Effects of dietary beef tallow on performance, rumen fermentation, carcass traits and meat quality of growing lambs

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Abstract

Growing lambs were evaluated for the effect of increasing dietary levels of beef tallow (BT) on performance, rumen fermentation, and carcass and meat characteristics. Twenty-one 5-month-old male Rambouillet lambs were assigned randomly to one of three diets with 0, 20, and 40 g BT/kg dry matter (DM) and similar energy and protein contents. Lambs were adapted to the diets for 15 days, followed by a 45-day evaluation trial. Data were analysed using a mixed model. Growth, feed intake and ruminal fermentation were not affected by the dietary level of BT. Daily metabolizable energy intake (MEI), carcass yield and degree of fatness, increased linearly as the BT level increased. Carcass classification and muscle conformation were not affected by BT. Most meat characteristics (texture, pH, myoglobin, protein content, colour, cathepsins, and chemical composition) five and eight days post-mortem were not affected, except for fat content in meat, which increased linearly as diet BT level increased. Fatty acid (FA) profiles of the meat from lambs fed the three diets were similar. In conclusion, addition of BT at 20 and 40 g/kg DM to diets for growing lambs allowed reduced grain usage and increased forage levels (from 0 to 270 g/kg DM), increasing energy intake, carcass yield and fatness, and intramuscular fat without causing harmful effects on growth, feed intake or ruminal fermentation characteristics.

Introduction

The proportion of grain in diets for finishing lambs has increased considerably. However, empirical evidence of potentially harmful effects of grain-based diets on rumen fermentation, feed intake, growth, and meat characteristics is still limited. Interestingly, the average growth rate of fattening lambs in Latin American countries is lower compared with the levels of performance that are achieved in North America, Europe, Asia, and Oceania. Although this can be explained from environmental, cultural and economic perspectives; nutritional limitations are also evident (Pinos, 2010). Fat can be used to increase dietary energy density and to increase the absorption of fat-soluble nutrients and reduce dust from the diet (NRC, 2001). Use of fat supplementation to increase dietary energy density and improve efficiency of milk production in dairy cattle is supported in the recent review of Palmquist and Jenkins (2017) and the meta-analysis of Weld and Armentano (2017). Tallow, yellow grease and mixtures are traditionally used as common sources of fat for beef cattle (Guerrero *et al.*, 2016). For beef cattle, supplemental oils that protect from rumen biohydrogenation modify muscle FA profiles while leaving growth performance unaffected (Scollan *et al.*, 2007). Several studies show that dietary lipids from vegetable oil in sheep generally do not affect growth rate, but lambs supplemented with linseed and sunflower oil show improvements in feed efficiency (Howes *et al.*, 2014; Van Cleef *et al.*, 2016). Most studies on dairy cows, beef steers and sheep have used oilseeds or ruminally inert fats. There is extremely limited work, however, on the impact of using free oil and tallow. In sheep, two studies have evaluated the effects of replacing grains with BT on growth of finishing lambs, finding variable effects of 20 and 40 g BT/kg DM on average daily gain (ADG) and feed conversion (Ružić-Muslić *et al.*, 2009; Ahmed *et al.*, 2015). Booyens *et al.* (2012) also indicated the possibility of altering the FA

profile of meat from lambs by feeding different sources of fat. When supplemental fat replaces grains and by-products the formulation of the entire diet should be considered. This is because replacing grain with saturated free FAs can result in increased energy intake simultaneous with reduced DM intake (DMI). Alternatively, increasing forage in diets with supplemented fat potentially leads to a forage effect whereby DMI would be increased while the added fat could also increase energy density of the diet (Weiss & Pinos-Rodríguez, 2009). Therefore, the purpose of the study was to evaluate the addition of BT to high-grain diets on growth, rumen fermentation, carcass characteristics and the chemical composition and FA profile of meat from lambs.

Materials and Methods

All procedures were reviewed and approved by the Academical Committee of Facultad de Agronomía y Veterinaria of Universidad Autónoma de San Luis Potosí, in compliance with Mexican laws enacted for the use of animals in experimentation (NOM-062-ZOO-1999).

Twenty-one five-month-old Rambouillet ram lambs weighing 28.8 kg ± 2.8 kg were assigned randomly to one of the three experimental diets containing 0, 20 and 40 g BT/kg DM. Diets (Table 1) were formulated according to NRC (1985) to meet the requirements for finishing lambs between four and seven months of age and weighing 30 kg.

Lambs were housed in an open barn in individual cages (0.8 m x 1.2 m) equipped with feeder and water trough. Lambs were fed twice a day (8h00 and 16h00) and had free access to feed and fresh water for a 60-day period (15 days for adaptation to experimental diets and individual cages, and 45 days for experimental sampling). Feed samples offered and refused were collected weekly and composited. They were then dried in a forced air oven at 90 °C for 48 hours to determine DM and subsequently ground to pass through a 1 mm screen (Thomas Wiley mill, Thomas Scientific LLC, Swedesboro, NJ, USA). Crude protein (CP), starch, ether extract (EE) and ash were determined according to procedures of the AOAC (2006). Neutral detergent fibre (NDF) was assayed with a heat stable amylase and expressed inclusive of residual ash (Mertens, 2002). Determination of acid detergent fibre (ADF) was by the method of Van Soest *et al.* (1991) with using an $AMKOM_{200}$ (AMKOM Technology Co., Fairport, NT, USA). In vitro organic matter digestibility (IVOMD) was determined using the two-stage technique of Tilley and Terry (1963). The IVOMD values were used to predict digestible energy (DE) (MJ/kg) as:

$$DE \text{ (MJ/kg)} = 0.27 + 0.04289(IVOMD) \qquad \text{(Fonnesbeck } et\ al., 1984)$$

Digestible energy values were then converted to metabolizable energy (ME) using the formula:

$$ME \text{ (MJ/kg)} = 0.8219DE \qquad \text{(NRC, 2001).}$$

Bodyweights were recorded before the morning feeding on days 1, 15, 30, and 45. Feed intake was measured daily as the difference between the amounts of feed offered and refused. Ad libitum intake was assured by offering 5% more feed than was previously consumed. Average daily gain was calculated from the changes in bodyweight. Feed conversion was calculated by dividing DMI by ADG. Metabolizable energy intake was calculated with DMI x ME energy in feed.

On day 45, three hours after the morning feeding, rumen samples were obtained via an oesophagus tube, and pH was determined immediately (pH Testr37®, Double Junction, Waterproof, USA). A subsample (10 mL) of filtered ruminal fluid was mixed with 1 mL of 25% metaphosphoric acid and frozen at -20 °C. For volatile fatty acid (VFA) quantification, ruminal fluids were centrifuged at 9 500 g for 15 min, and 1 µL of supernatant was injected into a gas chromatograph (HP® 6890 Agilent, USA) according to Erwin *et al.* (1961). The concentration of ammonia-nitrogen (NH_3-N) was determined using 20 µL of supernatant plus phenol and sodium hypochlorite incubated at 38 °C for 30 min (McCullough, 1967). Absorbance (Spectrophotometer UV/VIS Agilent® 8453, USA) was recorded at 540 nm.

On day 46, after a 24-hour fast, lambs were stunned with a captive bolt device and slaughtered at Facultad de Agronomía y Veterinaria abattoir, complying with the applicable Mexican regulation (NOM-033-ZAG/ZOO-2014). Carcass weight was recorded immediately, and dressing percentage was calculated by dividing carcass weight by final bodyweight and multiplying by 100 (Zimmerman *et al.*, 2008). The degree of fatness and carcass conformation of lambs were classified according to Mexican scale (NMX-FF-106-SCFI-2006). Carcass and leg length, width of buttock and thorax, and area at 12th rib were measured as described by Ruiz de Huidobro *et al.* (2005).

On the day of slaughter, the *Longissimus thoracis et lumborum* (LTL) of the left side between the tenth thoracic vertebra and the sixth lumbar vertebra was dissected, vacuum packed and frozen at -20 °C. On days 5 and 8 after slaughter, meat quality parameters (area pH, compression, colour and chemical composition) were determined. To calculate, LTL area at the 12th rib, the circumference of the chop eye was

drawn on acetate and its area was obtained by planimetry. Immediately after thawing the LTL meat sample, the pH was measured (pH meter, Thermo-Orion 410Aplus, Torrington, CT, USA). A compression test (Lepetit & Buffiere, 1993) was carried out with a texturometer applying up to 20% strain at a speed of 50 mm/min (Instron universal texturometer, Instron 3365, Grove City, Pennsylvania, USA). Luminosity (Hunter L* value), redness (Hunter a* value), and yellowness (Hunter b* value) were read with a colorimeter (Konica Minolta On Colour CM-2500d Online, Osaka, Japan). Moisture content of the LTL samples was determined by freeze drying and their fat content was determined using a Goldfisch fat extractor (Labconco Corp., Kansas City, MO, USA). Quantities of crude protein and ash in LTL samples were determined following (AOAC, 2006) and expressed as percentage of sample weight.

Table 1 Ingredients and chemical composition of isocaloric and isonitrogenous diets used to test effects of added beef tallow

	Dietary treatments		
	0	20	40
Ingredient (g/kg DM)			
Corn, whole grain	660	520	380
Soybean meal 48% CP	120	100	90
Cane molasses	80	80	80
Alfalfa hay	50	70	90
Wheat middling		120	230
Vitamin and mineral premix[1]	90	90	90
Beef tallow		20	40
Chemical composition			
DM (g/kg)	880	888	895
CP (g/kg DM)	141	141	144
NDF (g/kg DM)	95	132	167
ADF (g/kg DM)	46	61	76
Starch (g/DM)	327	283	239
Ash	122	127	132
IVOMD (g/kg DM)	766	775	778
ME (MJ/kg DM)	12.1	12.3	12.4
EE (g/kg)	30	46	62
Fatty acids (g/100 g fat)			
C12:0 Lauric acid	0.1	0.7	1.9
C14:0 Myristic acid	1.5	2.5	3.7
C16:0 Palmitic acid	8.6	20.8	26.1
C18:0 Stearic acid	4.8	8.5	15.6
C18:1 n-9 Oleic acid	40.5	35.4	29.4
C18:2 n-6 Linoleic acid	44.4	29.6	20.5
C18:3 n-3 Linolenic acid	0.1	2.5	2.8
Saturated fatty acids (SFA)	15.0	32.5	47.3
Unsaturated fatty acids (UFA)	85.0	67.5	52.7

DM: dry matter, CP: crude protein (Nx6.25), NDF: neutral detergent fibre, ADF: acid detergent fibre, IVOMD: *in vitro* organic matter digestibility; ME: metabolizable energy, EE: ether extract; SFA: ($\sum$ C12:0, C14:0, C16:0, C17:0, C18:0); UFA: ($\sum$ C18:1 n-9, C18:2 n-6; C18:3 n-6)
[1]Ca: 180 g; NaCl: 180 g; S: 5 g; K: 5.6 g; Mg: 8 g; Zn: 50 g; Fe: 20 g; I: 0.5 g; Mn: 36 g; Co: 90 mg; Se: 90 mg; Vit. A: 3000 MIU; Vit. D_3: 750 MUI; Vit. E: 25 MUI

The FA profiles of the diets and LTL samples determined after extraction with chloroform-methanol 2: 1(v/v) and the esterified samples were analysed using by a gas chromatograph (Agilent Technologies, Santa Clara, California, USA) following Berdeaux (1999). Individual FA peaks were identified by comparison with known reference methyl esters (Supelco 37 Component FAME Mix, 47885-U, Sigma-Aldrich Co.). All FA values were expressed as a weight percentage of total FAs. In LTL samples, myoglobin, cathepsin B and cathepsin B+L concentrations were quantified as proposed by Trout (1990) and Etherington & Wardale (1982).

The experimental design was a split plot that resulted from random assignment of seven lambs to each of the three diet treatments and repeated measurements that were collected over time. Data were analysed with a MIXED procedure using SAS release 9.1.3 (SAS Institute, Inc., Cary, North Carolina, USA) with the fixed treatment effects of BT level in the diet (two degrees of freedom (df)), and residual (lambs within treatment, 18 df) as the whole plot. The subplot consisted of time (repeated, 2 df) and interaction time*treatment which were included in the model with the AR(1) covariance structure. Polynomial contrasts were used to test the linear or quadratic effects of BT dietary level on the dependent variables. In addition, Tukey's test was used to detect differences between means. For those traits which were recorded at only one point in time, the model was reduced to contain only the whole plot. Categorical data (muscle conformation, fatness degree and perirenal fat) were analysed as a completely randomized design. The CATMOD procedure of SAS release 9.1.3 (SAS Institute, Inc., Cary, North Carolina, USA) was employed with the response functions considered as cumulative logits. The model included BT levels (treatments) in the diet (fixed, two df) and individual carcass as the experimental unit (residual 18 df). A probability of less than 0.05 was considered a statistically significant difference.

Results and Discussion

As expected, as additional BT was included in the diet, the EE and saturated fatty acid (SFA) contents, mainly C16:0 and C18:0, of the diet also increased and the contents of the unsaturated fatty acids (UFA) C18:1 n-9 and C18:2 n-6 decreased (Table 1). Perhaps because the diets were formulated to be isocaloric, lambs fed 0, 20, and 40 g BT/kg DM had similar final BW, total gain, ADG, DMI and feed conversion, and ruminal fermentation characteristics (pH values, VFA molar proportion and ammonia N concentration). However, daily ME intake increased linearly ($P < 0.05$) as the BT level in the diet increased. Carcass classification, carcass weight and length, muscle conformation, leg length, width of buttock and thorax and area of the longissimus muscle at 12th rib were not affected by dietary level of BT. Carcass dressing, fatness degree and dorsal fat at the 12th rib increased linearly ($P < 0.05$) as BT level in the diet increased (Table 2). In agreement with the current results, previous studies using grain-based diets and BT at 40 g/kg DM have shown that the added fat did not affect growth performance and ruminal fermentation in finishing beef cattle (Guerrero *et al.*, 2016; Lopez *et al.*, 2016). Moreover, in finishing lambs, saturated beef tallow (30 g/kg DM) has been shown not to affect DMI (Booyens *et al.*, 2013). Steers fed diets based on steam-flaked corn and supplemented with 0 and 40 g BT/kg DM also had similar rumen pH values, VFA molar proportion and ammonia N concentrations (Montgomery *et al.*, 2008). Thus, supplemental BT might not have influenced rumen fermentation characteristics of lambs and beef cattle, although it is uncertain if the same thing happens in dairy cows which are provided a diet that is high in C18 vegetable fats which reduce the digestibility of fibre (Palmquist & Jenkins, 2017).

To the authors' knowledge, this is the first study to show that lambs fed isocaloric diets that were augmented with BT and concurrently with fibre had higher daily ME intake than lambs fed an unaugmented control diet. In dairy cows, Weiss & Pinos-Rodríguez (2009) found that replacing corn grain with saturated free FAs was correlated with an increase in energy intake with that additional energy being mostly directed to body reserves. In growing lambs, an incremental increase in fat thickness at the 12th rib was noted with a diet that included 40 g BT/kg DM (Ahmed *et al.*, 2015). Taken together, these studies suggest that including BT in the diet leads to an increase in energy intake, which probably enables available energy to be directed mostly to increased fat deposition with consequent increases in carcass dressing and carcass conformation (Table 2).

Table 2 Means of growth performance, ruminal fermentation and carcass characteristics of finishing lambs fed diets with beef tallow

	Dietary treatment - amount of beef tallow (g/kg DM)			
	0	20	40	SEM
Growth performance				
Initial BW (kg)	28.9	29.0	28.5	0.57
Final BW (kg)	44.0	44.1	44.3	1.08
Total gain (kg)	15.1	15.1	15.8	0.81
ADG (g)	335.5	335.5	351.1	9.96
DMI (g/d)	999.4	1050.1	1060.7	22.91
ME intake (MJd) [L]	12.1[b]	12.9[a]	13.1[a]	0.07
DMI/ADG	2.98	3.12	3.02	0.39
Ruminal fermentation				
pH	5.81	5.90	5.95	0.10
Ammonia N (mg/L)	142.1	138.9	145.0	7.89
Total VFA (mmol/L)	91.3	92.9	92.8	3.77
Acetate (mol/100 mol)	46.2	47.4	47.6	3.52
Propionate (mol/100 mol)	37.0	36.3	35.0	2.01
Butyrate (mol/100 mol)	16.8	16.3	17.4	1.84
Acetate: propionate ratio	1.24	1.30	1.36	0.11
Carcass characteristics				
Carcass weight (kg)	21.2	22.9	23.0	0.51
Carcass dressing (%) [L]	48.2[b]	51.9[a]	51.9[a]	0.71
Carcass length (cm)	67.8	66.4	67.9	0.48
Carcass clasification[1]	3.5	3.5	3.6	0.11
Muscle conformation[2]	3.4	3.6	3.6	0.62
Fatness degree[3][L]	3.4[b]	4.1[a]	4.2[a]	0.38
Dorsal fat at 12th rib (mm) [L]	1.1[b]	1.4[a]	1.5[a]	0.12
Leg length (cm)	44.1	43.2	43.8	0.51
Width of buttock (cm)	19.0	19.2	19.4	0.41
Width of thorax (cm)	14.4	15.0	14.6	0.28
Area at 12th rib (cm^2)	20.0	20.3	20.9	0.47

BW: bodyweight; ADG: average daily gain; DMI: dry matter intake, ME: metabolizable energy, VFA: volatile fatty acids
[1] 1 = no classification; 2= Mexico 2; 3 = Mexico 1; 4 = Mexico extra
[2] 1: Poor; 2: normal; 3: good; 4: very good; 5: excellent
[3] 1: Very lean; 2: lean; 3: rather fatty; 4: fatty; 5: very fatty
[a,b] Row means with different superscripts differ at $P < 0.05$; [L] denotes a significant ($P < 0.05$) linear effect

Except for intramuscular fat, none of the characteristics of the meat measured on either day 5 or day 8 post-mortem were affected by the addition of BT to the diet. The absence of a significant relationship between BT and meat colour may resulted from the lack of differences in pH as indicated by Fiorentini *et al.* (2015). Intramuscular fat content increased linearly ($P < 0.05$) as BT level in the diet increased (Table 3). Although Wood *et al.* (2008) found a correlation between muscle lipid content (intramuscular fat) and meat tenderness, they also did not observe an effect of dietary BT on meat texture or colour. The inconsistent effects of dietary BT on meat characteristics were noted by Nelson *et al.* (2008).

Despite the differences in the FA profiles of the current experimental diets, none of the individual FAs in the meat was affected by the dietary BT (Table 4). This was not observed in another study, where inclusion of saturated tallow (30 g/kg DM) resulted in higher lamb muscle content of palmitoleic acid (C16:1c9) and monounsaturated fatty acids (MUFAs) (Booyens *et al.*, 2012). One explanation for the lack of

effect of dietary BT on fatty meat FA profile might be that FAs originating from BT are in the free form and not bound within triacyl glycerol molecules (Siebrits *et al.*, 2009).

Table 3 Means of carcass and *Longissimus dorsi* characteristics of finishing lambs at five and eight days post-mortem

Meat characteristic	Dietary treatment - amount of beef tallow (g/kg DM)			SEM
	0	20	40	
5 days post-mortem				
Texture (N/cm^2) [1]	8.8	8.7	8.9	0.93
Lightness index (L*)	39.8	40.3	40.4	1.01
Redness index (a*)	7.5	7.8	8.1	0.71
Yellowness index (b*)	11.7	12.0	11.3	0.95
pH	5.6	5.7	5.7	0.12
Cathepsin B [2]	266	275	269	17.31
Cathepsin B+L [2]	305	314	325	18.90
Myoglobin, mg/g	0.28	0.29	0.25	0.02
Protein Lowry (mg/mL)	0.38	0.39	0.42	0.10
Moisture (g/kg)	784	779	781	9.41
Crude protein (g/kg)	180	176	178	5.05
Fat (g/kg) [L]	47.1[b]	49.0[ab]	49.8[a]	0.95
Ash (g/kg)	36.4	34.9	35.6	2.11
8 days post-mortem				
Texture (N/cm^2) [1]	1.9	1.5	1.6	0.43
Lightness index (L*)	42.5	43.9	43.4	1.02
Redness index (a*)	7.0	6.8	6.5	0.83
Yellowness index (b*)	12.1	12.0	12.0	1.02
pH	5.7	5.5	5.5	0.11
Cathepsin B [2,]	321	322	324	16.31
Cathepsin B+L [2]	344	349	352	15.60
Myoglobin (mg/g)	0.31	0.30	0.28	0.02
Protein Lowry (mg/mL)	0.36	0.39	0.40	0.22
Moisture (g/kg)	784	769	777	9.41
Crude protein (g/kg)	180	172	175	5.89
Fat (g/kg) [L]	47.0[b]	48.3[ab]	49.7[a]	0.92
Ash (g/kg)	36.4	34.9	35.6	2.61

[1] Compression test at 20% of total compression
[2] Expressed as specific activity in nmol of NMec (amino-methylcoumarin) released per min/mg protein
[a,b] Row means with different superscripts differ at *P* <0.05; [L] and [Q] denote the significant (*P* <0.05) linear and quadratic effects, respectively

Table 4 Effect of tallow supplementation on fatty acid profile in *Longissimus dorsi* of finishing lambs

Fatty acid (g/100 g)	Dietary treatment - amount of beef tallow (g/kg DM)			SEM
	0	20	40	
C10:0, Capric acid	0.27	0.23	0.25	0.03
C12:0, Lauric acid	0.29	0.25	0.23	0.04
C14:0, Myristic acid	4.71	4.89	4.91	0.06
C15:0, Pentadecanoic acid	0.85	0.64	0.65	0.05
C16:0, Palmitic acid	27.20	27.11	26.40	0.61
C16:1, Palmitoleic acid	2.35	2.01	2.09	0.29
C17:0, Margaric acid	1.22	1.12	1.11	0.17
C17:1 c9 fatty acid	0.79	0.84	0.82	0.09
C18:0, Stearic acid	16.90	17.30	18.90	1.03
C18:1 t9, Elaidic acid	1.94	2.28	1.99	0.79
C18:1 c9, Oleic acid	37.90	37.80	37.20	1.59
C18:2 n-6, Linoleic acid	4.15	3.92	3.93	0.99
C18:3 n-3, Linolenic acid	0.42	0.46	0.42	0.11
C18:2 CLA cis9-trans11, Conjugated linoleic acid	0.45	0.51	0.49	0.09
C20:4 n-6, Arachidonic acid	0.56	0.65	0.61	0.09
SFA	51.40	51.50	52.40	1.99
MUFA	42.90	42.90	42.10	2.76
PUFA	5.58	5.54	5.45	1.28

SEM: standard error of means; SFA: saturated fatty acids ($\sum$ C10:0, C1:0, C14:0, C15:0, C16:0, C18:0, C20:0); MUFA: unsaturated fatty acid ($\sum$ C16:1, C17:1 c9, C18:1 t9, C18:1 c9); PUFA, polyunsaturated fatty acid ($\sum$C18:2 n-6, C18:3 n-3, C18:2 CLA cis9-trans11, C20:4 n-6).

Conclusion

Inclusion of BT (40 g BT/kg DM) as energy source, fibre supplement and grain replacement in finishing lamb diets correlated linearly with an increase in energy intake, carcass dressing and fatness, and muscle fat content, while DMI, growth performance and rumen fermentation were unaffected. In addition, BT does not have noticeable effects on texture, meat colour or FA profile. Therefore, dietary BT for finished lambs may be an option to minimize grains in diets without changing growth performance or meat traits.

Authors' Contributions

SLA and ADC conducted the experiment; JMPR conceived, designed and supervised the research; IADM evaluated the carcasses; JGV and HLR analysed the feed and meat samples.
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Conflict of Interest Declaration

The authors have declared that no competing interests exist

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