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Fattening performance and carcass characteristics of lambs supplemented with condensed tannins from *Acacia mearnsii* extract

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Abstract

Tannins are polyphenolic compounds with some beneficial properties for ruminants as they act as antioxidants, antimicrobials, and anthelmintics, which may improve animal performance. However, the results are variable, depending on the type, source, and dose. The objective of this research was to determine the effect on fattening performance and carcass characteristics of lambs supplemented with condensed tannins (CTs) from *Acacia mearnsii* extract. Thirty-six, three-month-old Dorset x Hampshire cross lambs of 20.8 ± 3.3 kg live weight were used in a completely randomized design with four treatments (n = 9), namely T1: basal diet, 0.0 g CT/kg DM^{-1}; T2: T1 + 1.75 g CT/kg DM^{-1}; T3: T1 + 3.5 g CT/kg DM^{-1}; and T4: T1 + 5.25 g CT/kg DM^{-1}. The daily weight gain was higher in T2 and T3 than in T1. The weight at slaughter and empty weight at slaughter were higher in T2 than in T1. The hot carcass weight was higher in T3 than in T1, but no different from T2 or T4. The hot carcass yield was higher in T3 than in the other treatments. The meat pH at slaughter and 24 hours postmortem was higher in T4 than in T1 and the meat protein percentage was higher in T3 than in T1. The inclusion of CTs from *Acacia mearnsii* extract in the diet of fattening lambs increased their daily weight gain, yield, and carcass weight.

Keywords: carcass yield, condensed tannins, fattening lambs, meat composition
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Introduction

Currently, the global population is increasing its preference for the consumption of cleaner and safer foods of animal origin and is concerned about the excessive use of antibiotics and other chemicals in the feed of domestic animals to improve their performance. For this reason, interest has arisen in the use of natural dietary phytochemicals as additives to improve the productive response of animals (Salami *et al*., 2018) without risk to human health. In recent years, the study of ruminant nutrition has been based on feeding alternatives focused on the manipulation of the rumen microbiota to improve the efficiency of its metabolism and the animal's productive performance (Patra & Saxena, 2011). In this context, the use of secondary metabolites such as tannins, which are present in plants, is highlighted as a modulating additive of ruminal fermentation (Tontini *et al*., 2021). Tannins have been widely studied in animal nutrition and production, especially their biological activities and the response of animals to their consumption, which is relevant in modern livestock (Mueller-Harvey, 2006; Waghorn, 2008; Huang *et al*., 2018).

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Tannins are polyphenolic compounds with some beneficial properties for ruminants, as they act as antioxidants, antimicrobials, and anthelmintics, which may improve animal performance. However, the results reported are variable, depending on the type, source, and dose used. The most notable effects attributed to condensed tannins (CTs) in ruminant nutrition refer to the mitigation of ruminal methane production (Piñeiro-Vázquez *et al*., 2015), anthelmintic effects in small ruminants (Katiki *et al*., 2013), antioxidant activity in meat (Luciano *et al*., 2009, 2011; Cunha *et al*., 2018), and protein binding to avoid their degradation in the rumen. However, not all CTs are biologically active, and may not have the ability to complex with proteins (Naumann *et al*., 2018). Some studies have shown that the inclusion of natural sources of polyphenols such as CTs in the diet of small ruminants improves productive variables, preserves the quality of the meat, prolongs shelf life, and prevents deterioration (Dawson *et al*., 2011; Elmer *et al*., 2017; Mkhize *et al*., 2018). The results have been variable, depending on the source of tannins and the dose used in the ration (McNabb *et al*., 1993; Luciano *et al*., 2009, 2011; Liu *et al*., 2016; Guerra-Rivas *et al*., 2016; Ortiz-López *et al*., 2016; Lobón *et al*., 2017; Kamel *et al*., 2018; Guerreiro *et al*., 2020). Therefore, the objective of this study was to determine the fattening performance and characteristics of the lambs supplemented with CTs from the extract of *Acacia mearnsii* (EAM).

Materials and Methods

Animal care and management procedures involving lambs were conducted according to the guidelines approved by the Mexican official norms (NOM-051-ZOO-1995: Humanitarian Care of Animals during Mobilization; NOM-033-SAG/ZOO-2014: Slaughter of Domestic and Wild Animals). In addition, the lambs were managed according to the regulations for the use and care of animals destined for research at the College of Postgraduates, Texcoco, Mexico. This study was carried out at the Ruminant Metabolic Unit of the experimental farm and in the Animal Nutrition Laboratory of the Livestock Postgraduate Programme of the Postgraduate College, Texcoco, Mexico.

A productive behaviour test was carried out for 85 days, 15 for adaptation and 70 for data collection. Thirty-six Dorset x Hampshire cross lambs were used with an initial live weight of 20.8 ± 3.3 kg. The lambs were dewormed (Closantil oral® 5%; 1 ml.5 kg^{-1} LW oral route), vaccinated (Biobac® 11, 2.5 $mL.animal^{-1}$), and vitaminized (Vigantol® ADE; 0.025 $mL.kg^{-1}$ LW). The animals were housed in individual, elevated metabolic cages (1.0 × 1.5 m) (animal density of 1.5 m^2 per lamb), equipped with a feeder and automatic drinker. Food was offered *ad libitum,* twice a day in quantities of 60% (0.700 kg) in the morning, and the remaining 40% (0.500 kg) in the afternoon (09h00 and 17h00).

The EAM (SETA®, Estância Velha, Brasil) was a commercial product with a fine, brown, powdery appearance. The content of CTs in the extract was 700 g/kg DM^{-1} and 62 g/kg dry matter. An isonitrogenous and isoenergetic basal diet was formulated for a daily weight gain of 300 g for lambs (NRC, 2007). The treatments consisted of T1 = basal diet; T2 = T1 + 0.25% EAM (CT = 1.75 g/kg DM^{-1}); T3 = T1 + 0.5% EAM (CT = 3.5 g/kg DM^{-1}); and T4 = T1 + 0.75% EAM (CT = 5.25 g/kg DM^{-1}) (Table 1). During the behaviour test, samples were collected every 15 days from each of the diets, and at the end of the experiment, they were mixed to obtain a sample composed of each treatment to determine the content of dry matter (DM) (method 934.01), crude protein (CP) (method 2000.11), ash (method 942.05), ether extract (EE) (method 920.39) (AOAC, 2005), acid detergent fibre (ADF), and neutral detergent fibre (NDF) (Van Soest *et al*., 1991). The laboratory tests were undertaken in the Animal Nutrition Laboratory of the Colegio de Postgraduados, México.

The dry matter intake (DMI) (g/day) was determined by the difference in weight between the feed offered and that rejected from the previous day. The daily weight gain (DWG) was calculated by weighing the animals at the beginning of the experiment and subsequently every 14 days before offering the morning feed, divided by 14 days. The feed conversion (FC) was calculated by dividing the DMI by the DWG. The final weight (FW) was recorded on the last feeding day of the experimental phase and the weight at slaughter (WS) was recorded 12 hours prior to slaughter.

Table 1 Ingredients and proximate analyses of the basal diet for fattening lambs

Ingredient	g/kg DM	Nutrient	Quantity
Ground corn	650	Dry matter (DM), %	90
Soybean meal	80	Ash, % DM	4.80
Oat hay	200	Crude protein, % DM	13.40
Molasses	50	Ether extract, % DM	7.80
Mineral premix and vitamin[1]	20	Neutral detergent fibre	22.67
		Acid detergent fibre	9.25
		Metabolizable energy, MJ/kg DM	11.74

[1]Calcium: 24%, chlorine: 12%, magnesium: 2%, phosphorus: 3%, potassium: 0.50%, sodium: 8%, sulfur: 0.50%, chromium: 5 mg/kg DM, cobalt: 60 mg/kg DM, iodine: 100 mg/kg DM, iron: 2000 mg/kg DM, manganese, 4000 mg/kg DM, selenium: 30 mg/kg DM, zinc: 5000 mg/kg DM, lasalocid: 2000 mg/kg DM, vitamin A: 500 000 IU/kg, vitamin D: 150 000 IU/kg, vitamin E: 1000 IU kg^{-1}

The lambs were slaughtered in accordance with the official Mexican standard NOM-033-SAG/ZOO-2014. After 12 hours of fasting and prior to transport, the lambs were weighed with a digital scale (Torrey, CRS-HD, 200 kg capacity).

At slaughter, blood, head, skin, extremities, red viscera (lungs, liver, heart), full and empty green viscera (rumen, reticulum, omasum, abomasum, and intestines) were weighed and defined as gastrointestinal content and viscera (GC) The empty weight at slaughter (EWS) was calculated by subtracting the weight of the GC from the WS and the hot carcass weight (HC) was recorded. The yield of hot carcass (YHC) and the biological yield of hot carcass (BYHC) were obtained using these formulae:

YHC = [(HC / WS) * 100]

BYHC = [(HC / EWS) * 100], according to Rios Rincóna *et al.* (2012).

The back fat (BF) was measured on each animal using a Sonovet 600 ultrasound (Universal Medical System, Inc.) with a 7.5 MHz transducer, between the twelfth and thirteenth ribs, at the beginning of the experiment and three days before slaughter. The pH was measured twice in a sample of the *Longissimus dorsi* muscle from the hot carcass, at 30 minutes and 24 hours after slaughter using a portable potentiometer (HANNA, mod. HI99163), with a penetrating electrode. The content of DM, CP, ash, and EE of the meat samples was determined according to the methodology described by the AOAC (2005).

A completely randomized experimental design was used and the data were analysed with PROC GLM (SAS, 2002 Version 9.0. Cary, NC: Institute Inc. Campus Drive.), regarding the initial live weight (IW) as a co-variate and each lamb as an experimental unit. The means of results were compared using Tukey's test ($P \leq 0.05$) (Steel *et al*., 1997).

Results and Discussion

The inclusion of CT in the diet of lambs increased ($P < 0.05$) the FW, DWG, and DMI in T2 and T3 more than in T1 but was no different from T4. However, FC was better in T2 than in T1, although no different from the other treatments with CT. There were no differences ($P > 0.05$) in BF among treatments (Table 2).

The WS and EWS were higher ($P < 0.05$) in T2 than in T1 but no different from T3 or T4. However, the HC was higher ($P < 0.05$) in T3 than T1 but no different from T2 or T4. The YHC was higher in T3 than in the other treatments and the BYHC was higher (P <0.05) in T3 and T4 than in T2 or T1. The GC and pH of the meat were higher ($P < 0.05$) in T4 than in T1 but no different from the other CT treatments (Table 3).

Table 2 Productive performance of lambs supplemented with condensed tannins

Item	Treatments				
	T1	**T2**	**T3**	**T4**	**SE**
Initial weight, kg	20.32	20.76	21.41	20.58	0.551
Final weight, kg	35.12[b]	39.28[a]	39.48[a]	37.62[ab]	0.884
Daily weight gain, g/d	211[b]	269[a]	260[a]	251[ab]	0.008
Dry matter intake, kg/d	0.947[b]	1.051[a]	1.095[a]	1.040[ab]	0.023
Feed conversion	4.55[b]	4.08[a]	4.26[ab]	4.36[ab]	0.085
Backfat depth, mm	3.11	3.00	3.60	3.00	0.089

T1: control, T2: 1.75 g condensed tannins/kg DM, T3: 3.25 g condensed tannins/kg DM, T4: 5.25 g condensed tannins/kg DM
[ab] Within a row, means with a common superscript did not differ with probability P <0.05

Table 3 Weight and yield of lamb carcasses supplemented with condensed tannins

Item	Treatments				
	T1	**T2**	**T3**	**T4**	**SEM**
Slaughter weight, kg	38.14[b]	41.51[a]	40.85[ab]	39.60[ab]	0.837
Empty body weight, kg	35.43[b]	38.40[a]	37.38[ab]	35.82[ab]	0.775
Hot carcass weight, kg	18.83[b]	20.53[ab]	21.66[a]	20.28[ab]	0.511
Hot carcass yield, %	49.29[b]	49.42[b]	52.92[a]	50.98[b]	0.388
Biological yield, %	53.05[b]	53.38[b]	57.83[a]	56.40[a]	0.447
Gastrointestinal contents, kg	2.71[b]	3.10[ab]	3.46[ab]	3.78[a]	0.122
pH at slaughter	6.35[b]	6.36[ab]	6.44[ab]	6.66[a]	0.042
pH 24 h postmortem	5.75[b]	5.78[ab]	5.90[ab]	5.98[a]	0.029

T1: control, T2: 1.75 g condensed tannins/kg DM, T3: 3.25 g condensed tannins/kg DM, T4: 5.25 g condensed tannins/kg DM
[ab] Within a row, means with a common superscript did not differ with probability P <0.05

The CP percentage in the lamb meat was higher (P <0.05) in T3 than in T1 and the ash content was lower (P <0.05) in T4 than in T1. There were no differences in the percentage of moisture or EE between treatments (Table 4).

Table 4 Chemical composition of lamb meat supplemented with condensed tannins

	Treatments				
Item	**T1**	**T2**	**T3**	**T4**	**SEM**
Moisture, %	73.43	72.09	71.21	71.65	0.5598
Crude protein, %	18.64[b]	19.15[ab]	20.44[a]	19.50[ab]	0.2549
Ether extract, %	3.63	4.17	4.45	5.02	0.2538
Ash, %	3.93[a]	3.90[a]	3.31[ab]	2.91[b]	0.1313

T1: control, T2: 1.75 g condensed tannins/kg DM, T3: 3.25 g condensed tannins/kg DM, T4: 5.25 g condensed tannins/kg DM
[ab] Within a row, means with a common superscript did not differ with probability P <0.05

The results of the present study showed an increment of up to 13.5% in DMI, 21.5% in DWG, and 11.0% in the FW of lambs supplemented with CT. As a result, FC was improved by up to 10.0%. These improvements were reflected in an increment of 9.1% WS, 7.7% EWS, 13% HC, 6.8% in the YHC, and 8.2% in the BYHC. The effect of improving the productive variables and carcass characteristics because CTs were added to the diets may be explained by the formation of a tannin–dietary protein complex. In addition, tannin–microbial enzyme complexes occur at the ruminal level because of the pH (5.8–6.8) protecting it from enzymatic degradation by proteolytic microorganism DM. This promotes it reaching the abomasum, where the tannin–protein complexes dissociate because of the acidic pH (< 3.5). Once dissociated, and through the action of pancreatic and intestinal enzymes, proteolysis occurs and amino acids are absorbed at intestinal level (Mueller-Harvey, 2006; Patra & Saxena, 2011; Chikwanha *et al*., 2019; Ebrahim & Negussie, 2020), providing a greater amount of metabolizable amino acids to promote muscle synthesis.

However, some studies with higher doses of CTs, negative effects on DWG, DMI, and digestibility were reported. Costa *et al*. (2021) reported that increasing doses of CT from 20 to 80 g/kg DM^{-1} from *Acacia mearnsii* affected DMI negatively and reduced DWG by up to 51.3% with a dose of 80 g CT/kg DM^{-1}. Gerlach *et al*. (2018) showed that a dose of 6.2 g CT/kg DM^{-1} of EAM decreased the digestibility of organic matter by 21% and concluded that an excess of phenolic compounds in the diet could have affected some microorganism DM and enzymes in the rumen, affecting the intake and digestibility of organic matter. The level of CT inclusion was not high enough to affect some microorganisms or their enzymatic activity, which was reflected in the productive response of the lambs.

The consumption of DM in ruminants is affected by the astringency that tannins present and the formation of tannin–protein complexes that are initially generated by the presence of salivary proteins (Costa *et al*., 2021). However, the reports on the effect of tannins on the consumption of DM are varied and depend largely on the source, types of tannin, and the dose, although it seems that most researchers agree that doses higher than 40 g CT/kg DM^{-1} negatively affect the intake and digestibility of DM (Gerlach *et al*., 2018; Mergeduš *et al*., 2020; Orlandi *et al*., 2020; Costa *et al*., 2021). Huang *et al*. (2018) stated that a low to moderate concentration of CTs in the diets (< 50 g/kg of DM^{-1}) did not affect the consumption of DM or the digestibility of nutrients. On the other hand, Costa *et al*. (2021) reported that in lambs fed diets with EAM in doses greater than 40 g/kg DM^{-1}, animal performance was affected, with reductions in weight gain, feed intake, and digestibility. In the current study, the doses of CTs from *Acacia mearnsii* did not affect consumption and weight gain of the lambs but stimulated it. These results may be because the doses of CT from *Acacia mearnsii* were relatively low. In the present study, the FW, YHC, and percentage of protein in the meat were higher with a dose of 3.5 g CT/kg DM^{-1}. Orlandi *et al*. (2020) stated that relatively low doses of tannins in the diet could improve the contribution of metabolizable protein in ruminants.

Dentinho *et al*. (2020) reported a positive response in carcass performance, DWG, and feed and protein efficiency from adding 15% *Cistus ladanifer* extract as a source of CTs per kilogram of soybean. These results agree with the current study, although the type of tannin was different. Dentinho *et al*. (2020) explain that this effect was attributed to the tannin–protein complex, which made it possible to protect the soybean protein from ruminal degradation, resulting in an increased rumen bypass outflow to the posterior tract, increasing its absorption in the small intestine. The reaction that generates the formation of tannin–protein complexes at ruminal level may reflect a greater contribution of metabolic protein, since the contribution of protein of microbial origin oscillates at 64%. On the other hand, the CP from the diet, which is not degraded in the rumen, could contribute up to 80% (Elmer *et al*., 2017).

The passage of more abundant metabolic protein to the small intestine is reflected in a greater availability of amino acids to be absorbed and used in the synthesis of protein and muscle mass by the body of the animal (Zhao *et al*., 2018; Chikwanha *et al*., 2019). Barry and McNabb (1999) stated that a concentration of 1 or 2 g CT/kg DM^{-1} was not enough to prevent protein degradation in the rumen, and that at least 5 g CT/kg DM^{-1} was needed to achieve the formation of tannin–protein complexes so that these could pass into the abomasum–duodenum. In the current study, the best productive responses of lambs were obtained with concentrations of 3.5–5.25 g CT/kg DM^{-1}. Similarly, a pH value ≥6 in meat affects its quality (Devine *et al*., 1993) by altering the water retention capacity (Bouton *et al*., 1971). In the current study, the pH at slaughter and the pH of the meat at 24 hours postmortem was improved as the dose of CT in the diet increased, although the pH of the meat at 24 hours was slightly lower than at the time of slaughter. The final pH value at 24 hours postmortem depends on factors such as the degree of ante-mortem stress (duration of transport and handling during slaughter), age of the animal, climate, nutritional status, temperament, and the health of the animal (Ponnampalam *et al*., 2017). However, those results were different from those reported by Liu *et al*. (2016), who used Quebracho tannins up to 1% in the diet of lambs subject to heat stress and reported that the meat pH decreased as the tannin dose increased. Meat with a higher pH can maintain its tenderness for longer and the shear force is lower (Priolo *et al*., 2000).

The use of CTs as a supplement in the feeding of ruminants can be an alternative to chemical supplements that are used to increase the animals' response and thus can provide cleaner and safer foods of animal origin for human consumption. However, research on the use of tannins as a supplement in ruminant feeding must be increased, since the results in the scientific literature are variable, depending on the source, dose, chemical structure, type of tannin, and the diet. Nevertheless, CTs offer a good alternative to improve the productive response of ruminants.

Conclusions

The results of the present research showed that it was possible to improve the performance of lambs under an intensive fattening production system when supplemented with relatively low doses of CTs from *Acacia mearnsii* extract since the lambs showed better weight gains and carcass performance.

Authors' Contributions

AGS conducted the research and wrote the manuscript, under the supervision of JRBG. For laboratory analyses LAM and MACP collaborated. For lamb handling, slaughter, and carcass characteristics, DHS and SSGM collaborated. HVH led the data analysis and interpretation of results. Critical revision of the version to be published was performed by JRBG, SSGM and AGS.

Conflict of Interest Declaration

The authors certify that they have no affiliations with any organization or entity with any financial or non-financial interest in the subject matter or materials discussed in this manuscript.

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