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Physico-chemical characteristics of meat after dietary supplementation of lambs with essential oils and calcium malate

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Abstract

The objective of this study was to determine the effect of a mixture of essential oils (EO), calcium malate (CM), or their combination (EO + CM) on the physico-chemical characteristics of lamb meat. A total of 36, two-month-old, 20 ± 3 kg live weight, male lambs were allocated to individual cages and randomly distributed into four treatment groups: 1, basal diet (BD); 2, BD + 150 g of EO ton^{-1}; 3, BD + 2.5 kg of CM ton^{-1}; and 4: BD + 150 g EO + 2.5 kg CM ton^{-1}. Feed and fresh water were available *ad libitum* to the lambs during the 56-day experiment. There was no difference in the chemical composition of the lamb meat although collagen was higher in the EO + CM treatment. The percentage of omega-3 and eicosapentaenoic acid was higher in the OE + CM group, but there were no differences in the total percentage of saturated or unsaturated fatty acids. The shear strength of raw meat was higher in the BD group, but the shear strength of cooked meat was lower in the EO group than in the BD or CM groups. There was no difference in water retention in raw meat or in water loss in cooked meat between groups. The colour of the meat was not affected by treatment. The EO and EO + CM groups had an increase in meat tenderness and the concentration of some unsaturated fatty acids, but this did not affect the colour or chemical composition of the lamb meat.

Keywords: chemical composition, fatty acids, meat colour, tenderness
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Introduction

The demand for animal protein will increase as the world population increases, which is projected to exceed nine billion by 2050. Meat consumption in developed countries has become reduced out of consideration for human health and the environment, but in developing countries, such as China, India, Russia, Mexico, and Brazil, the population with higher incomes seeks to acquire more and better products for consumption, such as meat, milk, and cheese (Chriki and Hocquette, 2020). As the population has greater access to meat consumption, they demand higher quality in terms of colour, flavour, softness, juiciness (Mancini and Hunt, 2005; Garmyn, 2020) and nutritional composition, mainly in the amount of saturated fatty acids present in meat that can affect human health (Junkuszew *et al*., 2020).

In Mexico, chemical additives, mainly ionophores, are used as modifiers of ruminal fermentation for the finishing of lambs with diets high in concentrate (Castillo *et al*., 2004). These compounds prevent the appearance of metabolic diseases such as acidosis and can increase the efficiency of dietary utilisation with better feed conversion, weight gain, and carcass yield (Garcia-Galicia *et al*., 2020). However, consumers prefer cleaner meat, without the excessive use of chemical additives.

Essential oils (EO) and organic acid salts, such as calcium malate, have been studied as substitutes for chemical additives and can be used to preserve ruminal health and improve animal

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performance (Ortuño *et al*., 2014; Malekkhahi *et al*., 2015; Loya-Olguin *et al*., 2019; García-Galicia *et al*., 2020; Haro *et al*., 2020). The published results on the use of EO in the finishing of lambs, regarding the physico-chemical characteristics of the meat, are highly variable. Smeti *et al*. (2013) observed no effect of rosemary EO on lipid oxidation or meat discolouration but did observe an improved polyunsaturated fatty acid level and acceptability (Smeti *et al*., 2018). Elsewhere, rosemary extract reduced discolouration and lipid oxidation and improved the shelf life of meat (Ortuño *et al*., 2014). Garcia-Galicia *et al*. (2020) reported that oregano oil improved meat tenderness; however, Simitzis *et al*. (2008) reported no changes in smoothness with oregano oil use. Studies with calcium malate and its effect on meat quality are scarce. Haro *et al*. (2020) reported an improvement in the colour but not in the chemical composition, pH, or fatty acid profile of the meat. These additives have the potential to modify rumen fermentation when fed to finishing lambs and, thus, modify the chemical composition, colour, tenderness, and unsaturated fatty acid content of the meat. Due to this potential, there is interest in the use of these additives to improve the productive response of animals (Salami *et al*., 2018), without risks to human health. However, the variability of the results requires that research be intensified to generate more reliable information.

Therefore, the objective of the present study was to determine the effect of calcium malate, a mixture of standardized EO (thymol, carvacrol, eucalyptol, and cinnamaldehyde), and their combination on the physico-chemical characteristics related to the quality of meat from lambs finished with diets high in concentrate.

Material and Methods

This research was conducted in accordance with the regulations for the use and care of animals intended for research at the Colegio de Postgraduados, and the Mexican Standard for Animal Care (NOM-062-ZOO-1999).

A total of 36 male lambs (Dorset/Hampshire cross), with an initial live weight (LW) of 20 ± 3 kg at three months of age, were randomly housed in individual indoor pens of 1.50 × 2 m, with a cement floor and a bedding of sawdust, provided with a feeder and drinker. The lambs were dewormed with 1 mL 50 kg^{-1} (LW) of subcutaneous Ivermectin (Ivomec F®) and had an adaptation period to the diets of 5 d, and 56 d in finishing. The diets were offered *ad libitum*, twice a day, with 60% in the morning (08:00) and the remaining 40% in the afternoon (17:00). The lambs were weighed at the beginning and end of the experimental period.

A basal diet (BD) (Table 1) was formulated for lambs for a daily weight gain of 300 g d^{-1} (NRC, 2007). A mixture of essential oils (EO; thymol, carvacrol, eucalyptol, and cinnamaldehyde; Emerald®, Nutryplus, Mexico), calcium malate (CM; Rumalate®, Norel, Mexico) or the combination of these was added to the basal diet. The treatments were: 1) BD; 2) BD + 150 g t^{-1} EO; 3) BD + 2.5 kg t^{-1} CM and 4) BD + 150 g t^{-1} EO + 2.5 kg t^{-1} CM.

Table 1 Ingredients and chemical composition of the basal diet

Ingredients)	**(g kg^{-1} DM)**
Corn grain ground	304
Sorghum grain ground	300
Soybean meal	169
Corn stover	150
Cane molasses	50
†Vitasal Ovine Plus®	20
By-pass fat	5
Chemical composition	
Dry matter	880.0
Crude protein	150.0
Metabolizable Energy (MJ kg^{-1})	11.3
Acid detergent fibre	110.6
Neutral detergent fibre	194.2
Ether extract	34.8
Ca	11.5
P	4

† 240 g Ca; 30 g P; 20 g Mg; 80 g Na; 120 g Cl; 5 g K; 5 g S; 5 mg Cr; 4,000 mg Mn; 2,000 mg Fe; 5,000 mg Zn; 100 mg I; 30 mg Se; 60 mg Co; 500,000 IU vitamin A; 150,000 IU vitamin D; 1,000 IU vitamin E

The lambs were weighed 24 h before slaughter and then slaughtered according to the Mexican Standard for Animal Care (NOM-062-ZOO-1999). The carcasses of the lambs were washed with drinking water and left to ventilate for 2 h. Subsequently, the carcasses were weighed and 7-cm-wide

samples of the *Longissimus dorsi* muscle (300 g) were taken and refrigerated at 4 °C for 24 h to record the post-mortem cold weight. In the *Longissimus dorsi* samples, pH, temperature, chemical composition, concentration of long-chain fatty acids, water retention capacity, water loss due to cooking, shear strength (cooked and raw meat), and colour were determined.

The pH was measured using a portable potentiometer with automatic temperature compensation (HANNA® instruments, HI 99163, EU), and the temperature was recorded in three different areas of the muscle (*Longissimus dorsi*) at 0 h and 24 h post-mortem.

The content of fat, moisture, protein, and collagen was determined in the meat using near infrared (NIR) spectroscopy equipment (FoodScan®, Foss, Denmark) in the region of 850–1050 nm, equipped with a calibration of artificial neural networks.

The determination of long-chain fatty acids (FA) in meat was conducted according to the method of Jenkins (2010), with esterification and trans-esterification catalysed by 5% methanolic hydrogen chloride. The prepared meat samples were analysed using gas chromatography (HP6890 gas chromatograph with FID detector, with automatic injector) with a chromatographic column (100 m × 0.25 mm × 0.20 µm; SUPELCO SP-2560). FA were quantified as methyl esters (FAME) (French *et al*., 2000). The FAME were separated and the amounts of saturated FA (SFA), monounsaturated FA (MUFA), and polyunsaturated FA (PUFA) were determined using gas chromatography. The indices of fatty acids of interest for human health were calculated as:

P/S = PUFA/SFA ratio, (1)

P/S2 = PUFA/(SFA–C18:0) ratio, (2)

(the contribution of C18:0 was subtracted because it is not considered to be hypercholesterolemic; Diaz *et al*., 2005), and

ATT = antithrombotic potential (C20:3 + C20:5 n–3)/(C20:4 n–6). (3)

The water retention capacity (WRC) was determined using the centrifugation method (Leal *et al*., 2015). Five-gram samples of finely minced meat without fatty tissue were placed in 50 mL centrifuge tubes (Nalgen), homogenised with 8 mL of 0.6 M sodium chloride solution, and refrigerated at 4 °C. Subsequently, they were centrifuged for 15 min at 15,652 × *g* in a centrifuge (model J2-HS, BECKMAN). The excess was decanted and the final volume was measured using the following equation:

$$\text{Volume of 0.6 M NaCl (mL)/100 g of meat} = (V_i - V_s)/m \times 100 \quad (4)$$

where V_i = initial volume of 0.6M NaCl solution (8 mL); V_s = volume recovered from the supernatant (after centrifugation); and m = sample weight (5 g). The results were reported as the volume of solution (in mL) retained in 100 g of meat.

The amount of water lost during cooking was determined according to the method of Bejerholm and Aaslyng (2004). Fillets of 5 g and 1.5 cm thickness were cut from raw meat after 24 h of maturation and were fried in a pan at 155 °C, turning the meat every 2 min until it reached an internal temperature of 70 °C. Subsequently, the samples were cooled for 15 min at room temperature (20–25 °C) and weighed. The following equation was used to determine the water loss:

$$LWC = ((P_i/P_f)/P_i)) \times 100 \quad (5)$$

where: LWC is the percentage cooking loss; P_i = initial weight of the sample; and P_f = the final weight of the sample.

The shear strength of both raw and cooked meat was measured using the Warner–Bratzler (SFWB) method and was determined in accordance with the provisions of AMSA (1995). The fillets were cooked and then the same methodology described above was used to determine the PPC. Raw and cooked fillets were perforated transversely (parallel to muscle fibres) at three different sites. Shear force was measured using a Warner–Bratzler (WB) cell with a triangular groove cutting edge, mounted on a Salter Model 235 (Warner–Bratzler meat shear, GR manufacturing Co. 1317 Collins LN, Manhattan, Kansas, 66502, USA) associated with a texture meter (TAXT Express Standard, Stable Micro Systems Surrey, UK). This method was used to measure the force required (kg cm^{-2}) to cut the fillet of meat. The data are expressed in kg force.

Fat tissue was manually removed from each meat sample and three colour measurements were made in different areas at 0, 3, 6, 12, 24, 48, 72, 96, 120, 144, 168 and 192 h post-mortem, using a brand meat colorimeter (Konica Minolta CM-700 d).

A randomised complete block design was used, and all data were analysed using the Shapiro–Wilk test to determine normality. The data on chemical composition, fatty acid profile, shear force, water retention capacity, and meat cooking losses were analysed using the PROC GLM in SAS 9.4, with the type of diet, block (with two experimental units per treatment), inside block, and their interactions as fixed effects. The statistical model was:

$$Y_{ijk} = \mu + \tau_i + \beta_j + (\tau\beta)_{ij} + \varepsilon_{ijk} \quad (6)$$

where: Y_{ijk} = observation k in treatment i and block j; μ = the overall mean; τ_i = the effect of treatment, i; β_j = the effect of block, j; $(\tau\beta)_{ij}$ = the interaction effect of treatment i and block j; and ε_{ijk} = the random error with mean 0 and variance σ^2.

Meat colour data were analysed with the PROC MIXED tool in SAS 9.4 in a design of measures repeated over time, where the type of diet (TRAT: BD, EO, CM, EO + CM), hours of storage (TIME: 0 to 192 h), and its interaction (TRAT×TIME) were considered as fixed effects, while an individual lamb within treatment was considered as a random effect. The covariance structure was compound symmetry. The statistical model was:

$$Y_{ijk} = \mu + \tau_i + \delta_{ij} + t_k + \beta_j + (\tau^*t)_{ik} + \varepsilon_{ijk} \quad (7)$$

where: μ = the overall mean; τ_i = the effect of treatment i; t_k = the effect of time k; $(\tau^*t)_{ik}$ = the effect of the interaction between treatment i and time k; δ_{ij} = random error with mean 0 and variance σ^2 q, the variance between animals (subjects) within treatment, equal to the covariance between repeated measures within animals; and ε_{ijk} = the random error with mean 0 and variance σ^2, the variance between measurements within animals. When differences were detected, the comparison of means was performed by Tukey's test considering differences with $\alpha = 0.05$ and trends when $\alpha = 0.1$.

Results and Discussion

There were no differences in meat moisture, protein, and fat content (P >0.05) between treatments (Table 2). The average values concurred with those reported by Junkuszew *et al*. (2020), and the absence of effects of EO or CM concurred with those reported by Yagoubi *et al*. (2018), Smeti *et al*. (2018), and Rivaroli *et al*. (2016) in cattle; and by Haro *et al*. (2020) in lambs. It is likely that the amounts used of the additives were not sufficient to substantially modify ruminal fermentation to affect the rate of fat or muscle protein deposition (Malekkhahi *et al*., 2015). Collagen is the most abundant protein molecule in the body and maintains the health of the muscles. It is important to determine collagen content as it varies depending on the breed, productive aptitude, precocity, and age of the subject. In animals with a high growth rate, the rate of protein and collagen replacement is high (Rivaroli *et al*., 2016). In this study, collagen content was lower (P <0.05) in the BD and CM groups than in the EO or EO + CM groups, which may be related to the lower weight gain of lambs supplemented with these same additives (Ortiz *et al*., 2021).

The pH at 24 h post-mortem was higher than the initial pH (P <0.05) in the meat of the lambs supplemented with EO or EO + CM (Table 2). The final pH values were similar to those reported by Yagoubi *et al*. (2018), but lower than those reported by Smeti *et al.* (2018). Post-mortem pH is a highly important parameter in meat quality, since after 24 h, glycogen reserves are depleted and pH stabilizes (Yagoubi *et al*., 2018). A pH at 24 h post-mortem within 5.5–5.6 is associated with a light colour of the meat and shows an adequate slaughter process, while a pH >5.6 is associated with dark meat, which is common in heavy animals in a state of stress at slaughter with less muscle glycogen stores (Smeti *et al*., 2018). McGeehin *et al*. (2001) reported that the most important factors affecting pH at 24 h post-mortem were sex, cold carcass weight (R^2 = 0.80) and, to a lesser extent, age, and environmental temperature. The pH at 24 h post-mortem in the meat of lambs from the EO or EO + CM groups was probably related to the higher (P <0.05) weight of the cold carcass (Ortiz *et al*., 2021).

There were no differences (P >0.05) between groups in the total fatty acid content of lamb meat. No differences (P >0.05) between groups were observed in the content of saturated, mono-unsaturated, or polyunsaturated fatty acids between treatments (Table 3). However, the content of C20: 5 n-3 and omega 3 in the group with EO+CM was higher (P <0.05) than in the group with EO, but there were no differences (P >0.05) in the total content of omega 6 fatty acids or omega 9 between groups.

Table 2. Chemical composition (g/100 g) and pH of lamb meat from lambs supplemented with essential oils, calcium malate, and a combination of these.

	Treatments					
Item	BD	EO	CM	EO + CM	SEM[X]	P value
Humidity	71.91	71.31	71.79	71.72	0.35	0.66
Protein	21.86	21.72	22.18	22.23	0.19	0.34
Fat	4.77	5.31	4.57	4.19	0.36	0.18
Collagen	1.43[b]	1.49[ab]	1.40[b]	1.58[a]	0.05	0.02
pH at 0 h	5.66[d]	6.22[a]	5.77[c]	6.07[b]	0.03	<0.0001
pH at 24 h	5.39[b]	5.86[a]	5.51[b]	5.72[a]	0.04	<0.0001

[ab]Means in the same row with different letters are significantly different (P <0.05).
BD = basal diet without additives; EO = mixture of essential oils (thymol, carvacrol, eucalyptol); CM = calcium malate; EO + CM = combination of essential oils and calcium malate. [X]SEM = standard error of the mean

Table 3. Mean content plus standard error of fatty acid contents in lamb meat from lambs supplemented with essential oils, calcium malate, or a combination of these

	Treatments				
Item[Y]	BD	EO	CM	EO+CM	P value
C14: 0	1.90 ± 0.11	1.89 ± 0.09	1.89 ± 0.10	1.84 ± 0.11	0.99
C16:0	22.07 ± 0.54	23.44 ± 0.48	22.70 ± 0.51	23.07 ± 0.51	0.35
C18: 0	15.72 ± 0.59	14.79 ± 0.52	16.33 ± 0.56	15.06 ± 0.56	0.27
C14: 1	0.06 ± 0.01	0.05 ± 0.01	0.05 ± 0.01	0.06 ± 0.01	0.53
C15: 1 n-5	0.37 ± 0.03	0.40 ± 0.03	0.40 ± 0.03	0.33 ± 0.03	0.40
C16: 1 n-7	2.15 ± 0.09	2.24 ± 0.08	2.11 ± 0.08	2.21 ± 0.08	0.67
C17: 0	1.69 ± 0.15	1.68 ± 0.14	1.82 ± 0.15	1.48 ± 0.15	0.47
C17: 1	1.36 ± 0.10	1.17 ± 0.08	1.15 ± 0.09	1.22 ± 0.09	0.41
C18: 1 n-9*trans*	3.16 ± 0.43	4.45 ± 0.38	3.68 ± 0.41	3.29 ± 0.41	0.17
C18: 1 n-9*cis*	42.04 ± 1.03	40.96 ± 0.91	40.68 ± 0.97	40.99 ± 0.97	0.79
C18: 2 n-6*cis*	4.19 ± 0.32	3.96 ± 0.28	3.77 ± 0.30	4.69 ± 0.30	0.23
C18: 3 n-6	0.11 ± 0.01	0.12 ± 0.01	0.10 ± 0.01	0.12 ± 0.01	0.24
CLA[y]	0.07 ± 0.01	0.08 ± 0.01	0.08 ± 0.01	0.08 ± 0.01	0.90
C20: 1 n-9	0.06 ± 0.01	0.06 ± 0.00	0.06 ± 0.01	0.07 ± 0.01	0.82
C20: 3 n-6	0.12 ± 0.01	0.10 ± 0.01	0.13 ± 0.01	0.14 ± 0.01	0.09
C20: 4 n-6	1.30 ± 0.11	1.10 ± 0.10	1.04 ± 0.10	1.31 ± 0.10	0.22
C20: 5 n-3	0.91 ± 0.07[ab]	0.77 ± 0.06[b]	0.83 ± 0.07[ab]	1.07 ± 0.07[a]	0.05
C20: 3 n-3	0.21±0.04	0.21±0.03	0.21±0.04	0.22 ± 0.04	0.99
Total, SFA	39.69 ± 0.63	39.80 ± 0.56	40.92 ± 0.60	39.66 ± 0.60	0.43
Total, MIFA	51.26 ± 0.77	50.96 ± 0.68	50.43 ± 0.73	49.67 ± 0.73	0.48
Total, PUFA	7.17 ± 0.52	6.00 ± 0.46	6.23 ± 0.49	7.76 ± 0.49	0.10
Total, ω9	47.45 ± 0.81	47.66 ± 0.76	46.52 ± 0.76	46.54 ± 0.76	0.64
Total, ω6	5.91 ± 0.39	5.61 ± 0.37	5.12 ± 0.37	6.33 ± 0.37	0.22
Total, omega 3	1.12 ± 0.08[ab]	0.98 ± 0.07[b]	1.04 ± 0.07[b]	1.37 ± 0.07[a]	0.02
P/S[X]	0.18 ± 0.01	0.15 ± 0.01	0.15 ± 0.01	0.20 ± 0.01	0.08
P/S2[W]	0.30 ± 0.02	0.24 ± 0.02	0.25 ± 0.02	0.32 ± 0.02	0.09
ATT[V]	0.85 ± 0.12	0.98 ± 0.13	0.99 ± 0.11	1.17 ± 0.11	0.32

[ab]Means in the same row with different letters are significantly different (P <0.05). BD = basal diet without additives; EO = mixture of essential oils (thymol, carvacrol, eucalyptol); CM = calcium malate; EO + CM = combination of essential oils and calcium malate; [y]CLA = conjugated linoleic acid; SFA = saturated fatty acids; MIFA = mono-unsaturated fatty acids; PUFA = polyunsaturated fatty acids; [X]P/S = PUFA/SFA ratio; [W]P/S2 = PUFA/(SFA–C18:0) ratio; VATT = antithrombotic potential (C20:3+C20:5 n–3)/(C20:4 n–6)

The P/S and PUFA/SFA ratios tended to be higher (P <0.1) in the fat of the lambs from the EO+CM group than in the other groups. These results concur with those reported by Diaz *et al.* (2005) in the intramuscular fat of lambs fed diets without additives. The total concentration of omega 3 was higher than the 0.66% reported by Haro *et al*. (2020) in lambs supplemented with malic acid, but lower than the 1.87% reported by García-Galicia *et al*. (2020) in lambs supplemented with oregano essential oil. A low omega 6/omega 3 ratio is desirable as a means of reducing high-incidence chronic diseases in Western societies and developing countries (Junkuszew *et al*., 2020). In the current study, the omega 6/omega 3 ratio of the control group was 5.3, but in the group with EO + CM, it was 4.6. Junkuszew *et al*. (2020) found an omega 6/omega 3 ratio of 8.83 in lambs but in adult ewes, the ratio was 4.5, similar to that observed in the current study. The omega 6/omega 3 ratio depends mainly on nutrition and, especially, on the sources of fatty acids and their metabolism in the rumen, as reported by Romero-Bernal *et al*. (2017), who supplemented lambs with ryegrass hay, fishmeal, or soybean meal. The profile of predominant fatty acids in the rumen depends on whether the lambs are finished on grazing or on high-starch diets, as well as the type of ruminal fermentation modifier included in the diet (García-Galicia *et al*., 2020). High omega 3 with an omega 6/omega 3 ratio of 4 has been associated with a 70% decrease in human mortality from cardiovascular disease, and a ratio of 2.5 reduces rectal cancer cell proliferation (Simopoulos, 2008). The P/S and PUFA/SFA ratios indicate the antithrombotic potential (ATT), which is an index that takes into account the specific roles of individual fatty acids on human health.

The shear strength of cooked meat from lambs supplemented with EO was lower (P <0.05) than in the BD or CM groups but similar to the EO+CM group. The shear strength of raw meat in the BD group was higher (P <0.05) than in the EO group but similar to that of the other groups (Table 4).

Table 4. Shear strength, water retention capacity, and water loss due to cooking of lamb meat from lambs supplemented with essential oils, calcium malate, or a combination of these

	Treatments				SEM[X]	
Item[Y]	BD	EO	CM	EO + CM		P value
SFWB Cooked meat, kg/cm^2	3.56^a	2.60^b	3.39^a	3.10^{ab}	0.16	<0.01
SFWB raw meat, kg/cm^2	5.77^a	4.84^b	5.33^{ab}	5.24^{ab}	0.17	<0.01
WRC, %	24.73	27.47	27.20	27.53	1.20	0.33
LWC, %	33.13	33.37	37.07	33.15	1.42	0.19

[ab]Means in the same row with different letters are different (P <0.05). BD = diet without additives; EO = Emerald® essential oil blend (thymol, carvacrol, eucalyptol); CM = Rumalate® calcium malate; EO + CM = combination of Emerald® and Rumalate®. SFWB = Warner-Bratzler shear force; WRA = water retention capacity; LWC = loss of water by cooking; [X]SEM = standard error of the mean.

The reduction in the shear strength of cooked lamb meat from lambs supplemented with essential EO concurs with that reported by García-Galicia *et al*. (2020) in lambs supplemented with oregano essential oil but is different to that reported by Simitsiz *et al*. (2008), who did not observe differences using the same supplement. Some of the intrinsic factors that affect meat texture are collagen content and solubility, sarcomere diameter, intramuscular fat content, and calpain proteolysis during aging (García-Galicia *et al*., 2020). There is a positive association between fat content and meat tenderness (Watkins *et al*., 2013). In the current investigation, an analysis of contrasts showed that the fat content in the meat of lambs supplemented with EO tended (P = 0.06) to be 18% higher than in the other groups, which may explain the greater softness of the lamb meat from the EO group.

There were no differences between treatments (P >0.05) in the water retention capacity or water loss due to cooking in the meat (Table 4). This agrees with the report of Haro *et al*. (2020) in lambs supplemented with EO, and by Abdelmalek *et al*. (2019) and Smeti *et al*. (2020) when using myrtle or rosemary oil as supplements. Atti *et al*. (2013) wrote that the more acidic the pH of the meat, the higher the water loss. In this investigation, the meat did not show a decrease in pH 24 h post-mortem, which may explain the observed result.

There were no differences between treatments (P >0.05) in the measured parameters (L*, a*, b*, and C*) of meat colour or treatment × time interactions. These results agree with those reported by Ortuño *et al*. (2014), Smeti *et al*. (2018), and Yagoubi *et al*. (2018), who did not observe differences in the colour parameters (L*, a*, b*, C*, and H*) when using rosemary oil, but are in contrast to those of Haro *et al*. (2020), who reported differences in the meat colour parameters of lambs supplemented with oregano essential oil. Ortuño *et al*. (2014) reported that time modifies all colour parameters, turning the meat from bright red to brown. Here, this was reported in terms of CIELab coordinates, with an increase

in L*, b*, and H* accompanied by a reduction in a* and C* in meat from all groups. The colour of the meat is affected by numerous factors such as breed, diet, age, weight and stress at slaughter, and the storage conditions of the meat, among others (Haro *et al*., 2020). In the current study, the conditions of diet, breed, management in the finishing process, and slaughter were similar; thus, it is thought that the doses of EO or CM used were insufficient to generate a detectable change. The colour of the meat is crucial in ensuring positive consumer appreciation, as consumers normally associate a bright red colour with fresh, quality meat (Wood *et al*., 2008). Meat with an L* value ≥34 is acceptable, and meat with an L* close to 44 is considered the ideal acceptability value for 95% of consumers (Khliji *et al*., 2010). In this study, the parameter, L*, had an average of 40.8, indicating that the meat was within a range of medium acceptability.

Conclusions

Essential oils, either alone or combined with calcium malate, in the diet of finishing lambs improve the pH of meat, increase the tenderness of cooked meat, and increase the total content of omega 3 fatty acids, all without modifying the colour, chemical composition or water retention capacity. This means that these additives can improve the quality of lamb meat. It is suggested that the investigation of these additives continue, and that the quantity as a supplement in high concentrate diets be increased for finishing lambs.

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Authors' Contributions

MAOH conducted the research and wrote the manuscript, under the supervision of JRBG. PAMH collaborated in laboratory analyses. JDGR collaborated in lamb handling, slaughter, and carcass characterisation. OVVM led the data analysis and interpretation of results. A critical revision of the version to be published was performed by JRBG, SSGM, and MAOH.

Conflict of Interest Declaration

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

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