

## Short Communication



## Effects of Long-Term Supplementation of Different Levels of Tannins Extract on Meat Quality and Carcass Traits of Hairy Lambs

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**Abstract** | Forty male hairy lambs ( $\frac{1}{4}$  Pelibuey and  $\frac{3}{4}$  Katahdin,  $31.53 \pm 3.8$  kg initial LW) were fed during 70 days with total mixed ration corn-based finishing diet supplemented with 0, 2, 4, or 6 g tannins extract/kg dietary dry matter (Control, TAN2, TAN4, and TAN6, respectively) in order to evaluate the effects of long-term tannin supplementation on meat quality. The tannins extract (TAN) contained a minimum of 70% tannin, comprised of a 50:50 blend of both condensed and hydrolysable forms. Average net intake of tannin were 1.71, 3.45, and 5.2 g of tannin/day for TAN2, TAN4, and TAN6, respectively. Lambs, were slaughtered at  $48.14 \pm 4.8$  kg averaging  $27.77 \pm 2.78$  kg of carcass weight. Records and meat samples of each carcass were obtained and analyzed (10 samples/treatment). There were no treatment effects on carcass traits. Tannin supplementation did not affect pH, drip loss or cook loss. However, tannin supplementation linearly increased ( $P \leq 0.04$ ) the *L* value and shear force, and tended to increase ( $P=0.08$ ) water holding capacity being maximal at high supplementation level (6 g TAN/kg dietary DM). It is concluded that long-term supplementation of tannins may affect color and tenderness of meat lamb when is supplemented beyond 4 g/kg DM. However, further research must be performed in order to establish the maximum dose level of supplementation with positive effects on meat quality but without detrimental effects of tannins on feed intake, nutrient utilization and health.

**Keywords** | Tannins, Lambs, Finishing phase, Meat quality, Carcass characteristics

**Received** | August 06, 2021; **Accepted** | September 06, 2021; **Published** | October 01, 2021

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**Citation** | Pérez BIC, Rojas-Román LA, Estrada-Angulo A, Muro OC, Barreras A, Plascencia A (2021). Effects of long-term supplementation of different levels of tannins extract on meat quality and carcass traits of hairy lambs. *Adv. Anim. Vet. Sci.* 9(11): 1973-1977.

**DOI** | <http://dx.doi.org/10.17582/journal.aavs/2021/9.11.1973.1977>

**ISSN (Online)** | 2307-8316; **ISSN (Print)** | 2309-3331

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## INTRODUCTION

Tannins are a complex group of polyphenolic compounds found in many plant species commonly consumed by ruminants. They are conventionally classified into two major groups: Hydrolysable and condensed tannins (Khanbabaee and van Ree, 2001). Even when tannins be toxic when it consumes at high concentrations (reduce voluntary feed intake and nutrient digestibility), at moderate level of intake has been shown benefits on

growth performance and/or dietary energy utilization in cattle (Rivera-Méndez *et al.*, 2016) and lambs (Rojas-Román *et al.*, 2017). In this sense, the use of supplemental tannins as a feed additive it has been used in recent years in growing-finishing rations. Recent studies have showed that type of dietary tannins (soluble or condensed) can affect in a different way ruminal microbial metabolism by several mechanism including inhibition of extracellular microbial enzymes, inhibition of oxidative phosphorylation or metal ions deprivations (Huang *et al.*, 2018). This cause changes

in end-products of ruminal fermentation can be advantage of some meat quality attributes in wool lambs (Biondi et al., 2019; Luzardo et al., 2019). Tannins extract dose from, 2.7 up to 7.3 g/kg diet DM have been positive effects on meat quality of wool lambs (Vasta et al., 2007; Gravador et al., 2015), but in hairy finishing lambs the information about of the tannins supplementation on meat quality is limited. It's well recognized that breed play an important role on lamb meat quality (Ramírez-Retamal and Morales, 2014). In this sense, there is information regard of the supplementation effects of moderate levels of tannins (0.43 g/kg diet DM) on hairy lambs during short-period (i.e. <40 d; Sánchez et al., 2019), but information of long-term supplementation period (> 63-d) in finishing hairy lambs on meat quality is limited. It has been reported that the period of supplementation can determine the magnitude of the effects of tannins supplementation (Rojas-Román et al., 2017); therefore, the aim of the present study was to investigate the dietary supplementation of a different levels (4 and 6 g TAN/kg diet DM) concentrate source of hydrolysable and condensed tannins on meat quality of hairy lamb finished with a high-energy diet during 70 days. Due to the nature of lamb's growing, longer term finishing phase are from 9 to 11 weeks, we chose 70-d because the initial weight at start of the experiment and target final weight to slaughter (45 kg LW).

This experiment was conducted at the Universidad Autónoma de Sinaloa Feedlot Lamb Research Unit. All animal management and care procedures were in accordance with the guidelines approved by the Universidad Autónoma de Sinaloa Animal Use and Care Committee (Protocol # 07082019). Forty Pelibuey × Katahdin (¼ Pelibuey and ¾ Katahdin, 175 ± 18 d age; 31.53 ± 3.8 kg initial LW) intact male lambs were used in a 70-d growth-performance experiment to evaluate the long-term tannins ingestion effects on meat quality. Lambs were placed in 20 pens (2 lambs/pen; 10 lambs/treatment). Description of diets and the management of experimental units prior and during the experiment were previously described by Rojas-Román et al. (2017). Briefly, dietary treatments consisted of a total mixed corn-based finishing diet (16.23% crude protein, 17.45% neutral detergent fiber, 3.57% ether extract, and 2.01 Mcal of net energy for maintenance/kg) supplemented with 0, 2, 4 or 6 g tannin extract/kg dietary dry matter. The tannin extract (TAN) consisted of a 50:50 blend of condensed (quebracho) and hydrolysable (chestnut) phenolic polymers (minimum 70% tannin; ByPro, Silvateam, Ontario, CA). Supplemental tannins was hand-weighed using a precision balance (Ohaus, mod AS612, Pine Brook, NJ), and premixed for 5 min with the other minor dietary ingredients (urea, limestone and trace mineral salt) before incorporation into complete mixed basal diet using a 2.5 m<sup>3</sup> capacity paddle mixer (mod 30910-7, Coyoacán, México). To avoid contamination, the

mixer was thoroughly cleaned between each treatment. All lambs were harvested on the same day (18-h fasted). Lambs were stunned (captive bolt), exsanguinated and skinned, the gastrointestinal organs were separated, and hot carcass weight (HCW) was registered. After carcasses (with kidneys and internal fat included) chilled in a cooler at -2 to 1°C for 24 h, the following measurements were obtained: 1) cold carcass weight (CCW); 2) subcutaneous fat (fat thickness) was taken over the 12<sup>th</sup> to 13<sup>th</sup> thoracic vertebrae; 3) LM surface area, measure using a grid reading of the cross-sectional area of the *longissimus* muscle between 12<sup>th</sup> and 13<sup>th</sup> rib, and 4) kidney, pelvic and heart fat (KPH) was removed manually and afterward weighed and reported as a percentage of the cold carcass weight. Meat pH and temperature were measured at 1 and 24 h post-slaughter between the 4<sup>th</sup> and 5<sup>th</sup> lumbar vertebra with a portable pH meter (MP230, Mettler Toledo, Plano, Tx, USA.). Calibration was performed using buffer pH 4 and 7. After carcasses (with kidneys and internal fat included) were chilled in a cooler at -2°C to 1°C for 48 h. At 24-h of chilling, two longissimus muscle (LM) steaks (3-cm thick) from each carcass (10 per treatment) were removed between 12<sup>th</sup> and 13<sup>th</sup> rib interface, preserved immediately on dry ice and shipped to the Meat Quality Laboratory for storage at 4°C until days postmortem. At 14 days postmortem, steaks were frozen at -20°C vacuum packaged, and stored for subsequent meat quality trait analysis. Variables measured included water holding capacity (WHC), color, purge loss (PL) at 24 and 48-h, cook loss (CL), and shear force (SF). The color values *L*<sup>\*</sup> (lightness), *a*<sup>\*</sup> (redness), and *b*<sup>\*</sup> (yellowness) were determined using a Minolta CR-410 spectrophotometer (Konica Minolta Camera Co., Ltd, New Jersey, USA). The chroma (*C*<sup>\*</sup>) and hue angle (*h*<sup>°</sup>) were estimated as  $C^* = [(a^*)^2 + (b^*)^2]^{1/2}$ , and  $h^\circ = \tan^{-1}(b^*/a^*)$ . The 10-cm-thick steaks previously obtained from the rib were thawed and cooked at 21-d postmortem followed the procedures described by López-Carlos et al. (2014), previously cooked steaks were aged at 4°C for 24 h. To obtain SF values, 1×1×3 cm cores were taken from each cooked steak parallel to the orientation of the muscle fibers. The SF measurements (kg/cm<sup>2</sup>) were determined using a Lloyd texturometer (Lloyd Instruments, Fareham, Hampshire, UK) equipped with Warner-Bratzler shear blades with a crosshead speed of 50 mm/min. Water-holding capacity was determined using a modified compression technique from the method termed press-juice, in which 0.3 kg of a meat sample is positioned between 2 layers of filter paper and 2 plaques of acyclic Plexiglas, and compressed at a force of 5 newton (5 N) for 60 s using the Lloyd texturometer. The WHC was estimated as juice lost divided by the initial sample mass. Drip loss was measured using the technique described by López-Carlos et al. (2014).

All the data were tested for normality using the Shapiro-

Wilk test. Data were analyzed by ANOVA using the MIXED procedure of SAS Software (2007). The results were analyzed according to a randomized complete block design with subsampling, with pen as the experimental unit and animal as the observational unit. Treatment effects were tested for linear, quadratic and cubic components of the TAN level. In addition, means separations using the honestly difference test (Tukey's Test) were performed between TAN levels. Treatment effects were considered significant when the value of  $p \leq 0.05$ , and were identified as trends when the  $p$ -value was  $> 0.05$  and  $\leq 0.10$ . between TAN levels were performed. Contrasts were considered significant when the  $P$ -value was  $\leq 0.05$ , and tendencies were identified when the  $P$ -value was  $> 0.05$  and  $\leq 0.10$ .

Weight at slaughter were 48.34, 48.03, 48.30, and 47.87 kg for Control, TAN2, TAN4, and TAN6, respectively. The carcass responses to the treatments were previously reported by Rojas-Román et al. (2017), in which tannin supplementation did not affect ( $P > 0.25$ ) hot carcass weight, dressing percentage, kidney-pelvic-heart fat, fat thickness, LM area, nor shoulder muscle composition in which the average values were  $27.77 \pm 2.8$  kg,  $57.71 \pm 1.2\%$ ,  $2.87 \pm 0.8\%$ ,  $3.02 \pm 0.44$  mm,  $19.91 \pm 1.6$  cm<sup>2</sup>,  $60.7 \pm 0.02\%$ , and  $20.6 \pm 0.1\%$ , respectively.

Treatments effect on meat quality are shown in Table 1. Tannin supplementation did not affect pH, drip loss or cook loss. However, tannin supplementation linearly increased ( $P \leq 0.04$ ) the  $L$  value and shear force, and tended to increase ( $P = 0.08$ ) water holding capacity being maximal at high supplementation level (6 g TAN/kg dietary DM). The variation in meat pH influences factors such as color and the ability of the meat to retain water. A low ultimate pH results in meat proteins having decreased water-holding capacity and a lighter color. Conversely, a higher ultimate pH will give a darker color and less drip loss. In our experiment, tannin supplementation did not affect pH recorded at 1 and 24-h postmortem. The absence of effects on meat pH when tannins is supplemented is consistent with previous report (Biondi et al., 2019). In contrast, Min et al. (2012) reported a faster decline in muscle pH within 8 h postmortem in meat goats receiving diets supplemented with 4% tannins, although final pH at 24 h postmortem was similar with controls. The inconsistencies on the effects of tannins on meat pH could be mediated by factors such as time of sample taken and received tannins dose concentration level. Consistent with the pH registered in our experiment, drip loss or cook loss were not affected by tannin supplementation. Similarly, Garcia et al. (2019) did not observed effects of tannins supplementation (up to 4% g/kg diet DM) on final pH and drip loss.

Along with the amount of fat (marbling), meat color is of great importance because these two characteristics

will be the first to determine consumer purchasing decisions. Changes brightness ( $L^*$ ), in  $a^*$  (redness) and  $b^*$  (yellowness) values over a period of time means a meat color deterioration from red to brown, and reflect the myoglobin concentration and its redox state in meat, rejection of acceptance is highly associated with excessively darken color of meat. In our experiment, lambs that receive tannins showed high  $L^*$  values (linear effect,  $P < 0.01$ ). The higher  $L^*$  values in meat lambs that consumed tannins may be because of high muscle glycogen levels in those animals. Blood hemoglobin level have been negative correlated with supplemental tannins, which in turn is correlated with muscle lightness (Priolo et al., 2000). Increased meat lightness in some experiments is probably a consequence of reduced muscle iron content in animals fed CT-rich diets. According to these findings, it is likely that a reduced biosynthesis of hemoglobin caused by dietary tannins could result in a lighter color in meat. According to the above, the increase on  $L^*$  value is not rare response to tannin supplementation (Luzardo et al., 2019). Even when some reports indicate no effects of supplemental condensed tannins on color of meat lamb (García et al., 2019; Valenti et al., 2019). Anyway, the effect of dietary CT on meat color of lamb have been controversial, the absence of effects of tannins on color meat could be explained by a short tannins supplementation period (i.e.,  $< 40$  days), low dose of tannins, and type of tannins supplemented since condensed tannins are less degraded or absorbed in the gastrointestinal tract than solubles (Hervás et al., 2004; dos Santos et al., 2021). The average value of shear force registered in the present experiment ( $3.6 \pm 0.57$ ) is within range for lambs finished with high grain diets (Hopkins et al., 2010). Supplemental tannin tended ( $P = 0.08$ ) to linearly increase shear force. This effect of supplemental tannins is not consistent. A decreased of meat lamb tenderness have been reported by high-tannins supplementation (dos Santos et al., 2021), but others (Bonanno et al., 2011; Sánchez et al., 2019), did not report effects of tannins on shear-force of meat. Most of inconsistencies regard to tannin supplementation on performance and meat quality could be explained by factors such as supplementation level (net intake of tannins/d), diet composition (energy concentration and associative factor of tannins with other diet ingredients), and type of tannin supplemented (condensed, hydrolysable, or they combination). Regarding the latter, Luzardo et al. (2019) tested the effects of chestnut and chestnut + quebracho as sources of supplemental tannins offered in grazing lambs supplemented with concentrates. They reported that when chestnut was include in diet not differences in shear force was detected when compared with Controls, but when chestnut was combined with quebracho shear force was numerically increased on 6.5%. Further research must be conducted to elucidate mainly the role of dose and tannins type on its effect on lamb meat quality.

**Table 1:** Treatment effects on *m. longissimus thoracis et lumborum* meat quality of hairy lambs received 0, 2, 4, or 6 g tannins/kg diet DM during 70-d.

Item	Tannin level (g/kg diet DM)				SEM	P <sup>1</sup> value	
	0	2	4	6		L	Q
Observations	10	10	10	10			
pH							
1 h	6.60	6.58	6.50	6.54	0.07	0.33	0.60
24 h	5.95	5.80	5.86	5.79	0.06	0.13	0.49
Water holding capacity, %	33.47	35.43	35.76	36.34	1.07	0.08	0.52
Drip loss, %	5.30	4.65	4.96	4.57	0.51	0.14	0.84
Color							
L	38.73b	38.41b	38.44b	41.43a	0.84	0.04	0.08
a*	20.89	20.71	21.00	21.74	0.36	0.09	0.22
b*	14.51	14.64	14.52	14.35	0.29	0.89	0.77
Cook loss, %	18.08	18.07	18.17	19.09	0.65	0.29	0.48
Shear force, kg/cm <sup>2</sup>	3.10c	3.19c	3.85b	4.26a	0.09	0.01	0.07

Rows with different literal differ ( $P < 0.05$ ); SEM, standard error of the mean; LM, *longissimus* muscle. <sup>1</sup>P = observed significance level for linear and quadratic effects of supplemental tannin.

It is concluded that, under the experimental conditions of our experiment, a long-term supplementation of tannins (70-d) may affect color and tenderness when is supplemented beyond 4 g/kg DM. The increases on shear force and  $L^*$  value was notable when tannin was supplemented at 6 g tannin/kg diet DM. Increases on lightness can be a visual advantage for the potential consumers. However, further research must be performed in order to establish the maximum dose level of supplementation with positive effects on meat quality but without detrimental effects of tannins on feed intake, nutrient utilization and health.

## NOVELTY STATEMENT

According to the style and format (and articles previously published), short communication not include this section. However, in order to accomplish with the requested the novelty statement is: The use of tannins extracts as feed additive in lambs have become popular in several regions. Tannins extract dose from, 3 up to 7 g/kg diet DM have been positive effects on meat quality of wool lambs, but in hairy finishing lambs the information about of the tannins supplementation on meat quality is very limited. The findings that are expose here shown that, in hairy lambs, the long-term supplementation may affect color and tenderness of meat lamb when is supplemented beyond 4 g/kg DM.

## AUTHOR'S CONTRIBUTION

BICP, LRR, and OCM run the experiment, collected and analyzed the samples. AB analyzed the data. AEA advice the experiment, write the first version of the manuscript.

AP designed the research work, data curation, corrected, interpreted and finalized the manuscript.

## CONFLICT OF INTEREST

The authors have declared no conflict of interest.

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