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Carcass characteristics of feedlot lambs fed crude glycerin contaminated with high concentrations of crude fat

J.F. Lage [a,*], P.V.R. Paulino [b], L.G.R. Pereira [c], M.S. Duarte [b], S.C.Valadares Filho [b], A.S. Oliveira [d], N.K.P. Souza [b], J.C.M. Lima [b]

[a] Departamento de Zootecnia da Universidade Estadual Paulista Julio de Mesquita Filho (UNESP), Campus Jaboticabal, CEP: 14884-900, Jaboticabal, SP, Brazil
[b] Departamento de Zootecnia da Universidade Federal de Viçosa (UFV), Campus Universitário, CEP: 36571-000, Viçosa, MG, Brazil
[c] Embrapa Gado de Leite, CEP: 36038-330, Juiz de Fora, MG, Brazil
[d] Departamento de Zootecnia da Universidade Federal do Mato Grosso, Campus Sinop, CEP: 78557-267, Sinop, MT, Brazil

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ABSTRACT

Thirty non-castrated male lambs with 20 ± 2.3 kg average body weight (BW) were randomly assigned to five treatments consisted of different dietary concentrations of crude glycerin (CG; 0, 3, 6, 9 and 12% on DM basis) to evaluate the effects on performance, carcass and meat quality traits. A quadratic effect was observed for performance ($P = 0.04$), final BW ($P < 0.01$) and hot carcass weight ($P < 0.01$). No effects of CG were observed ($P > 0.05$) on carcass pH neither on shear-force, cooking loss and ether extract content in longissimus. The inclusion of CG tended to reduce the Zn content in meat ($P = 0.09$). The data suggests that CG (36.2% of glycerol and 46.5% of crude fat) may be used in diets of finishing lambs with concentrations up to 3% without negative effects on performance and main carcass traits. Moreover, inclusion of CG seems to not affect quality and safety of meat for human consumption.

1. Introduction

The increase of biodiesel production has led to increased stocks of glycerol with a subsequent price reduction, making glycerol a potential high energy feed source for ruminants (Avila et al., 2011). The inclusion of glycerol in ruminant diets modifies the ruminal acetate: propionate ratio due to the increase of propionate levels (Drouillard, 2008). Therefore, glycerin may partially replace starch-based ingredients, in the diet because glycerol (a constituent of crude glycerin) is converted to propionate in the rumen and acts as a precursor for hepatic glucose synthesis. Consequently, the inclusion of crude glycerin in ruminant diets may increase marbling since glucose is used as a source of carbon for fatty acid synthesis (Schoonmaker, Fluharty, & Loerch, 2004).

Recent efforts have evaluated the effects of the inclusion of crude glycerin (above 88% of glycerol) in diets on intake, performance, carcass and meat quality traits of sheep reporting acceptable inclusion of 15%, 21% and 12% respectively on diet dry matter (Avila-Stagno et al., 2013; Gunn, Neary, Lemenager, & Lake, 2010; Meale, Chaves, Ding, Bush, & McAllister, 2013). However, a limit number of studies have evaluated the effects of crude glycerin contaminated with high crude fat contents in diets fed to sheep.

We hypothesized that crude glycerin containing 36.2% of glycerol and 46.5% of crude fat in dry matter (DM) basis may be used as an energy source in diet of feedlot finishing lambs at concentrations up to 12% on DM basis without compromising animal performance, carcass and meat quality traits. Thus, the objectives of this study were to evaluate the effects of crude glycerin (containing 36.2% of glycerol and 46.5% of crude fat in DM basis) on feed efficiency, average gain daily, dressing percentage, commercial cut yield, meat quality and concentrations of heavy metals in meat and liver from lambs fed in feedlot.

2. Material and methods

2.1. Animals, diets and experimental design

This experiment was carried out at the Animal Laboratory of the Department of Animal Science at the Federal University of Viçosa (*Universidade Federal de Viçosa*-UFV) in Viçosa-MG, Brazil. Experimental protocols were approved by the Institutional Animal Care and Use Committee of the Federal University of Viçosa.

Thirty weaned Santa Ines non-castrated male lambs, weaned at 3 months of age with 20 ± 2.3 kg of average initial body weight (BWi) were used. The animals were confined in individual pens with feeders and water suppliers. Initially, lambs were submitted to a period of 10 days of adaptation to the experimental diets. After the adaptation period, animals were randomly assigned to one of

* Corresponding author. Tel.: +55 16 8111 1888, +1 402 705 9517.
E-mail address: josilage@gmail.com (J.F. Lage).

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the five experimental treatments, which consisted of five concentrations of crude glycerin (0, 3, 6, 9 and 12% of DM) as a substitute of ground corn, with six replicates per treatment. Experimental diets were composed of 30% of corn silage and 70% of concentrate (DM basis). Diets were isonitrogenous (18% crude protein on a DM basis) in order to supply the nutritional requirements of a lamb with 10 to 30 kg of BW with the growth potential of 250 g/day (NRC, 2007). Diets were not isoenergetic because crude glycerin was contaminated with higher concentration of crude fat (46.5%). A mixture of urea and ammonium sulfate was used to adjust the crude protein content (CP) of the diets because of the differences in CP content of the protein sources used. Composition of ingredients and the experimental are presented in Table 1.

The crude glycerin (CG) used in this study originated from vegetable oils of castor bean, soybean, cottonseed and sunflower. It was produced by methylic route and obtained from *Brasil Ecodiesel Indústria e Comércio de Biocombustíveis e Óleos Vegetais S.A, Iraquara-BA, Brazil.* The composition of the CG is presented in Table 2. Lambs were fed twice a day at 8 a.m. and 4 p.m. Amounts of feed offered to animals were calculated according to previous dry matter intake (DMI) and adjustments were made when needed so that refused feed did not exceed 10% of daily intake. Ort daily weights and samplings were performed for the diet quantities provided from each animal. To estimate intake, samples of orts from each animal were first composited weekly and then composited every 14 days. Feed samples were frozen at −18 °C for later analysis. All diet samples were analyzed for DM, organic matter, and ether extract as described by the Association of Official Analytical Chemists (AOAC, 1990). For analyzing the neutral detergent fiber (NDF) concentration, the samples were treated with alpha thermostable amylase without sodium sulfite and corrected for ash residue (Mertens, 2002) and residual nitrogen compounds (Licitra, Hernandez, & Van Soest, 1996). The non-fiber carbohydrate content was calculated as proposed by Hall (2000).

At the beginning of the trial all the animals were weighed after a 16-h solid fast before the first feeding in the morning. Average daily gain (ADG) was obtained by weighing the animal at the beginning and the end of the experiment, always after a 16-h solid fast.

2.2. Slaughter, carcass trait data collection and sampling procedures

After 56 days of feeding lambs were slaughtered by cerebral concussion followed by jugular and carotid venesection. Pre-harvest handling was in accordance with good animal welfare practices, and slaughtering procedures followed the Sanitary and Industrial Inspection Regulation for Animal Origin Products (Brasil, 1997). After the slaughter, the initial pH (pH_i) was measured on the left carcass of each animal using a pH-meter with automatic endpoint and buffer recognition as well as temperature compensation equipped with a penetrating electrode (Model SG2 — ELK, Seven Go™, Mettler Toledo International Inc.). The pH-meter was calibrated before use to pH 7.0 and 4.01. The pH was measured at approximately 4 cm deep on longissimus muscle of the left side of each carcass (12th rib). The carcass was split into two identical longitudinal halves and weighed to determine the hot carcass weight (HCW). Liver were weighed and samples of each animal (150 g) were collected for further heavy metal analysis. All carcasses were refrigerated at 4 °C for approximately 24 h and then taken from the cooling chambers and weighed again in order to determine the cold carcass weight (CCW). The difference between the chilled carcass weight and HCW was used to calculate carcass shrink loss (CSL). Carcass yield was calculated using hot carcass weight (HCW) and cold carcass weight (CCW) divided by empty body weight (EBW) and shrunk body weight (SBW) and then multiplying the result by 100.

After 24 h postmortem chill, final pH (pHf), 12th rib fat thickness (RFT) and 12th rib longissimus muscle area (LMA) were measured on the left side of each carcass. The final pH was also measured at approximately 4 cm deep on longissimus muscle of the left side of each carcass (12th rib). Longissimus muscle area was traced on transparencies and measured later with a planimeter and RFT measurements were taken 3/4 the length ventrally over the longissimus muscle by using a digital paquimeter (Greiner, Rouse, Wilson, Cundiff, & Wheeler, 2003). In the right half of each carcass commercial cut yield was measured by separating the carcass half into neck, shoulder, rib, loin and leg.

Longissimus muscle was completely removed from the left side of each carcass and standardized into three 2.54 cm thick steak samples for the Warner–Bratzler shear force (WBSF; AMSA, 1995). Steak thawing and cooking losses were measured in the same steak samples used for WBSF evaluation. For chemical composition and heavy metal evaluation a 200 g of longissimus muscle sample was also taken from the left half of each carcass. All longissimus muscle samples were vacuum-packaged and kept at −20 °C for 10 days until the analysis was performed.

2.3. Warner–Bratzler shear force measurement

Warner–Bratzler shear force steaks were thawed at 4 °C for a period of 24 h and oven broiled in an electric oven pre-heated to 150 °C. Internal steak temperature was monitored with 20-guage copper-constantan thermocouples (Omega Engineering, Stamford, CT) placed in the approximate geometric center of each steak and attached to a digital monitor. When internal steak temperature reached 35 °C, it was turned over and allowed to reach an internal temperature of 70 °C before removal from the oven. Cooked WBSF steaks were cooled for 24 h at 4 °C (AMSA, 1995). Two round cores

Table 1
Ingredient proportion and chemical composition of the experimental diets.

Item	Concentration of crude glycerin, % DM				
	0	3	6	9	12
Ingredients proportion, % DM					
Corn silage	30.0	30.0	30.0	30.0	30.0
Corn	43.7	40.6	37.5	34.4	31.3
Soybean meal	22.5	22.5	22.5	22.5	22.5
Crude glycerin	–	3.0	6.0	9.0	12.0
Urea/ammonium sulfate	0.5	0.6	0.7	0.8	0.9
Commercial premix[a]	3.3	3.3	3.3	3.3	3.3
Chemical composition, % DM					
Dry matter	70.8	71.1	71.4	71.6	71.9
Organic matter	96.2	96.2	96.1	96.1	96.1
Crude protein	18.8	18.8	18.9	18.9	18.9
NDFap	21.5	21.1	20.7	20.3	20.0
Ether extract	3.0	4.3	5.6	6.9	8.2
Non-fiber carbohydrate	53.7	52.9	52.0	51.2	50.4
Total digestible nutrients	78.9	75.9	79.2	80.4	81.3

[a] Composition (per kg): Ca, 110 g; P, 65 g; Na, 185 g; Cl, 300 g; Mg, 20 g; S, 20 g; Mn, 4660 mg; Zn, 4750 mg; Co, 120 mg; I, 72 mg; Se, 35 mg.

Table 2
Physical–chemical composition of crude glycerin included in the experimental diets.

Item	Value
Glycerol	36.2
Methanol	8.7
Total fatty acids	46.5
Water	6.2
Crude protein	0.4
Ash	2.0
Density, g/cm^3	0.9
Heavy metals (mg/kg)	
Cu	5.6
Cr	1.2
Ni	5.9
Pb	0.3
Zn	5.2
Cd	Level lower than detection limit (0.05 mg/L)

(1.27 cm diameter) were removed from each steak, with six round cores for each animal (three 2.54 cm thick steaks per animal and two round cores per steak), parallel to the long axis of the muscle fibers (AMSA, 1995). Each core was sheared once through the center, perpendicular to the fiber direction by a Warner–Bratzler shear machine (G-R Manufacturing Company, Manhattan, KS — USA).

2.4. Steak thawing and cooking loss

Steak thawing and cooking loss were evaluated on steaks also used for WBSF measurement. For thawing loss evaluation, each steak was weighed frozen and after a period of 24 h thawing at 4 °C. The cooking loss of each steak was recorded after steaks were oven-broiled during the WBSF processing. Cooking loss was calculated as the difference between the weight of the steaks before and after oven-broiling. The total cooking loss minus drip loss represented the evaporative loss. The total liquid loss was calculated by the difference between the weight of frozen and cooked steak.

2.5. Chemical composition and heavy metal analysis

Samples used for chemical analysis were lyophilizated for 72 h. All samples were then ground using a ball mill and analyzed for moisture, protein (Method 920.87; AOAC, 1990), ether extract (EE; Method 920.85; AOAC, 1990), and ash (Method 924.05; AOAC, 1990) in order to determine the chemical composition of each longissimus sample.

Heavy metal analysis of CG samples was evaluated by inductively coupled plasma optical emission spectroscopy (ICP-OES; Perkin Elmer, model Optima 2000DV), in axial configuration, with 1400 kW of radiofrequency, and a gas flow rate of 0.60 L/min for meat and liver samples; concentrations of Cd, Cu, Cr, Ni, Pb and Zn were measured by inductively coupled plasma atomic emission spectroscopy (ICP-AES; Perkin Elmer, model Optima 3300DV). The detection limits (3σ) obtained for the elements were (μg/L): Cd = 5.5; Cr = 6.6; Cu = 7.0; Ni = 16.5; Pb = 60.1; and Zn = 3.1.

2.6. Statistical analysis

Data were analyzed as a completely random design using the SAS (SAS Institute, Inc.) software. The GLM procedure was used to analyze the fixed effects of treatment on intake, performance, carcass and meat quality traits, with animal serving as the experimental unit. Initial body weight was used as a covariant and orthogonal contrasts were used to determine linear and quadratic responses to increasing concentrations of crude glycerin incorporation. For all the variables, *P*-values less than or equal to 0.05 were declared significant and values less than or equal to 0.10 were considered tendencies.

3. Results and discussion

Dry matter intake (g/day) decreased linearly ($P < 0.01$) with the increase of crude glycerin (CG) concentration in the diets, which is supported by previous studies (Gunn, Schultz, et al., 2010; Parsons, Shelor, & Drouillard, 2009; Ramos & Kerley, 2012). Most of the studies regarding the evaluation of inclusion of CG in diets of ruminant animals (Avila-Stagno et al., 2013; Mach, Bach, & Devant, 2009) have used CG with concentrations of fatty acids and methanol lower than 1%. In the present study CG had 46.5% of lipids and 8.7% of methanol. However, the high-risk to health associated to methanol consumption due to inclusion of CG in diets of ruminant animals is not expected since methanol is naturally produced in the ruminal environment as a result of pectin digestion (Pol & Demeyer, 1988). These authors have demonstrated that a continuous infusion of methanol (1 mol L^{-1}) at a rate of 10 mL h^{-1} into the rumen of ovine was completely converted to methane.

The greater concentration of lipid in diets of animals fed diets with the higher concentration of CG was likely to be the main factor that contributed for a reduction of dry matter intake. Ruminant animals are relatively intolerant to high concentrations of fat and feed intake usually decrease as fat content of the diets exceeds 6% DM basis (Palmquist & Jenkins, 1980). It was observed that ether extract (EE) content ranged from 6.9 to 8.2% in diets containing concentration of CG higher than 6%. Lipids represent a potential stimulator of cholecystokinin (Liddle, Goldfine, Rosen, Taplitz, & Williams, 1985) which is an appetite suppressant through the gastric emptying inhibition (Moran & McHugh, 1982). High lipid diets increase plasma levels of cholecystokinin while the decrease of passage rate of the digesta increases the reticulum-rumen distention leading to a stimulation of cholecystokinin receptors in these gastrointestinal compartments (Allen, 2000) consequently reducing appetite. According to Czerkawski (1972) the fatty acids may reduce the nutrient digestibility in the ruminal environment and thus, reduce the dry matter intake. Jenkins (1993) reported that inclusion of lipids in ruminant diets could negatively affect the neutral detergent fiber (NDF) digestibility and animal performance. Moreover, glycerol supplementation affects negatively the digestion of the more fibrous fraction of the feed (Krueger et al., 2010; Schroder & Sudekum, 1999). Thus, the association of crude glycerin with higher content of crude fat in diets decreased the DMI by the animals.

The final body weight (BWf; $P < 0.01$) and average daily gain ($P = 0.04$) have a quadratic effect with the increase of CG concentration in the diets ($P = 0.05$); therefore, no differences were observed ($P = 0.25$) for gain:feed ratio (G:F; Table 3). Animals fed higher concentrations of CG had lower DMI, thus showed lower average daily gain. Similar results were observed by Avila-Stagno et al. (2013) who reported that the inclusion of glycerol in the diets tended to reduce ADG and final body weights of lambs, but did not affect the G:F.

A quadratic effect was observed for HCW ($P < 0.01$) and CCW ($P < 0.01$; Table 4). The growth rate of the animal is highly affected by the diet composition which influences carcass tissue deposition. Thus, lambs that were fed diets with higher concentrations of CG had lower BW ($P < 0.01$) and consequently lighter carcasses.

There was a negative linear effect of CG concentrations on hot carcass yield in relation to shrunk body weight (HYSBW; $P = 0.04$) and cold carcass yield in relation to shrunk body weight (CCYSBW; $P = 0.04$). There was no effect of CG concentrations in hot carcass yield in relation to empty body weight (HYEBW; $P = 0.26$) and cold carcass yield in relation to empty body weight (CCYEBW; $P = 0.20$). Carcass yield is directly affected by carcass weight (Owens & Gardner, 2000). Thus, animals fed diets with higher concentrations of CG had lighter carcasses and consequently lower carcass yield in relation to shrunk body weight. On the other hand, diets with low fat levels may increase the dressing percentage, quality and yield grade of the

Table 3
Growth performance of lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
DMI, g[b]	1121	1115	899	942	783	0.03	<0.01	<0.01	0.73
ADG, g[c]	268	300	255	257	193	0.01	0.04	<0.01	0.05
G:F, g/kg[d]	238	267	286	275	258	0.01	0.25	ns	ns
Final BW, kg[e]	33.7	35.0	33.0	32.4	29.5	0.59	<0.01	<0.01	0.03

[a] Contrasts: L and Q — linear and quadratic effects.
[b] Dry matter intake.
[c] Average daily gain.
[d] Gain:feed ratio (feed efficiency).
[e] Final body weight.

Table 4
Carcass characteristics of lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
HCW, kg[b]	15.2	16.0	14.5	14.3	12.7	0.33	<0.01	<0.01	0.02
CCW, kg[c]	14.8	15.5	14.0	13.8	12.3	0.32	<0.01	<0.01	0.02
HYSBW, %[d]	45.2	45.6	44.0	44.0	42.8	0.33	0.04	<0.01	0.48
HYEBW, %[e]	52.1	52.5	51.8	51.3	50.6	0.30	0.26	ns	ns
CCYSBW, %[f]	43.9	44.2	42.6	42.5	41.4	0.34	0.04	<0.01	0.53
CCYEBW, %[g]	50.5	50.9	50.1	49.6	49.0	0.29	0.20	ns	ns
CSL, %[h]	3.0	3.1	3.2	3.4	3.2	0.07	0.49	ns	ns
RFT, mm[i]	1.0	0.8	0.8	1.1	0.8	0.06	0.33	ns	ns
LMA, cm[b,j]	11.5	14.6	12.1	11.9	10.2	0.44	0.02	0.04	0.02

[a] Contrasts: L and Q — linear and quadratic effects.
[b] Hot carcass weight.
[c] Cold carcass weight.
[d] Hot carcass yield shrunk body weight.
[e] Hot carcass yield empty body weight.
[f] Cold carcass yield shrunk body weight.
[g] Cold carcass yield empty body weight.
[h] Carcass shrunk loss.
[i] Rib fat thickness.
[j] Longissimus muscle area.

Table 5
Commercial cut weight and yield of lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
Commercial cuts, kg									
Neck	0.7	0.7	0.6	0.7	0.6	0.02	0.08	0.10	0.31
Shoulder	1.4	1.4	1.3	1.3	1.2	0.03	<0.01	<0.01	0.26
Rib	1.9	2.1	1.8	1.8	1.6	0.05	<0.01	<0.01	0.22
Loin	1.0	1.1	1.0	1.0	0.8	0.03	0.01	<0.01	0.04
Leg	2.2	2.3	2.1	2.0	1.8	0.05	<0.01	<0.01	0.04
Commercial cut yield, %[b]									
Neck	9.4	9.9	9.1	10.6	9.7	0.25	0.49	ns	ns
Shoulder	19.9	18.9	19.7	19.9	20.2	0.18	0.17	ns	ns
Rib	26.7	27.2	26.1	26.0	26.5	0.27	0.66	ns	ns
Loin	14.0	14.0	14.8	14.4	13.9	0.22	0.73	ns	ns
Leg	29.9	29.9	30.3	29.1	29.7	0.26	0.77	ns	ns

[a] Contrasts: L and Q — linear and quadratic effects.
[b] Percentage of cold carcass weight.

carcass while the high fat content diets tend to decrease these carcass traits (Owens & Gardner, 2000) as observed in the present study.

No effects of CG concentrations were observed on carcass shrink loss ($P = 0.49$) and 12th rib fat thickness ($P = 0.33$), which presented average values of 3.2% and 0.9 mm respectively (Table 4). Carcass shrink loss (CSL) is influenced both by the air speed in chilling, and especially by carcass fat thickness, which is also considered an important parameter in evaluating the final product. Generally, carcasses with high values of CSL also present low values of RFT and less juicy and tender meat. Nonetheless, RFT was not affected by the inclusion of CG and consequently, no differences were observed for CSL among the experimental treatments. Quadratic effect was observed for LMA ($P = 0.02$). According to Sainz (1996), LMA can accurately estimate muscular development of animal, being the final BW at the slaughter is the main factor that affects LMA. As observed in this study, lambs fed with diets containing higher concentrations of CG displayed lower BW at slaughter ($P < 0.01$) which might explain the results observed for LMA. Moreover, fat supplementation with levels up to 2% of the total diet or 4% total dietary fat seems to maximize longissimus muscle area (Owens & Gardner, 2000). This might explain the greater LMA of lambs fed diets with 3% of CG as this diet contained a total of 4.3% of EE (Table 1).

Commercial cut results are presented in Table 5. Increasing crude glycerin in the diet tended to reduce the weight of the neck ($P = 0.08$). However, a negative linear effect was observed on the weight of the shoulder ($P < 0.01$) and rib ($P < 0.01$) and a quadratic effect on the weight of loin ($P = 0.01$) and leg ($P < 0.01$; Table 5). Commercial cut weights are directly related to the carcass weight which might explain the results observed in this study since animals fed diets with higher concentrations of CG had lower weight of carcass and consequently presented lower weights of shoulder, loin and leg. Commercial cut yields represent the distribution of tissues in different regions of the carcass. Thus, the inclusion of 12% CG did not cause any changes in the distribution of the components of the carcass and any commercial cut yield (Table 5).

The inclusion of CG in diets did not affect the carcass pHi ($P = 0.43$) and pHf ($P = 0.33$; Table 6). According to Duarte et al. (2011) in unstressed animals the values of muscle ultimate pH usually range from 5.5 to 5.8. The average pH value observed in this study (6.0) is higher than normally recommended although there was no evidence of stress in the animals during the pre-slaughter. According to Immonen, Ruusunen, and Puolanne (2000) and Velasco, Cañeque, Lauzurica, Pérez, and Huidobro (2004) the value of pH is higher when animals showed carcass with lower rib fat thickness. The subcutaneous fat acts as a thermal insulator of the carcass during chilling, thus reducing the chilling rate of the carcass, allowing a normal pH decline. In this present study, animals showed an average rib fat thickness value of 0.9 mm which was not enough to allow a normal pH decline.

According to Freund et al. (1995) glycerol increases fluid retention through the reduction of free water in the organism. The effect on inclusion of glycerol in diet on muscle water retention was also reported by Parker, Dobson, and Fitzpatrick (2007), who worked with beef cattle that received glycerol during the transportation and observed that glycerol leads to a hyperhydration of the animal which implies a better quality of meat. As such, we have hypothesized that the inclusion of CG in the diets would reduce steak thawing loss as the dietary levels of CG increases. However, no differences were observed ($P = 0.11$) for thawing loss of meat among lambs fed different levels of CG in the diet (Table 6).

No differences were observed for cooking loss ($P = 0.42$), total loss ($P = 0.18$) and for WBSF ($P = 0.42$) among the experimental treatments. In this study the lamb meat showed acceptable tenderness (4.4 kgf) since according to Shackelford, Morgan, Cross, and Savell (1991) the WBSF values up to 4.5 kgf characterize a tender meat.

Concentrations of CG evaluated did not affect the concentration of ether extract ($P = 0.90$) in longissimus muscle (Table 7). In the present study, it was expected that the inclusion of CG in diets would decrease the acetate:propionate ratio in the rumen, possibly due to increases in propionate, which is the precursor of glucose. Consequently, it was also expected that the inclusion of CG would increase the intramuscular fat content since according to Schoonmaker et al. (2004) glucose is the main carbon source for deposition of fat tissue. The absence of an improvement in intramuscular fat accretion, despite overwhelming evidence of increased propionate synthesis with glycerin supplementation, seemingly refutes this belief, because the feeding glycerin may result in a suppression of intramuscular fat accretion (Drouillard, 2012) or tend to decrease deposition of intramuscular fat within the longissimus muscle (Elam, Eng, Bechtel, Harris, & Crocker, 2008).

Crude glycerin did not affect the values of moisture ($P = 0.29$) or ash contents ($P = 0.13$) of the longissimus muscle (Table 7). However, crude protein (CP) tended to reduce linearly ($P = 0.10$) with the increase of dietary levels of CG. The intake of glycerol by cattle during pre-slaughter decreases the energy deficit caused by the increase of the insulin concentration in plasma, which allows lower protein degradation in muscle and consequently preserves the carcass protein content after slaughter (Parker et al., 2007). The meat from lambs fed higher concentrations of CG presented lower values of CP. This

Table 6
Meat quality from lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
Thawing loss, %	9.1	10.5	11.2	9.4	8.0	0.42	0.11	ns	ns
Cook loss, %	18.2	20.5	20.1	16.7	20.3	0.73	0.42	ns	ns
Total loss, %	25.9	29.0	29.1	24.3	27.0	0.72	0.18	ns	ns
WBSF, N[b]	45.1	50.0	45.1	37.3	39.2	0.23	0.42	ns	ns
pH initial	6.6	6.5	6.7	6.6	6.5	0.04	0.43	ns	ns
pH final	6.0	6.1	6.0	6.1	6.0	0.02	0.33	ns	ns

[a] Contrasts: L and Q — linear and quadratic effects.
[b] WBSF: Warner–Bratzler shear force.

is most likely due to lower DMI that affected the performance and consequently lean tissue deposition.

The presence of Pb, Cd, Cr and Ni was not detected by the analytical method used on any of the samples analyzed. In this case, the concentration of those elements was lower than the detection limit of the analytical method used, which was (mg/kg): Cg = 0.5; Cr = 0.6; Ni = 1.6; and Pb = 6.0. No differences were observed ($P = 0.24$) among treatments with different concentrations of CG for the concentration of Zn in the liver (Table 8). However, the concentration of Zn in meat tended to decreased ($P = 0.09$) as the inclusion of CG in diets. Concentrations of Cu tended to increase ($P = 0.07$) in the liver of animals fed diets with CG while no differences were observed ($P = 0.43$) in concentrations of this element in meat among treatments with different concentrations of CG ($P = 0.25$).

The higher concentrations ($P = 0.04$) of Zn in meat of lambs fed diets without CG can be explained by a greater DMI and consequently a greater intake of mineral mixture present in the ration. Thus, the intake of Zn which was a component of mineral mixture of the diets was greater in lambs fed diets without CG, which allowed a greater deposition of this element in meat from those animals. Liver plays an important role in the metabolization of Cu, being responsible for the retention of up to 90% of Cu present in the organism (Hidiroglou & Ivan, 1993) which is a possible explanation for the low concentrations of Cu in meat observed in this study. Considering a daily consumption of 100 g of liver or lamb meat by humans, it can be inferred that concentrations of Zn and Cu are within the tolerable intake limit to avoid causing toxicity in humans.

Table 7
Chemical composition of longissimus muscle from lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
Moisture, %	74.5	74.2	74.6	74.5	74.9	0.09	0.29	ns	ns
Ashes, %	1.1	1.1	1.3	1.3	1.2	0.03	0.13	ns	ns
Crude protein, %	19.1	19.2	18.9	18.9	18.4	0.10	0.10	0.02	0.19
Ether extract, %	2.5	2.5	2.3	2.5	2.4	0.08	0.90	ns	ns

[a] Contrasts: L and Q — linear and quadratic effects.

Table 8
Levels of heavy metals in liver and longissimus muscle from lambs fed for 56 days with diets increasing in the concentration of crude glycerin contaminated with crude fat.

Item	Concentrations of crude glycerin, % DM					SEM	*P*-value		
							Treatment	Contrast[a]	
	0	3	6	9	12			L	Q
Liver, mg/kg									
Zn	34.7	39.3	32.3	37.3	34.1	1.06	0.24	ns	ns
Cu	58.2	62.1	72.9	55.4	92.7	4.73	0.07	0.04	0.29
Longissimus, mg/kg									
Zn	31.8	28.0	26.6	29.3	26.0	0.77	0.09	0.04	0.39
Cu	2.3	2.7	2.2	2.4	2.2	0.07	0.25	ns	ns

[a] Contrasts: L and Q — linear and quadratic effects.

4. Conclusions

The inclusion of crude glycerin (36.2% of glycerol) with higher concentration of crude fat and methanol, replacing corn in the diets of lambs does not affect negatively the safety of meat and liver for human consumption. Moreover, it had no effects on shear force and ether extract content on longissimus muscle, but performance and main carcass characteristics such as weight and longissimus muscle area are improved when lambs are fed up to 3% DM basis, suggesting that inclusion crude glycerin with higher crude fat would not exceed this concentration in diets for lambs finished in feedlot.

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