



Grain grinding size of cereals in complete pelleted diets for growing lambs: Effects on animal performance, carcass and meat quality traits

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ABSTRACT

The main goal of the present study was to clarify the effects of different grinding particle size of grains (2-mm vs. 6-mm) included in complete pelleted diets (CPD) for fattening lambs on animal performance, carcass and meat quality. Twenty male merino lambs (14.8 kg; $n = 10$ per group) were fed the corresponding diet ad libitum and slaughtered when they reached 27 kg. No differences were observed in the feed conversion ratio or carcass characteristics. However, lambs fed coarser diets (6 mm) were more efficient with less residual feed intake (-14.0 vs. 15.4 g DM/animal/d; $P < .05$) than lambs fed the 2 mm CPD. Lambs fed the 6-mm CPD showed higher levels of intramuscular fat and saturated fatty acids. Consequently, increasing the particle size of the grains included in CPD allows for improving feed efficiency and intramuscular fat in fattening lambs.

1. Introduction

Complete pelleted diets (CPD) simplify feedlot feeding management of fattening lambs in Mediterranean countries when compared to the traditional ad libitum supply of concentrates and cereal straw in separate feeding troughs. In order to avoid selective ingestion of certain ingredients and ensure enough fibre consumption, granulation of CPD in pellets has been deemed as necessary when CPD are to be fed to ruminants. It has been demonstrated that CPD result in similar feeding efficiency when compared to the traditional (Blanco et al., 2015). It must be considered, however, that CPD manufacturing requires cutting or grinding of all ingredients included in the formulation, so that most particles in the processed feed are of a variable and small size (Waldo, Smith, Cox, Weinland, & Lucas, 1971).

This is very important because ruminal digestion and fermentation processes can be impaired if feed is not sufficiently chewed and digesta comminuted through rumination and a minimum retention time of digesta in the forestomachs. In fact, mastication and rumination stimulate the production of saliva which buffers the rumen pH (thus minimising risk of ruminal acidosis) mainly due to its bicarbonate content (Grant, Colenbrander, & Mertens, 1990). Thus, feeding finely ground diets may be a hazard to reach these goals. Indeed, incidence of ruminal acidosis in fattening lambs is similar between CPD and with traditional separate feeding of concentrates and cereal straw (Blanco

et al., 2014). A possible alternative to enhance ruminal digestion of CPD would be to modify the cereal particle size (grinding size). A coarser grinding size of cereals incorporated to CPD would allow for longer residence times of digesta and promote a more favourable fermentation of starch (Blanco et al., 2014).

Although some previous studies have dealt with the effect of grain processing on ruminal fermentation and feed intake and digestibility (Andrés et al., 2018; Gimeno et al., 2015), the effects on animal performance, feed efficiency, carcass attributes and meat characteristics have not been studied in weaned lambs intensively fattened and fed CPD. A study by Oliveira et al. (2015) happens to be the only study found considering the effect of grain (corn) processing, i.e. whole grain, dry beans ground, and wet grain, on lamb carcass and meat quality, and they found no significant effects. Therefore, the present study was proposed to investigate the effect of different grinding sizes of cereals included in CPD on animal performance, feed efficiency and carcass and meat quality of fattening lambs.

2. Material and methods

All handling practices followed the recommendations of the Directive 2010/63/EU of the European Parliament and of the Council on the protection of animals used for scientific purposes and the IGM-CSIC Animal Experimentation Committee. The commercial slaughtering

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or killing process was carried out according to the Council Regulation (EC) No 1099/2009 on the protection of animals at the time of killing.

2.1. Animals and diets

Twenty intact male Merino lambs (age 47 ± 0.78 days, aged 6–8 weeks old) were allocated randomly to one of two different groups ($n = 10$ per dietary treatment) balanced by body weight (14.8 ± 0.16 kg). Animals were housed in individual sawdust-bedded concrete-floor pens (1.45 m width \times 1.40 m length \times 1.30 m height) where they received the experimental diets ad libitum at 09:00 h daily. Both groups were fed the same CPD, the only difference being the grinding size of the cereals (2-mm vs. 6-mm) included in the CPD before pelleting. Ingredients (43% barley, 15% corn, 24% soybean, 15% barley straw, and 3% vitamin-mineral premix), chemical composition (AOAC, 2003), particle size distribution (ADSA, 1970; Waldo et al., 1971) and physical characteristics of both CPDs are described in detail by Andrés et al. (2018). The dietary fatty acid profile (32% C16:0, 5% C18:0, 23% C18:1, 22% C18:2, 3% C18:3) can be found in Blanco et al. (2017). The total duration of the experiment was 48 days, including the adaptation period (7 days), until lambs reached a weight of 27 kg. Animals were weighed twice weekly before the morning feeding, in order to calculate the average daily weight gain, until the animals reached the targeted final body weight (BW).

2.2. Slaughter procedure and carcass characteristics

When an animal reached 27 kg, feed and water were withdrawn, and after 1 h each lamb was weighed again (slaughter BW). The animal was immediately stunned and slaughtered by exsanguination from the jugular vein, eviscerated and skinned. The dressed carcass (Colomer-Rocher, Morand-Fehr, Delfa, & Sierra Alfranca, 1988) was weighed before (hot carcass weight, HCW) and after chilling at 4 °C for 24 h (cold carcass weight, CCW). During chilling of the carcass, the pH value of the *longissimus thoracis* muscle was determined at the level of the sixth rib on the right side at 0 h, 45 min and 24 h post-mortem, using a pH meter equipped with a penetrating electrode and calibrated with buffers at 4 °C (pH meter Metrohm® 704, Switzerland). Chilling losses were calculated as the difference between HCW and CCW and were expressed as a proportion of the HCW. The carcass yield was calculated as the proportion CCW/BW recorded just before slaughtering. The left side of each carcass was divided into commercial cuts according to Colomer-Rocher et al. (1988); each cut was weighed to assess its proportion in the carcass. Then, the *longissimus thoracis* (LT) *et lumborum* (LL) muscle was removed from the right carcass side at 24 h post-mortem for chemical analysis (LT), colour (raw meat, LL), lipid oxidation and texture parameters (cooked meat, LL).

2.3. Meat quality characteristics

The LT samples were used for proximate analysis following the methods described by AOAC (2003), whereas the LL samples were cut into slices 2.5-cm thick. Five slices from each animal were placed on impermeable polypropylene trays and wrapped with an oxygen-permeable polyvinylchloride film (oxygen permeability of 580 ml/m² per h). The packaged meat was then stored under simulated retail display conditions (12 h daily illumination and 3 ± 1 °C). On each sampling day [0 (after 60 min), 1, 3, 6, 8 and 10 days] the polypropylene trays were unwrapped to measure lightness (L^*), redness (a^*) and yellowness (b^*) values (CIE, 1976) using a chromameter operating with a D65 illuminant and 10° observer angle (Minolta® Chroma Meter 2002, Germany) in the same two slices every single day. The hue angle (Hue), which defines colour (0° is red; 90° is yellow), was calculated as arctangent (b^*/a^*), and the chroma, a measure of colour intensity (0 is dull; 60 is vivid), was computed as $\sqrt{(a^{*2} + b^{*2})}$ (Young & West, 2001).

Additionally, another LL slice (2.5 cm thick) was sampled at day 0 of storage and cooked in a double-sided griddle (preheated at 220 °C) until a core temperature of 75 °C was reached, following the guidelines for cooking procedures of AMSA (1995). Then, cooked samples were placed on impermeable polypropylene trays, over-wrapped with an oxygen-permeable film (580 ml/m²/h) and stored under refrigeration (12 h daily illumination and 3 ± 1 °C) for 72 h. The concentrations of thiobarbituric-acid reactive substances (TBARS) in the cooked lamb samples before and after the storage were analysed according to the methodology described by Nam and Ahn (2003).

The remaining two slices from each LL muscle were sampled at day 0 of storage and one was used to determine the water holding capacity (WHC), via cooking losses, and the texture profile analysis (TPA) on cooked samples, and the other one was used to analyse the fatty acid (FA) content. Water holding capacity and TPA were determined according to the methodology described by Andrés et al. (2014). Additionally, the LL slice used for FA analysis was first triturated in a domestic food processor, and a 1-g sample portion was lyophilised and then submitted to in situ transesterification (Carrapiso, Timón, Petrón, Tejada, & García, 2000). The gas-chromatography mass-spectrometry (GC-MS) technique was used for FA determination. A 7890A gas chromatograph equipped with a HP 88 column (100 m \times 0.25 mm \times 0.20 mm film thickness) and coupled to a 5975C mass spectrometer (Agilent Technologies; Palo Alto, CA, USA) were used for the analysis. The chromatographic procedure was based on that described by Liu, Stephen-Inbaraj, and Chen (2007) with modifications. Briefly, helium was used as carried gas at a flow ratio of 1 ml/min, the injector and detector operated at 200 °C and 300 °C, respectively, the injection was carried out in split mode at a ratio 30:1, and the oven program temperature started at 170 °C (maintained for 24 min), the temperature was increased to 220 °C at 7.5 °C/min and then to 230 °C at 10 °C/min (maintained for 5 min). The transfer line temperature was 230 °C and the mass spectrometer operated in electron impact mode with an electron multiplier and ion voltages of 70 eV and ion voltage 1560 V, respectively. Detection was performed by total ion mode with a scanning range of 40–350 *m/z* mass units at 3.94 scans/s, and peak identification and quantification were carried out using a set of four standard solutions prepared from a mixture of 37 fatty acid methyl esters (FAME; Supelco 37 Component FAMEmix), plus the *c*11–18:1, *t*11–18:1, *c*9,*t*11–18:2 and *t*10,*c*12–18:2 methyl esters (Sigma Aldrich Química, S.L., Madrid, Spain). The identification was supported by comparison of the peaks' mass spectra with data available in a Mass Spectral Library (Hewlett Packard, Willey 275). The quantification of individual FAME was carried out by the external standard method. The concentration of the FA methyl esters (FAME) were first calculated as percentage of total FAME, then converted into percentage of FA in total FA (i.e. g FA/100 g total FA) using the corresponding coefficient of conversion ($F_{\text{FAMEi}} - F_{\text{Ai}} = M_{\text{FAi}}/M_{\text{FAMEi}}$), where M_{FAi} is the molecular weight of the FAi and M_{FAMEi} is the molecular weight of the FAMEi). Finally, the FA percentages were converted into mg FA/100 g of meat using both the lipid conversion factor (LCF) for lean lamb (0.916) to account for the presence of lipid components other than FA (Greenfield & Southgate, 2003), i.e. g of total FA = g of IMF \times LCF, and the amount of IMF in the meat samples (g IMF/100 g of meat).

2.4. Calculations and statistical analysis

Average daily weight gain (ADG, g/day) was estimated as the regression coefficient (slope) of BW against time using the REG procedure of the SAS package (SAS Inst. Inc., Cary, NC). The feed conversion ratio (FCR) was obtained by dividing the feed intake per day by the ADG (g/g). Residual feed intake (RFI) was calculated according to the procedure described by Santos et al. (2018), slightly modified. Thus ADG, mid-test metabolic body weight ($\text{MBW} = \text{BW}^{0.75}$) and intramuscular fat (IMF) content in LT of all the lambs were used as independent variables in a multiple linear regression model to predict DMI. The statistical model

used was: $DMI_i = \beta_0 + \beta_1 MBW_i + \beta_2 ADG_i + \beta_3 IMF_i + e_i$, where DMI_i represents the dry matter intake of the i th animal; β_0 is the intercept; β_1 , β_2 and β_3 are the regression coefficients on MBW, ADG and IMF, respectively, and e_i is the residual for the i th animal. Predicted DMI may be assumed as the “average” or expected intake for animals of similar weights, weight gains and body fatness. The difference between the actual daily feed and the predicted (expected) feed intake for each individual corresponds to the residual feed intake (RFI).

All performance, carcass, and meat data were subjected to one-way analysis of variance using the GLM procedure of the SAS package (1999) with the experimental diet (cereal particle size) as the only source of variation. In all cases, the individual lamb was considered as the experimental unit. The pH in *longissimus thoracis* muscle and meat colour data were analysed by repeated measurements using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). The model included fixed effects of the diet, time (hour for pH measurements and storage day for meat colour) and their interaction. The effect of animal nested in diet was used as the error term to test the effect of diet. Meat pH values recorded 24 h post-mortem were initially used as a covariate but finally removed from the models due to the lack of significant effects. Significance was declared at $P < .05$.

3. Results

3.1. Animal performance, feed efficiency and carcass characteristics

Animal performance parameters and carcass characteristics are summarised in Table 1. As previously reported (Andrés et al., 2018), daily DM intake was 7% greater ($P < .05$) in lambs fed the 2-mm CPD than in those fed the 6-mm CPD. However, no significant differences were observed for ADG or FCR. Carcass characteristics were not affected by grain particle size in CPD (Table 1). Residual feed intake (RFI) was significantly ($P < .05$) reduced when lambs were fed the 6-mm CPD compared with 2-mm CPD.

3.2. Meat pH, proximate composition and fatty acid profile

Neither the diet nor the interaction between diet and time revealed significant differences in the LT muscle pH (data not shown). Similar to previous studies (Blanco et al., 2018), only a steady decrease ($P < .05$) in meat pH was observed as time progressed in both groups. The final pH values (5.79 vs. 5.93 for 2-mm and 6-mm lambs, respectively; SED = 0.153; $P = .564$) were similar to those previously published for

Table 1

Dry matter intake, average daily gain, fattening period, feed conversion ratio and carcass characteristics of lambs receiving complete pelleted diets with cereals ground into different particle sizes (n = 10 per group).

	2-mm	6-mm	SED	P-value
Slaughtering live body weight (kg)	27.4	27.1	0.232	0.116
Average daily weight gain (g/day)	307	277	19.1	0.132
Feed conversion ratio (kg/kg)	2.93	2.98	0.149	0.771
Residual feed intake (g DM/ d)	15.4	- 14.0	13.74	0.046
Slaughter weight (kg)	27.4	27.1	0.235	0.110
Fattening period (days)	45.1	49.0	2.942	0.201
Hot carcass weight (HCW, kg)	13.1	12.8	0.228	0.177
Cold carcass weight (CCW, kg)	12.7	12.4	0.230	0.215
Chilling losses (%)	2.94	2.77	0.237	0.496
Carcass yield (%)	46.4	45.9	0.769	0.587
Carcass cuts (%)				
Shoulder	20.2	20.3	0.413	0.740
Loin-rib	16.7	17.3	0.772	0.491
Tail	1.19	1.21	0.097	0.844
Best-end	7.56	8.23	0.444	0.158
Scrag-end	9.03	8.65	0.496	0.452
Breast-flank	10.0	9.74	0.643	0.474
Leg	35.1	34.6	0.735	0.498

Table 2

Effect of grain particle size of the cereals included in complete pelleted diets on the proximate composition of meat samples (*longissimus thoracis*) expressed in g/kg of meat (n = 10 per group).

	2-mm	6-mm	SED	P-value
Humidity	756	748	6.8	0.259
Ash	13.0	11.3	0.54	0.008
Crude protein	199	201	2.8	0.610
Crude Fat	28.1	37.2	3.58	0.025

Table 3

Effect of grain particle size of the cereals included in complete pelleted diets on the total fatty acid profile (mg/100 g meat) in meat (*longissimus lumborum*) from fattening lambs (n = 10 per group).

	2-mm	6-mm	SED	P-value
SFA	975.9	1284.0	128.39	0.032
10:0	3.34	5.88	1.140	0.044
12:0	6.86	7.19	2.216	0.882
14:0	55.9	81.56	11.641	0.047
15:0	6.42	8.88	1.266	0.075
16:0	554.1	742.3	79.94	0.035
17:0	31.4	37.7	5.81	0.306
18:0	316.6	398.7	36.72	0.044
20:0	0.19	0.41	0.158	0.188
22:0	0.73	0.82	0.577	0.880
BCFA	7.32	10.38	2.133	0.182
iso-15:0	0.46	0.99	2.232	0.041
iso-16:0	0.75	1.24	0.361	0.199
iso-17:0	2.29	2.79	0.550	0.390
anteiso-17:0	1.43	2.08	0.477	0.206
iso-18:0	2.39	3.28	0.812	0.303
MUFA	1107.5	1392.4	140.83	0.066
c9-14:1	0.83	1.74	0.429	0.055
c9-16:1	33.5	42.7	7.55	0.252
c9-17:1	25.4	28.6	3.29	0.362
t10-18:1 + t11-18:1	47.3	70.7	12.89	0.106
c9-18:1	904.0	1139.8	114.66	0.061
c11-18:1	92.5	105.2	10.50	0.255
19:1 ^(*)	2.48	2.54	0.697	0.941
c11-20:1	1.43	1.10	0.585	0.590
PUFA	584.9	646.7	60.85	0.336
trans-18:2	2.90	3.71	0.869	0.375
18:2n - 6	353.4	401.7	39.74	0.242
18:3n - 6	3.89	3.03	0.648	0.216
18:3n - 3	23.0	23.3	3.05	0.903
c9,t11-18:2 ^(**)	2.30	4.39	0.711	0.011
t10,c12-18:2	1.16	3.35	0.664	0.005
20:2n - 6	3.74	4.77	0.601	0.113
20:3n - 6	16.9	16.4	2.064	0.813
20:3n - 3	8.74	10.17	1.377	0.324
20:4n - 6	138.8	146.7	15.04	0.612
20:5n - 3	21.1	20.1	2.549	0.708
22:4n - 6	7.03	6.57	1.028	0.679
22:5n - 3	2.01	2.41	0.860	0.658
P/S	0.65	0.51	0.079	0.097

FAME: fatty acid methyl esters; SFA: total saturated fatty acids; BCFA: branched-chain fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids (including conjugated linoleic acids); P/S: ratio PUFA/SFA; (*) unknown isomer; (**) coelutes with t7,c9-18:2.

lambs reared under similar conditions (Blanco et al., 2014).

Table 2 summarises the results regarding the chemical composition of LT muscle. A significant ($P = .008$) effect was detected for ash content, being greater in meat from lambs fed the 2-mm CPD (13.0 vs. 11.3 g/kg meat). There were also significantly higher values ($P = .025$) of IMF in meat from the 6-mm lambs (28.1 vs. 37.2 g/kg meat). Moreover, a higher amount ($P = .032$) of saturated fatty acids (SFA), was observed in the meat from lambs fed the 6-mm CPD compared to the 2-mm group (Table 3), mainly due to higher contents of 16:0 ($P = .035$) and 18:0 ($P = .044$). Similarly, the monounsaturated fatty

Table 4

Effect of grain particle size of the cereals included in complete pelleted diets for fattening lambs on the colour parameters of meat (*longissimus lumborum* muscle) refrigerated (4 °C) for up to 10 days (n = 10 per group).

		<i>L</i> *	<i>a</i> *	<i>b</i> *	Chroma	Hue
Diet	2-mm	41.5	10.8	4.91	12.0	24.2
	6-mm	40.1	11.4	4.40	12.3	20.6
Storage day	0	40.8 ^b	10.7 ^b	3.90 ^a	11.4 ^b	20.1 ^{bc}
	1	44.3 ^d	11.1 ^{bc}	6.48 ^c	12.9 ^c	30.4 ^c
	3	42.4 ^c	11.7 ^c	5.77 ^{bc}	13.0 ^c	26.3 ^d
	6	39.6 ^a	12.2 ^d	5.05 ^b	13.3 ^c	22.1 ^c
	8	38.8 ^a	11.3 ^b	3.48 ^a	11.9 ^b	16.6 ^a
	10	38.9 ^a	9.62 ^a	3.24 ^a	10.3 ^a	18.7 ^{ab}
RSD ₁		3.429	2.464	2.369	2.546	10.17
RSD ₂		1.693	1.189	1.126	1.189	5.292
P- values						
Diet		0.037	0.218	0.257	0.453	0.070
Storage day		0.001	0.001	0.001	0.001	0.001
Diet × day		0.895	0.929	0.026	0.521	0.029

RSD₁: Comparison between diets; RSD₂: Comparison between diets and diets × day; ^{a,b,c,d} Within each column, significant ($P < .05$) differences among storage days are represented by different superscripts.

acid (MUFA) content showed a trend towards higher amounts in the 6-mm lambs, probably as a consequence of higher levels of c9-18:1 ($P = .061$). No differences were observed in total polyunsaturated fatty acids (PUFA) ($P = .336$) but for CLA isomers [c9,t11-18:2 ($P = .011$) and t10,c12-18:2 ($P = .003$)], higher contents were observed for 6-mm lambs. The differences previously reported for the concentration of branched-chain and odd fatty acids fatty of the ruminal content in the same animals (Andrés et al., 2018) were not observed in the muscle fat, but a different evolution was detected for iso-15:0 ($P = .041$).

3.3. Colour, lipid peroxidation and texture parameters of meat

Table 4 summarises the results of LL muscle colour stability during aerobic storage under refrigeration (4 °C) for 10 days. Lightness (L^*) showed significant ($P = .037$) differences between diets with the lighter meat from the 2-mm lambs (41.5 vs. 40.1). Meat redness (a^*), yellowness (b^*) and colour intensity (chroma) were not affected. However, a trend towards significant ($P = .070$) differences in the hue angle was observed, with greater values in the LL muscle of the 2-mm group when compared to 6-mm lambs (24.2 vs. 20.6). All of the colour characteristics changed during the storage period ($P < .001$). The values increased during the first three-six days of storage and decreased afterwards. Significant interactions diet × day were observed for b^* and hue ($P < .05$). These consisted of significantly higher values of b^* and hue angle at day 10 of storage in 2-mm samples as compared with 6-mm (b^* : 4.83 vs. 1.64; hue: 26.14 vs. 11.24, respectively; data not shown in tables for brevity).

TBARS levels in cooked meat before storage were under 0.2 mg/kg. In the 72 h stored meat the TBARS values were similar in both groups [4.89 vs. 5.03 mg of malonaldehyde (MDA) per kg of meat for 2-mm and 6-mm, respectively; SED = 0.382; $P = .723$]. Similarly, the results for cooking losses and TPA parameters of non-aged meat after cooking did not show significant differences ($P > .05$) between treatments (Table 5). However, a statistical trend towards greater cooking losses in meat from the 6-mm group was observed ($P < .085$).

4. Discussion

A proper evaluation of the effects of increasing sieve grinding size of cereal grains included in CPD formulated for growing lambs on animal performance, feed efficiency, carcass characteristics and meat quality traits is necessary before any recommendation can be made for lambs raised in intensive production systems.

In agreement with Ferreira et al. (2011), carcass characteristics

Table 5

Effect of grain particle size of the cereals included in complete pelleted diets for fattening lambs on cooking loss (% of water loss) and texture profile analysis of not-aged cooked meat samples (*longissimus lumborum* muscle; n = 10 per group).

	2-mm	6-mm	SED	P-value
Cooking loss	30.2	31.9	0.91	0.085
Hardness (N)	218	220	8.5	0.849
Cohesiveness	0.456	0.445	0.0082	0.198
Springiness	0.487	0.475	0.0098	0.237
Chewiness (N)	48.5	46.4	2.22	0.360

N: Newtons.

were not modified by grain particle size (Table 1). Moreover, the values in the present study were similar to those described for animals grown under similar rearing conditions (Blanco et al., 2014; Galvani, Pires, Hübner, Carvalho, & Wommer, 2014; Jacques, Berthiaume, & Cinq-Mars, 2011; Rodríguez et al., 2008; Sormunen-Cristian, 2013). However, as previously reported by Andrés et al. (2018), the 6-mm animals might have been more efficient than 2-mm lambs due to a decreased fermentation of starch in the rumen, and hence a higher percentage of starch reaching the small intestine. These events might explain the lack of differences between treatments in FCR (Table 1) despite the lower DMI observed in the 6-mm group, as previously published in Andrés et al. (2018). Additionally, more starch digestion in the small intestine might have promoted higher levels of glucose absorbed into the blood stream of the 6-mm group, so we can speculate that the secretion of insulin (a hormone that promotes lipogenesis in all adipocyte tissue) might have been increased (Gathercole et al., 2011), thus enhancing IMF accumulation in this group of lambs (Table 2).

Also, it has been suggested that some compounds generated under acidogenic conditions (e.g., lipopolysaccharide from *E. coli* and t10c12-CLA) might reduce lipogenesis in the mammary gland (Oetzel, 2007; Wynn et al., 2006), although some contradictory results have been found (Colman et al., 2010). In any case, Wynn et al. (2006) demonstrated that supplying t10c12-CLA does not depress IMF. Accordingly, there should not be any association between the t10 shift caused by the more acidogenic conditions in the 2-mm lambs (Andrés et al., 2018) and the reduced IMF deposition observed in this group of animals when compared to the 6-mm group.

On the other hand, the lipogenesis reduction in IMF of lambs fed the most acidotic diet (2-mm CPD) seems to be consistent with the FA profile observed in meat samples (Table 3). Phospholipids are essential components of cell membranes (rich in PUFA) and its amount remains relatively constant regardless the body fatness. On the contrary, the neutral lipids (rich in SFA) increase as body fatness is raised (Wood et al., 2008). These facts support the differences between experimental groups in meat FA profiles, with significantly higher amounts of SFA in meat from lambs fed the 6-mm CPD compared with the 2-mm group. Unfortunately the fatty acid analysis employed for IMF did not resolve t10-18:1 and t11-18:1, but their combined content did not differ between diets. Based on rumen fatty acid profiles previously published (Andrés et al., 2018), it is likely that t10-18:1 would have dominated and a trend towards increased t11-18:1 would have likely occurred with increased grind size.

On the basis of the present experimental plan, it is difficult to explain the differences found in L^* between the dietary treatments, especially taking into account the lack of differences in a^* and b^* . Since the dry-matter intake was different between lambs from each treatment (Andrés et al., 2018), a nutritional-intake effect on those muscle characteristics affecting muscle lightness (e.g., levels of vitamin E, iron), might have contributed (Calnan, Jacob, Pethick, & Gardner, 2016). In addition, the pH value and lamb age are additional factors related (inversely) to L^* in lamb (Calnan et al., 2016); however, in this study neither pH nor age at slaughter were significantly different among the

experimental groups. Furthermore, since muscle L^* seems unaffected by lamb carcass fatness (Sañudo, Alfonso, Sánchez, Delfa, & Teixeira, 2000), L^* differences could not be explained by near-to-significant differences in IMF content. The higher lightness in 2-mm throughout the display period could represent a sensory advantage for this meat because L^* appears to be positively correlated with colour sensory scores in displayed meat from fattening lambs (Callejas-Cárdenas et al., 2014). This study suggested an L^* value of 42 as a critical value for consumer acceptance during lamb chop display. The study by Khiliji, van de Ven, Lamb, Lanza, & Hopkins (2010) found that average consumers considered lamb colour as acceptable when L^* and a^* values were higher than 34 and 9.5 respectively, although they should be higher than 44 and 14.5 to have a positive result with a 95% confidence level. Accordingly, the L^* values in the present study would indicate lamb colour deterioration ($L^* < 42$) over 3 days of display. Subsequently, L^* values at day 6 of storage decreased respectively to 40.3 and 39.1 for 2-mm and 6-mm lamb (data not shown in Tables for brevity).

In contrast, the greater b^* and hue angle in the 6-mm LL muscle after eight days of refrigeration, which explains the significant interaction between diet and storage day, may have been due to a higher oxidation rate of ferrous haem-iron (Fe^{2+}) into its ferric form (Fe^{3+}) in this meat, resulting in a colour shift from bright red to a less desirable brownish (metmyoglobin) colour (AMSA, 2012; Andrés et al., 2014). This lower oxidative stability might be related to the lower amount of SFA in the fat of lambs fed the 2-mm CPD, which may have promoted an increased susceptibility to lipid peroxidation, and subsequently haem-Fe oxidation. However, the possible increased susceptibility of raw meat from 2-mm CPD fed lambs to lipid oxidation was not corroborated by meat TBARS values in cooked meat stored for 72 h.

5. Conclusions

Even though no changes in FCR or carcass characteristics were observed, the meat of animals fed CPD formulated with cereals ground more coarsely (6-mm sieve) showed higher levels of IMF, a greater proportion of saturated fatty acids in fat and a better colour stability during refrigeration. All these effects may be partially related to changes in ruminal fermentation resulting in more starch leaving out the rumen to be digested in the small intestine, which may improve feed efficiency in lambs fed a CPD including cereal grains ground more coarsely (6-mm sieve).

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Declaration of Competing Interest

The authors have no conflict of interests to declare.

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