninu.2017.09.004

LEMUS, C., J. BUGARÍN, F. GRAGEOLA, J. G. RODRÍGUEZ, K. MEJÍA, R. VALDIVIA (2017): Chemical characteristics of the Hass avocado paste full fruit (*Persea americana* Mill.) Mexican from Nayarit intended for animal feed. Rev. Comp. Prod. Proc. 24, 112-118 (in Spanish).

MANEERAT, W., S. PRASANPANICH, S. TUMWASORN, V. LAUDADIO, V. TUFARELLI (2015): Evaluating agro-industrial by-products as dietary roughage source on growth performance of fattening steers. Saudi J. Biol. Sci. 22, 580-584.

DOI: 10.1016/j.sjbs.2015.01.015

MARTÍNEZ-MARÍN, A. L., M. PÉREZ-HERNÁNDEZ, L. M. PÉREZ-ALBA, D. CARRIÓN-PARDO, A. G. GÓMEZ-CASTRO (2012): Adding vegetable oils to the diet of dairy goats: effect on digestibility and productive results. Arch. Med. Vet. 44, 21-28 (in Spanish).

DOI: 10.4067/S0301-732X2012000100004

MENKE, K. H., H. STEINGASS (1988): Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. Animal Research and Development 28, 7-55.

MLAMBOA, V., J. L. N. SIKOSANA, F. L. MOULD, T. SMITH, I. OWEN, E. MUELLER-HARVEY (2007): The effectiveness of adapted rumen fluid versus PEG to ferment tannin-containing substrates *in vitro*. Anim. Feed Sci. Tech. 136,128-136.

DOI: 10.1016/j.anifeedsci.2006.08.014

NOM-062-ZOO (1999): Technical specifications for the production, care and use of laboratory animals. Published in the Official Gazette of the Federation. 12/100/2000 22/08/2001 (in Spanish).

OKORUWA, M. I., E. T. E. EHEBHA, E. OMORAGBON (2015): Replacement value of avocado seeds with orange peels meal for guinea grass on growth performance and nutrient digestibility of growing sheep. Int. J. Res. Agric. Forest. 2, 35-40.

ORTIZ-LÓPEZ, B., M. A. MARIEZCURRENA-BERASAÍN, R. BARAJAS-CRUZ, P. LÓPEZ-PEREA, H. B. ESCALONA-BUENDÍA, J. L. BÓRQUEZ-GASTELUM, M. D. MARIEZCURRENA-BERASIN (2016): Influence of tannins on physical and sensory characteristics of bovine meat in fattening. Rev. Mex. Cien. Pec. 7, 309-320 (in Spanish).

DOI: 10.22319/rmcp.v7i3.4212

PATRA, A. K., J. SAXENA (2010): Review: A new perspective on the use of secondary plant metabolites to inhibit methanogenesis in the rumen. Phytochemistry 71, 1198-1222 (in Spanish).

DOI: 10.1016/j.phytochem.2010.05.010.

ROSSI, C. A., M. DE LEÓN, G. L. GONZÁLEZ, P. C. DIB, A. M. PEREYRA (2008): Chemical composition, total polyphenol content and nutritional value in browsing species of the silvopastoral system of the arid Argentine Chaco. Zootecnia Tropical 26, 105-115 (in Spanish).

SANTANA, G., H. J. CORREA (2016): Inclusion of three lipid sources in the diet of lactating cows under grazing on consumption and digestibility. Liv. Res. Rur. Dev. 28, 38 (in Spanish).

SAS (2004): Statistical Analysis System. User´s guide. SAS Institute Inc., SAS Campus Drive, Cary, North Carolina, pp. 421-480.

SIAP (2017): Agri-Food and Fisheries Information Service. Agrifood Atlas First edition, Mexico City, Mexico. www.gob.mx/siap (in Spanish).

TARAZONA, A. M., M. C. CEBALLOS, J. F. NARANJO, C. A. CUARTAS (2012): Factors affecting forage intake and selectivity in ruminants. Rev. Colomb. Cienc. Pecu 25, 473-487 (in Spanish).

THEODOROU, M. K., B. A. WILLIAMS, M. S. DHANOA, A. B. MCALLAN, J. FRANCE (1994): A simple gas production method using a pressure transducer to determine the fermentation kinetics of ruminant feeds. Anim. Feed Sci. Tech. 48, 185-197.

DOI: 10.1016/0377-8401(94)90171-6

TILLEY, J. M. A., R. A. TERRY (1963): A two-stage technique for the *in vitro* digestion of forage crops. Grass Forage Sci. 18, 104-111.

DOI: 10.1111/j.1365_2494.1963.tb00335.x

UCHENNA, U. E., A. B. SHORI, A. S. BABA (2017): Inclusion of avocado (*Persea americana*) seeds in the diet to improve carbohydrate and lipid metabolism in rats. Rev. Arg. Endocri. Metabol. 54, 140-148.

DOI: 10.1016/j.raem.2017.07.005

VASTA, V., G. LUCIANO (2011): The effects of dietary consumption of plants secondary compounds on small ruminants' products quality. Small Rumin. Res. 101, 150-159 (in Spanish).

DOI: 10.1016/j.smallrumres.2011.09.035

WANAPAT, M., S. KANG, K. PHESATCHA (2013): Enhancing buffalo production efficiency through rumen manipulation and nutrition. Buffalo Bulletin 32, 258-275.

DOI: 10.2174/978168108417611701009

WU, D., L. XU, S. TANG, L. GUAN, Z. HE, Y. GUAN, Z. TAN, X. HAN, C. ZHOU, J. KANG, M. WANG (2016): Influence of oleic acid on rumen fermentation and fatty acid formation *in vitro*. PLoS ONE 11, 1-13.

DOI: 10.1371/journal.

YANG, S. L., D. P. BU, J. Q. WANG, Z. Y. HU, D. LI, H. Y. WEI, L. ZHOU, J. J. LOOR (2009): Soybean oil and linseed oil supplementation affect profiles of ruminal microorganisms in dairy cows. Animal 3, 1562-1569.

DOI: 10.1017/S1751731109990462

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LEMUS-FLORES, C., J. O. BUGARIN PRADO, F. GRAGEOLA NUÑEZ, R. VALDIVIA BERNAL, I. RUIZ DIMAS, J. A. BONILLA CARDENAS, J. C. SEGURA CORREA: Učinak praha avokada, suncokretova ulja te različitih omjera voluminozne i koncentrirane krme u obroku na unos hrane, probavljivost hrane i proizvodnost mladih ovnova. Vet. arhiv 90, 353-364, 2020.

SAŽETAK

Cilj rada bio je istražiti utjecaj različitih omjera između praha avokada i suncokretova ulja kao izvora energije, te različitih omjera između voluminozne i koncentrirane krme u obroku na unos hrane i proizvodnost mladih ovnova. Osim toga, in vitro istražena je probavljivost suhe tvari, probavljivost organske tvari i proizvodnja plina. Unos hrane mjeren je u obrocima koji su sadržavali prah avokada ili suncokretovo ulje uz omjer između voluminozne i koncentrirane krme od 60:40% ili u obrocima sa različitim omjerom voluminozne i koncentrirane krme uz dodatak 10% praha avokada ili suncokretova ulja. Dnevni prirast bilježen je tijekom 84 dana. Probavljivost suhe tvari in vitro određivana je 48 h, a proizvodnja plina in vitro 72 h od uzimanja uzoraka. Za određivanje metaboličke energije i probavljivosti organske tvari korišteni su parcijalna proizvodnja plina 24-ti sat, sirovi protein, sirova mast i pepeo. Niske razine praha avokada i suncokretova ulja u obroku rezultirale su najvišim unosom hrane te najvišom in vitro probavljivošću suhe tvari i in vitro proizvodnjom plina. Dnevni prirast bio je 0,28 kg u životinja kojima je davano 10% praha avokada, 0,30 kg u životinja kojima je davano 10% suncokretova ulja, pri čemu je omjer voluminozne i koncentrirane krme iznosio 40:60%. Dodavanje visokih razina praha avokada ili voluminozne krme u obrok dovelo je do nižeg unosa hrane kod mladih ovnova pa je zaključeno da je najpovoljnija razina dodanog praha avokada 10% uz omjer voluminozne i koncentrirane krme 40:60%.

Ključne riječi: prihvatljivost; probavljivost; proizvodnja plina; avokado; ovnovi