

ORIGINAL ARTICLE

Effects of dietary oil blend on fatty acid composition, oxidative stability and physicochemical properties of Longissimus thoracis et lumborum muscle in goats

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ABSTRACT

This study examined the effects of dietary blend of 80% canola oil and 20% palm oil (BCPO) on the physicochemical properties, antioxidant status, oxidative stability and fatty acid composition of Longissimus thoracis et lumborum (LTL) muscle from goats during chill storage. Over a 14-week feeding trial, 24 Boer bucks were randomly assigned to and supplemented with diets containing 0, 4 or 8% BCPO on a dry matter basis, slaughtered and the LTL was subjected to a 7 day chill storage. Neither diet nor post mortem ageing influenced ($P > 0.05$) antioxidant enzyme activities, chemical composition and cholesterol. Diet had no effect on the carbonyl content, free thiol content, water-holding capacity, tenderness, pH and glycogen. Oil-supplemented goats had higher ($P < 0.05$) C18:1 *trans*-11, C18:3n-3 and C20:5n-3, carotenoid, tocopherol and redness, and lower thiobarbituric acid reactive substances values than the control goats. Post mortem ageing decreased ($P < 0.05$) shear force and oxidative stability of chevon. No significant ($P > 0.05$) changes were found in the proportion of individual fatty acids throughout storage. Total polyunsaturated fatty acids (PUFA) decreased while total saturated fatty acids increased as storage progressed. Dietary BCPO enhanced n-3 PUFA without compromising the quality attributes of chevon.

Key words: ageing, antioxidant, carbonyl, cholesterol, thiol.

INTRODUCTION

Ruminant meat contributes appreciable amount of fat to the human diet (Givens *et al.* 2006). In recent years, this fat component has been under the limelight due to its implications for human health and meat quality (Givens 2010). Ruminant meat contains relatively high amounts of saturated fatty acids (SFA) and low polyunsaturated fatty acids (PUFA) (Givens 2010; Najafi *et al.* 2012). The purported imbalance in the fatty acid composition of ruminant meat has been implicated in the incidence of chronic diseases such as cardiovascular diseases, diabetes and various forms of cancer (WHO 2003). This necessitates the need to modify the fatty acid profile of ruminant meat. Dietary unsaturated fats have been accentuated as an effective means of modifying the fatty acid profile of ruminant meat (Bessa *et al.* 2007; Kim *et al.* 2007; Jerónimo *et al.* 2009; Najafi *et al.* 2012). Nonetheless, dietary unsaturated fats can cause oxidative stress in animals (Slim *et al.* 1996). It can also predisposes the meat to protein and lipid oxidation (Renner *et al.* 1999) which can affect the sensory properties, shelf life and the nutritive value of the meat (Bekhit *et al.* 2013; Sola-Ojo *et al.* 2013). In addition, consumption

of lipid and protein oxidation products could affect consumers' health (Romeo *et al.* 1998; Soladoye *et al.* 2015). Thus, enhancing the unsaturated fatty acid profile of ruminant meat warrants concomitant enhancement of the antioxidant status to counteract lipid and protein oxidation. Synthetic antioxidants are effective in curbing oxidative spoilage (Fasseas *et al.* 2008; Adeyemi *et al.* 2013) but there are concerns about their safety in animals and humans (Karre *et al.* 2013). Besides, synthetic antioxidants are expensive and scarce, especially in low-income countries (Adeyemi & Olorunsanya 2012; Olorunsanya *et al.* 2012). Thus, the use of natural antioxidants could be a feasible alternative. Dietary supplementation of vegetable oils rich in antioxidants could suffice to manipulate the fatty acid profile of ruminant meat and prevent oxidative spoilage. Compared to lipid oxidation, little

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attention has been paid to the impact of dietary oil on protein oxidation in ruminant meat.

The fatty acid composition and the antioxidant contents of palm oil (Edem 2002) and canola oil (Ghazani *et al.* 2014) have been documented. Previous companion studies have shown that the blend of 80% canola oil and 20% palm oil did not have deleterious effects on the *in vitro* (Adeyemi *et al.* 2015a) and *in vivo* (Adeyemi *et al.* 2015c) rumen fermentation, feed intake and growth performance in goats (Adeyemi *et al.* 2015c). Thus, we propose that a blend of 20% palm oil and 80% canola oil will enhance the beneficial fatty acids in goat meat and prevent oxidative spoilage. Although, antioxidant vitamins such as vitamin E play a vital role in the regulation of oxidation of unsaturated fatty acids in a biological system, various protective endogenous antioxidant enzymes such as glutathione peroxidase, catalase and superoxide dismutase are also involved (Renner *et al.* 1999) in the maintenance of tissue oxidative stability. There is a dearth of information on the effect of blend of palm oil and canola oil on the activities of these antioxidant enzymes in goat meat. Thus the objective of this study was to determine the effect of dietary blend of canola oil (80%) and red palm oil (20%), on fatty acid composition, cholesterol, antioxidant vitamins and enzymes, physicochemical properties and protein and lipid oxidation of Longissimus thoracis et lumborum (LTL) muscle in goats during a 7 day post mortem ageing.

MATERIALS AND METHODS

Animal welfare

This study was conducted following the guidelines of the research policy of the Universiti Putra Malaysia on animal welfare and animal ethics.

Animals and diets

Twenty-four, 4–5-month-old Boer bucks with initial body weight of 20.54 ± 0.47 kg (mean $\pm$ standard deviation) were used in a feeding trial at ar-Raudhah Biotech Farm Pty. Ltd. Kuang, Selangor, Malaysia. Each animal was individually housed in a wooden slatted-floor pen equipped with drinking and feeding facilities. The goats were randomly assigned to and supplemented with diets containing 0, 4 or 8% blend of 80% canola oil and 20% palm oil (BCPO) on a dry matter basis. The feeding trial lasted 100 days after 2 weeks of adaptation. All diets were formulated to meet the nutritional requirements of growing goats following the recommendation of the NRC (2007). The experimental diets consisted of 50% forage (oil palm fronds) and 50% concentrate. The experimental diets were isonitrogenous and isocaloric. This was achieved by adjusting the ingredients in the concentrate portion of the diet. The proximate composition of diets and LTL muscle was determined following the AOAC (2007) methods. The neutral detergent fiber (NDF) and acid detergent fiber (ADF) contents of the diet were analyzed by the methods of Van Soest

et al. (1991). The chemical and fatty acid composition and antioxidant contents of the diets are presented in Table 1.

Slaughtering procedure and muscle sampling

The animals were fasted overnight with *ad libitum* access to water. The animals were weighed and slaughtered in a commercial abattoir according to the halal procedure as outlined in MS1500:2009 (Department of Standards, Malaysia 2009). After evisceration and carcass dressing, the right LTL between the sixth to 12th lumbar vertebrae was excised at 0, 1, 4 and 7 days post mortem and used for all analyses. About 30 g of the LTL was dissected on day 0 and divided into two parts. The first part was snap frozen and pulverized in liquid nitrogen, and assigned for the determination of pH and glycogen, antioxidants, fatty acids, cholesterol, lipid and protein oxidation at day 0. The second part was stored in a chiller at 4 ± 1 °C for the determination of drip loss. The carcasses were hung in the chiller at 4 ± 1 °C for subsequent sampling. On 1, 4 and 7 days post mortem, 40 g of LTL muscle was sampled from the carcasses and divided into two portions. The first portion (15 g) was snap frozen and pulverized in liquid nitrogen and assigned for the determination of pH, glycogen, antioxidants, fatty acids, cholesterol, lipid and protein oxidation. The second portion (25 g) was assigned for the determination of color, cooking loss and shear force.

Determination of muscle glycogen content

The glycogen content in muscle samples was determined using EnzyChrom™ Glycogen Assay kit (Cat#: E2GN-100; BioAssay, Hayward, CA, USA) following the manufacturer's procedure.

Determination of muscle pH

The pH of the muscle was read using a precalibrated portable pH meter (Mettler Toledo, AG 8603, Greifensee, Switzerland). Each pulverized sample (0.5 g) was homogenized (Wiggen Hauser® D-500, Hohenbrunn, Germany) for 30 s with 10 mL of a 5 mmol/L sodium iodoacetate, 150 mmol/L KCl solution to prevent further glycolysis. The pH of the resultant homogenate was measured at 20 ± 1 °C using the electrode attached to the pH meter.

Determination of antioxidant enzyme activity

Glutathione peroxidase (GPX) activity was measured using Cayman Glutathione Peroxidase Assay Kit (cat#: 703102.; Cayman Chemical Company, Ann Arbor, MI, USA), the superoxide dismutase (SOD) activity was measured using Cayman SOD Assay kit (cat#: 706002.; Cayman Chemical Company, Ann Arbor, MI, USA) while the catalase activity was measured using Cayman

Table 1 Chemical and fatty acid composition and antioxidant content of dietary treatments

Parameter	Levels of BCPO [†] (%)		
	0	4	8
Chemical composition (% dry matter (DM))			
DM	67.70	67.90	68.07
Crude protein	14.27	14.37	14.39
Ether extract	2.30	6.35	11.11
Organic matter	93.16	93.42	93.55
Nitrogen free extract	16.56	13.97	12.45
Acid detergent fibre	35.04	33.28	32.52
Neutral detergent fibre	63.52	62.67	62.06
Metabolizable energy, MJ/kg DM	11.59	11.61	11.62
Ca	1.02	1.05	1.04
P	0.52	0.54	0.54
Fatty acid composition, g/kg DM			
C12:0, lauric	0.01	0.03	0.04
C14:0, myristic	0.53	0.51	0.51
C16:0, palmitic	2.79	5.98	7.78
C16:1, palmitoleic	0.08	0.11	0.15
C18:0, stearic	0.56	1.12	1.43
C18:1n-9, oleic	3.82	14.87	26.32
C18:2n-6, linoleic	7.05	11.87	12.06
C18:3n-3, linolenic	1.06	2.61	4.13
n-6:n-3	6.66	4.54	2.92
Total fatty acids	15.83	37.09	52.27
Antioxidant contents (mg/kg DM)			
Total carotenoid	14.81	16.71	19.86
α-tocopherol	101.12	112.47	123.21
γ-tocopherol	10.22	34.55	49.17
δ-tocopherol	1.21	3.45	5.93

[†]Blend of 80% canola oil and 20% palm oil.

Catalase Assay Kit (cat#: 707002.; Cayman Chemical Company, Ann Arbor, MI, USA) following the manufacturer's procedure.

Determination of total carotenoid content

The carotenoid contents in feed and muscle samples were extracted and quantified following the method described by Okonkwo (2009). Two grams of each sample was homogenized with 10 mL acetone. The contents were stirred for 30 min and two 5 mL aliquots of acetone were used to rinse the flask and re-extract the residue. The extracts were pooled and 1 mL of deionized water was added. The mixture was transferred into 5 mL n-hexane and centrifuged at 3000 × g for 10 min. The absorbance of the hexane layer was read at 450 nm using a spectrophotometer (Secomam, Domont, France). The total carotenoid content was calculated by the following formula:

$$\text{Conc. } (\mu\text{g/g}) = A \times V \times 10^4 / (2592 \times W)$$

Where A = absorbance
V = Volume of n-hexane (mL)
W = Sample weight

Determination of tocopherol content

Extraction of tocopherol from feed and tissue samples followed the method of Kamal-eldin *et al.* (2000).

Quantification of tocopherol contents was done with Agilent 1200 series high-performance liquid chromatography (HPLC) as described by Pegg and Amarowicz (2009). The column used was C₃₀ YMCTM carotenoid (250 mm × 4.6 mm, internal diameter 5 μm) (YMC, Allentown, PA, USA). An isocratic mobile phase made up of 99% n-hexane and 1% isopropanol was used. The flow rate was 0.5 mL/min and the injection volume was 20 μL. UV detection was monitored at 295 nm. The isomers of tocopherol were quantified by comparing the peak area of sample with those of tocopherol standards in the HPLC controller software.

Determination of lipid oxidation

Lipid oxidation was measured as 2-thiobarbituric acid reactive substances (TBARS) using QuantiChromTM TBARS Assay Kit (cat#: DTBA-100., BioAssay Systems, Hayward, CA, USA) following the description of the manufacturer.

Determination of carbonyl and free thiol contents

Protein thiol was quantified according to Elman's method using 2, 2-dithiobis(5-nitropyridine) (DTNP) (Winterbourn 1990). The results were expressed as nmol/mg protein. The carbonyl content in muscles was

determined using Cayman protein carbonyl colorimetric assay kit (cat#: 10005020, Cayman Chemical Company, Ann Arbor, MI, USA) following the manufacturer's procedure. Carbonyl was expressed as nmol/mg protein.

Cholesterol

Cholesterol content in meat was determined using the method of Rudel and Morris (1973). One gram of pulverized meat sample was homogenized with 3 mL 95% ethanol and 2 mL 50% potassium hydroxide solution. The homogenates were incubated in a water bath at 60 °C for 10 min and cooled at room temperature. Five milliliters hexane and 3 mL deionized water was added to the homogenates, mixed for 20 s and allowed to settle at room temperature until complete phase separation. About 2.5 mL of the upper phase (hexane layer) was transferred into a clean glass tube and evaporated to dryness under nitrogen gas flow at 60 °C. The residue was re-suspended with 4 mL of o-phthalaldehyde reagent and kept at room temperature for 10, followed by adding 2 mL of concentrated sulfuric acid slowly, mixing and allowing to stand for an additional 10 min at room temperature. Cholesterol standards (Sigma L-4646) were prepared to make final concentrations of 0, 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 µg/mL cholesterol. The absorbance of the standards and samples were read at 550 nm using a spectrophotometer (Secomam, Domont, France). An absorbance-concentration calibration curve was plotted for the cholesterol standards and the cholesterol concentration of the sample was estimated from the derived equation.

Color

Meat color coordinates were determined using a Color Flex spectrophotometer (Hunter Lab, Reston, VA, USA) based on the International Commission on Illumination (CIE) Lab-values (also known as lightness (L*), redness (a*) and yellowness (b*)) with D65 illuminant and 10° standard observer, tristimulus values (X,Y,Z) and reflectance at specific wavelength (400–700) nm. The device was calibrated against black and white reference tiles prior to use. The meat samples were placed at the base of the color flex cup. For each sample, a total of three readings for L*, a* and b* values were recorded and then averaged.

Drip loss and cooking loss

Drip and cooking losses were measured according to the methods of Sabow *et al.* (2015b). For drip loss, the fresh meat samples obtained at day 0 were individually weighed (approximately 15 g) and recorded as initial weight (W1). The weighed samples were placed into polyethylene plastic bags, labeled, vacuum packaged and stored at 4 °C. After 1, 4 and 7 days post mortem, the samples were removed from the bags, gently blotted dried, weighed and recorded as W2.

Drip loss was calculated and expressed as the percentage of difference between initial and final weight of samples after storage divided by initial weight of samples.

$$\text{Drip loss (\%)} = [(W1 - W2)/W1] \times 100$$

For cooking loss, the samples were placed in water-impermeable polyethylene bags and vacuum packed. The samples were cooked in a pre-heated water bath set at 80 °C. When the internal temperature of the samples reached 80 °C as monitored using a stabbing temperature probe (HI 145-00 thermometer; HANNA® Instruments, Smithfield, RI, USA) inserted into the geometric center of the sample, the samples were removed from the water bath, equilibrated to room temperature and removed from the bags, blotted dry without squeezing and reweighed (W2). The percentage cooking loss was calculated using the following equation:

$$\text{Cooking loss (\%)} = [(W1 - W2)/W1] \times 100$$

Shear force

The samples used for cooking loss were used to determine the shear force values. Meat textural assessment was conducted using the TA.HD plus® texture analyzer (Stable Micro System, Surrey, UK) equipped with a Volodkevitch bite jaw. The equipment was calibrated at 5 kg for weight, 10 mm return distance for height and the blade speed was set at 10 mm/sec. The sample preparation was conducted following the procedure of Sazili *et al.* (2005). From each sample, three replicate blocks (1 cm height × 1 cm width × 2 cm length) were cut parallel to the direction of the muscle fibers and each block was sheared in the center perpendicular to the longitudinal direction of the fibers. Shear force was reported as the average peak positive force values for all blocks of individual sample.

Fatty acid (FA) analysis

The total lipids in feed and muscle (devoid of subcutaneous and intermuscular fats) samples were extracted with chloroform: methanol (2:1, v/v) mixture following the method of Folch *et al.* (1957) modified by Rajion *et al.* (1985). The extracted FAs were transmethylated to their fatty acid methyl esters (FAME) using 0.66N KOH in methanol and 14% methanolic boron trifluoride (Sigma Chemical Co., St. Louis, MO, USA) according to the methods of the AOAC (2007). Heneicosanoic acid was used as internal standard. The FAME was separated in a gas chromatograph (Model 6890 Agilent Technologies, Santa Clara, CA, USA) equipped with a flame ionization detector. The column used was fused silica capillary (Supelco SP-2560, 100 m, 0.25 mm internal diameter, 0.20 mm film thickness). Helium was the carrier gas and the split ratio after the FAME injection was 10:1.

The temperature of the injector and detector was programmed at 250 °C and 300 °C respectively. To facilitate optimal separation, the column temperature was set at 100 °C, held for 2 min and warmed to 170 °C at 10 °C/min, held for 2 min, warmed to 230 °C at 5 °C/min, and then held for 20 min. A reference standard mix (C4–C24 methyl esters; Sigma-Aldrich, Inc., St. Louis, MO, USA) and conjugated linoleic acid (CLA) standard mix (CLA *cis*-9, *trans*-11 and CLA *trans*-10, *cis*-12, Sigma-Aldrich, Inc., St. Louis, MO, USA) were used to determine recoveries and correction factors for the determination of individual FA composition.

Statistical analysis

The experiment followed a completely randomized design. Data obtained for all parameters were subjected to the GLM procedure of SAS (SAS 2003) where diet, post mortem ageing and interaction between diet and post mortem ageing were fitted as fixed effects in a repeated measure analysis of variance. Means were separated using Tukey Honestly Significant Difference test at a significance level of $P < 0.05$.

RESULTS

Fatty acids and cholesterol

Table 2 shows the effect of graded levels of BCPO and post mortem ageing on the FA composition and cholesterol content of LTL muscle from goats. Diet did not affect ($P > 0.05$) the proportion of C14:0, C16:0 and C18:0 in LTL muscle from goats. Similarly, the proportion of C18:1n-9, C16:1n-7, C18:2n-6 and C20:4n-6 did not differ ($P > 0.05$) between the treatments. The LTL muscle of oil-supplemented goats had higher ($P < 0.05$) C18:1 *trans*-11, C18:3n-3 and C22:5n-3 and lower ($P < 0.05$) n-6/n-3 compared with the control goats. Total SFA, total monounsaturated fatty acid (MUFA), total PUFA and PUFA/SFA were similar across dietary treatments. No significant changes ($P > 0.05$) were observed in the proportion of individual FA during post mortem ageing. The total SFA increased ($P < 0.05$), while the total n-6, n-3 and PUFA decreased ($P < 0.05$) as post mortem storage increased. Neither diet nor post mortem ageing influenced ($P > 0.05$) cholesterol content of LTL muscle in goats. The interaction between diet and post mortem ageing for the FA and cholesterol was not significant ($P > 0.05$).

Physicochemical properties and chemical composition

Effects of dietary BCPO and post mortem storage on the physicochemical properties and chemical composition of the LTL muscle are presented in Table 3. Diet did not affect ($P > 0.05$) the pH, glycogen, drip loss, cooking loss, tenderness, yellowness and lightness of the LTL muscle from goats. The LTL muscle of goats supplemented with 4 and 8% BCPO had higher ($P < 0.05$) redness

compared with the control goats. The pH, glycogen and shear force decreased ($P < 0.05$) while the drip loss increased as storage progressed. However, the pH and glycogen did not decline ($P > 0.05$) beyond day 1 post mortem. Post mortem storage did not affect cooking loss, lightness and yellowness of the LTL muscle. Neither diet nor post mortem storage influenced ($P > 0.05$) the chemical composition of the LTL muscle in goats.

Antioxidants, lipid and protein oxidation

Table 4 shows the effect of graded levels of BCPO and post mortem storage on antioxidants and the oxidative stability of the LTL muscle in goats. Neither diet nor post mortem ageing influenced ($P > 0.05$) the SOD, CAT and GPX activities. The concentration of total carotenoids, α and δ -tocopherol were higher ($P < 0.05$) while the TBARS value was lower ($P < 0.05$) in LTL muscle from goats supplemented with 4 and 8% BCPO compared to the control goats. Diet did not influence ($P > 0.05$) the concentration of γ -tocopherol in the LTL muscle from goats. The TBARS value increased ($P < 0.05$) while the concentration of total carotenoids and tocopherols decreased ($P < 0.05$) as ageing progressed. There was no significant ($P > 0.05$) interaction between diet and post mortem ageing for the antioxidant compounds and enzymes and TBARS value.

Diet had no effect ($P > 0.05$) on the free thiol and carbonyl contents. Regardless of dietary treatment, the free thiol content decreased ($P < 0.05$) while the carbonyl content increased ($P < 0.05$) from day 1 onward. The free thiol and the carbonyl contents observed on day 0 were not significantly different ($P < 0.05$) from those measured on day 1. Interaction between diet and post mortem ageing on the free thiol and carbonyl contents was not significant ($P > 0.05$).

DISCUSSION

Fatty acid composition

Dietary supplementation of BCPO was not a significant source of variation influencing the proportion of C14:0, C16:0 and C18:0 in the LTL muscle from goats. This observation suggests that dietary BCPO did not affect the activities of lipogenic enzymes required for the synthesis of medium and short chain FA (Kim *et al.* 2007). This observation is consistent with the findings of Jerónimo *et al.* (2009) who observed that various blends of soybean and linseed oils did not alter the proportion of the short and medium chain FA in the polar, neutral and total lipids of Longissimus lumborum muscle from lambs. Similarly, Karami *et al.* (2013) observed that dietary canola oil compared with palm oil did not affect the proportion of C14:0, C16:0 and C18:0 in the Longissimus lumborum muscle from goats.

Dietary BCPO enhanced the proportion of C18:1 *trans*-11 in LTL muscle from goats. The C18:1 *trans*-11 is an intermediate product of biohydrogenation of

Table 2 Effect of graded levels of BCPO[†] on fatty acid composition[‡] and cholesterol content of Longissimus thoracis et lumborum muscle in goats during postmortem ageing

Parameter	Levels of BCPO [†] (%)				Storage days				P-value			
	0	4	8	SEM	0	1	4	7	SEM	D	S	D*S
C14:0, myristic	2.09	2.04	1.91	0.18	2.01	2.32	2.40	2.62	0.17	0.780	0.062	0.310
C16:0, palmitic	23.33	21.73	23.29	1.73	22.76	23.34	24.00	24.03	1.23	0.409	0.073	0.231
C16:1n-7, palmitoleic	2.50	2.62	2.00	0.24	2.37	2.40	2.38	2.38	0.42	0.200	0.403	0.569
C18:0, stearic	20.88	20.84	20.07	0.79	20.59	21.40	22.00	22.20	0.86	0.313	0.090	0.274
C18:1n-9, oleic	23.67	23.36	22.88	1.42	22.37	22.37	22.43	22.48	1.35	0.216	0.712	0.437
C18:1 <i>trans</i> -11 vaccenic	1.21 [‡]	2.43 [‡]	3.15 [‡]	0.29	2.26	2.20	2.30	2.22	0.35	0.046	0.801	0.123
CLA <i>cis</i> -9 <i>trans</i> -11	1.02	0.92	1.27	0.17	1.05	1.06	1.20	1.15	0.23	0.504	0.290	0.397
CLA <i>trans</i> -10 <i>cis</i> -12	0.97	1.02	1.21	0.13	1.09	1.20	1.22	1.20	0.24	0.277	0.324	0.156
C18:2n-6, linoleic	12.31	12.35	13.40	0.65	12.69	11.75	10.94	11.00	0.76	0.109	0.064	0.490
C18:3n-3, linolenic	0.65 [‡]	1.03 [‡]	1.25 [‡]	0.11	0.97	0.90	0.82	0.80	0.14	0.006	0.085	0.239
C20:4n-6, arachidonic	7.30	7.04	6.37	0.43	6.90	6.52	6.23	6.00	0.34	0.136	0.076	0.560
C20:5n-3, eicosapentaenoic	1.72	1.95	1.94	0.28	1.87	1.59	1.30	1.11	0.30	0.463	0.201	0.657
C22:5n-3, docosapentaenoic	1.75 [‡]	2.00 [‡]	2.21 [‡]	0.27	1.99	1.75	1.70	1.65	0.22	0.007	0.312	0.731
C22:6n-3, docosahexanoic	1.00	1.29	1.48	0.21	1.26	1.20	1.20	1.16	0.22	0.082	0.410	0.175
ΣSFA	46.31	44.63	45.27	1.77	45.40 [‡]	47.06 ^{ab}	48.40 [‡]	48.69 [‡]	1.90	0.663	0.046	0.108
ΣMUFA	27.38	28.42	28.04	1.63	28.18	26.97	27.11	27.08	1.56	0.294	0.217	0.560
ΣPUFA	26.74	27.64	29.16	1.41	27.95 [‡]	25.97 [‡]	24.61 [‡]	24.07 [‡]	1.32	0.179	0.030	0.230
Σn-3	5.13	6.28	6.89	0.51	6.10 [‡]	5.44 ^{ab}	5.02 [‡]	4.72 [‡]	0.60	0.130	0.014	0.970
Σn-6	19.61	19.40	19.78	0.98	19.60 [‡]	18.27 ^{ab}	17.17 [‡]	17.00 [‡]	0.89	0.121	0.023	0.172
n-6:n-3	4.09 [‡]	3.08 [‡]	2.87 [‡]	0.31	3.34	3.35	3.42	3.60	0.41	0.013	0.721	0.123
UFA:SFA	1.16	1.25	1.22	0.11	1.21	1.24	1.12	1.05	0.13	0.570	0.263	0.237
PUFA:SFA	0.57	0.61	0.64	0.03	0.60	0.55	0.51	0.49	0.44	0.851	0.289	0.121
Total FA (mg/g of muscle)	29.80	32.30	32.51	4.24	31.51	32.42	31.03	34.05	5.11	0.434	0.793	0.250
Cholesterol (mg/100 g)	54.32	50.96	47.71	4.46	50.91	50.00	48.01	49.28	4.71	0.605	0.397	0.625

[†], [‡] means having different superscripts along the same row for each factor are significantly different ($P < 0.05$). ΣSFA = (C14:0 + C16:0 + C18:0), ΣMUFA = (C16:1 + C18:1 + C18:1 *trans*-11), ΣUFA = (C16:1 + C18:1 + C18:1 *trans*-11 + CLA *cis*-9 *trans*-11 + CLA *cis*-12 *trans*-10 + Σn-3 + Σn-6), ΣPUFA = (C18:1 *trans*-11 + CLA *cis*-9 *trans*-11 + CLA *trans*-10 *cis*-12 + Σn-3 + Σn-6), Σn-3 = (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3), Σn-6 = (C18:2n-6 + C20:4n-6), n-6:n-3 = (C18:2n-6 + C20:4n-6) ÷ (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3), UFA:SFA = (ΣUFA)/ΣSFA, PUFA:SFA = (ΣPUFA)/ΣSFA. Blend of 80% canola oil and 20% palm oil. SEM, standard error of mean; D, diet; S, storage; D*S, interaction between diet and storage. [‡]Data expressed as percentage of total identified fatty acids. CLA, conjugated linoleic acid; FA, fatty acid; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; UFA, unsaturated fatty acid.

C18:1n-9, C18:2n-6 and C18:3n-3 (Bauman *et al.* 2003). Therefore, the significant increase in the concentration of C18:1 *trans*-11 in the LTL muscle of oil-supplemented goats could be due to the higher total unsaturated FAs in their diets compared with that of the control goats. A companion feeding trial (Adeyemi *et al.* 2015c) showed that goats supplemented with 4 and 8% BCPO had higher intake of C18:1n-9, C18:2n-6 and C18:3n-3, suggesting that the unsaturated FA might have undergone partial biohydrogenation in the rumen. The current observation concurs with that of DePeters *et al.* (2001) who observed that dairy cows fed canola oil had higher C18:1 *trans*-11 in their milk compared with those fed control diet.

Dietary BCPO had no effect on the proportion of C18:2n-6 and C18:1n-9 in the LTL muscle from goats. This finding was unexpected given the

increase in the intake of C18:2n-6 and C18:1n-9 as BCPO increased in the diet (Adeyemi *et al.* 2015c). This observation could be due to ruminal hydrolysis and biohydrogenation of C18:1n-9 and C18:2n-6 (Bauman *et al.* 2003; Kim *et al.* 2007). This observation lends credence to the result of a companion *in vitro* study (Adeyemi *et al.* 2015a) which showed that the apparent biohydrogenation of C18:1n-9 increased as the level of BCPO increased in the substrate. In addition, the ruminal concentration of C18:2n-6 and C18:1n-9 tended to decrease with an increase in dietary BCPO (Adeyemi *et al.* 2015c).

Dietary BCPO enhanced the concentration of C18:3n-3 in the LTL muscle from goats. This observation reflects the concentration of C18:3n-3 in the diet. It also reflects the *in vitro* (Adeyemi *et al.* 2015a) and *in vivo* (Adeyemi *et al.* 2015c) ruminal concentration of C18:3n-3, suggesting

Table 3 Effect of BCPO[†] on physicochemical properties and chemical composition of Longissimus thoracis et lumborum muscle in goats during postmortem ageing

Parameter	Levels of BCPO [†] (%)				Storage days				P-value			
	0	4	8	SEM	0	1	4	7	SEM	D	S	D*S
Physicochemical properties												
Glycogen (mg/g)	1.89	1.89	1.91	0.14	1.94 ¹	0.72 ^b	0.70 ^b	0.71 ^b	0.11	0.759	0.039	0.711
pH	5.69	5.72	5.70	0.05	6.20 ¹	5.69 ^b	5.70 ^b	5.69 ^b	0.05	0.621	0.023	0.624
Cooking loss	32.05	29.40	32.56	1.48		30.95	27.59	33.39	0.96	0.176	0.120	0.067
Drip loss	7.02	6.84	6.81	0.99		5.92 ¹	1.20 ^b	0.53 ^b	0.68	0.131	0.048	0.090
Shear force	1.02	1.10	0.98	0.18		1.15 ¹	0.88 ^b	0.80 ²	0.03	0.166	0.034	0.700
Lightness (L*)	32.33	33.17	31.80	2.47		32.26	35.45	34.40	1.75	0.925	0.433	0.791
Redness (a*)	12.96 ^b	14.32 ¹	15.54 ¹	0.77		12.64 ¹	11.35 ^b	11.04 ^b	0.41	0.037	0.645	0.648
Yellowness (b*)	13.27	12.95	12.48	0.57		12.37	13.49	14.04	0.72	0.297	0.709	0.621
Chemical composition (%)												
Moisture	74.81	74.79	73.45	0.65		75.87	75.30	74.24	0.65	0.112	0.433	0.297
Protein	19.64	19.18	20.80	1.34		16.23	16.80	17.50	1.64	0.401	0.791	0.645
Crude fat	4.16	4.33	4.50	1.24		4.66	4.34	4.70	1.23	0.982	0.137	0.106
Ash	1.48	1.69	1.24	0.55		1.50	1.56	1.59	0.70	0.851	0.106	0.921

¹, ^b ²Means having different superscripts along the same row for each factor are significantly different ($P < 0.05$). [†]Blend of 80% canola oil and 20% palm oil. SEM, standard error of mean; D, diet; S, storage; D*S, interaction between diet and storage

Table 4 Effect of BCPO[†] on antioxidant status and lipid and protein oxidation of Longissimus thoracis et lumborum muscle in goats during post mortem ageing

Parameter	Levels of BCPO [†] (%)				Storage days				P-value			
	0	4	8	SEM	0	1	4	7	SEM	D	S	D*S
Carotenoid (mg/kg)	0.12 [‡]	0.23 [‡]	0.40 [‡]	0.08	0.35 [‡]	0.23 [‡]	0.17 [‡]	0.12 [¶]	0.08	0.034	0.004	0.742
α-tocopherol (mg/kg)	2.29 [‡]	3.45 [‡]	4.43 [‡]	0.31	4.14 [‡]	3.57 [‡]	3.23 [‡]	2.62 [‡]	0.11	0.021	0.002	0.106
δ-tocopherol (mg/kg)	0.66 [‡]	0.75 [‡]	1.02 [‡]	0.05	0.78	0.75 [‡]	0.70 [‡]	0.67 [‡]	0.04	0.001	0.045	0.561
γ-tocopherol (mg/kg)	0.06	0.07	0.09	0.01	0.09	0.08	0.06	0.05	0.02	0.292	0.993	0.751
Glutathione peroxidase [‡]	90.52	81.50	72.64	12.32	88.84	84.71	79.67	75.22	7.25	0.561	0.289	0.107
Superoxide dismutase [§]	2.29	2.63	2.83	0.44	2.55	2.62	1.98	2.14	0.61	0.212	0.435	0.156
Catalase [¶]	1772.1	1700.6	1680.4	40.20	2010.2	1987.7	1860.0	1801.9	44.15	0.316	0.222	0.167
TBARS (mg MDA/kg)	0.21 [‡]	0.12 [‡]	0.06 [‡]	0.09	0.13 [¶]	0.26 [‡]	0.35 [‡]	0.55 [‡]	0.16	0.010	0.002	0.561
Carbonyl (nmol/mg protein)	3.20	3.21	3.26	0.02	1.20 [‡]	1.26 [‡]	3.54 [‡]	5.86 [‡]	0.06	0.579	<.001	0.338
Free thiol (nmol/mg protein)	53.98	54.05	54.29	0.99	54.19 [‡]	54.14 [‡]	46.75 [‡]	42.01 [‡]	0.89	0.103	0.001	0.160

[‡], [‡] [¶] [§]Means having different superscripts along the same row for each factor are significantly different ($P < 0.05$). [†]Blend of 80% canola oil and 20% palm oil. [‡]Glutathione peroxidase activity is expressed as nmoles nicotinamide adenine dinucleotide glycohydrolase oxidized /min/mg protein. [§]One unit of superoxide dismutase activity corresponds to the amount of enzyme needed to exhibit 50% dismutation of the superoxide radical. [¶]Catalase activity is expressed as nmol. H₂O₂/min/mg protein. SEM, standard error of mean; D, diet; S, storage; D*S, interaction between diet and storage; TBARS, 2-thiobarbituric acid reactive substances, MDA, malondialdehyde

that some C18:3n-3 escaped rumen biohydrogenation. This observation agrees with that of Karami *et al.* (2013) who observed an increase in the concentration of C18:3n-3 in the Longissimus lumborum muscle from goats fed canola oil compared with those fed palm oil. The increase in the concentration of C18:3n-3 was

accompanied by an increase in the concentration of C22:5n-3, suggesting *in vivo* elongation of C18:3n-3. Similar increase in C18:3n-3 and its long chain derivatives following dietary supplementation of vegetable oil blends has been reported (Kim *et al.* 2007; Jerónimo *et al.* 2009).

The increase in the concentration of n-3 FA in the LTL muscle from goats supplemented with 4 and 8% BCPO was accompanied by a decrease in the n-6/n-3 ratio. This observation reflects the dietary n-6/n-3 ratio. Raes *et al.* (2004) asserted that the n-6/n-3 ratio of meat depends to a large extent on the dietary C18:2n-6/C18:3n-3 ratio. Regardless of the treatment, the n-6/n-3 ratio of the LTL muscle of goats was within the normal range (< 4) specified for a healthy diet (WHO 2003).

There was no difference in the cholesterol content of LTL muscle due to diet. This could be due to the similar muscle SFA across dietary treatments. As found in the current study, the cholesterol content of Longissimus dorsi of bulls fed different oils was similar to those fed the control diet (Oliveira *et al.* 2012).

Post mortem ageing did not affect the proportion of individual FA. However, the total n-3 and n-6 PUFA decreased while the total SFA increased as post mortem storage progressed. This observation could be due to the increase in lipid oxidation over storage. Similarly, Mahecha *et al.* (2009) observed a reduction in the n-3 and n-6 PUFA and an increase in SFA in beef Longissimus aged for 14 days.

Physicochemical properties and chemical composition

Diet had no effect on the muscle pH and glycogen. This observation could be due to the similarity in the metabolizable energy of the diets. Dietary energy affects muscle glycogen at slaughter which is an important factor influencing post mortem muscle glycogen and pH (Lawrie & Ledward 2006). The muscle pH and glycogen observed on day 0 (45 min post mortem) was higher than those observed on other post mortem storage days. The decline in muscle glycogen and pH could be due to post mortem glycolysis, which necessitates the conversion of glycogen to lactic acid, thus lowering the pH (Adeyemi & Sazili 2014; Salwani *et al.* 2015). The ultimate pH (pH at 24 h post mortem) observed in the current study is consistent with the ultimate pH of Longissimus dorsi muscle from different Omani breeds of goats (Kadim *et al.* 2004). The similarity in the muscle glycogen and pH observed on days 1, 4 and 7 indicates that post mortem glycolysis was completed by 24 h post mortem.

Fatty acids are involved in various nutritional, technological and eating aspects of meat quality (Wood *et al.* 2008). Thus, accessing the role of dietary fat on the physicochemical properties of meat is essential.

Tenderness is one of the most important meat quality parameters and appears to be sought at the expense of flavor or color (Lawrie & Ledward 2006). Dietary BCPO had no effect on shear force value of the LTL muscle from goats. The similarity in tenderness is expected since no changes were observed for the intramuscular fat of the LTL muscle across the dietary treatments. High

intramuscular fat increases the expansion of fat cells in the perimysial connective tissue which forces muscle bundles apart, thus opening up the muscle structure (Lawrie & Ledward 2006). The current observation is in agreement with that of Mir *et al.* (2003) who observed no difference in the shear force value of Longissimus dorsi muscle of cattle fed 3 and 6% sunflower oil compared to those fed the control diet. In contrast, Zinn (1989) observed that steers fed 4 and 8% animal-vegetable fat blend had lower shear force compared to the control steers.

Regardless of diet, the shear force value decreased over storage. This observation could be due to the degradation of myofibrillar proteins in the course of ageing. Similar findings were found in chevon (Sabow *et al.* 2016) and beef (Marino *et al.* 2013). During post mortem, the accumulation of Ca^{2+} into the muscle sarcoplasm leads to the activation of μ -calpain which in turn destabilizes the intact myofibrillar structure by degrading myofibrillar proteins involving titin, filamin, troponin-T and desmi (Lomiwes *et al.* 2014).

Water-holding capacity is an important meat quality trait because it affects the appearance of meat prior to cooking, its characteristics during cooking and juiciness on mastication (Lawrie & Ledward 2006; Farouk *et al.* 2014). Diet had no effect on the drip loss of LTL muscle from goats. This finding agrees with that of Mir *et al.* (2003) who observed that the drip loss in steers fed 3 and 6% sunflower oil with vitamin E was similar to that of the control steers (without oil) and those fed sunflower oil without vitamin E. Dietary manipulation of muscle fatty acid can induce lipid and protein oxidation. Protein oxidation can alter the functionality of myofibrillar protein via polymerization and aggregation and these can affect the physicochemical properties of meat, such as water-holding capacity (Huff-Lonergan & Lonergan 2005). Irrespective of dietary treatment, the drip loss increased over storage. This observation could be due to the degradation of the myofibrillar proteins which affects the ability of the muscles to hold water (Lawrie & Ledward 2006).

Cooking loss is an agglomeration of liquid and soluble matters lost from meat during cooking (Lawrie & Ledward 2006). Neither diet nor post mortem ageing affected cooking loss of the LTL muscle. This could be due to the overriding effect of heat denaturation of myofibrillar protein which masked the effects of other factors.

Color is an important meat quality attribute being the first quality attribute that consumers consider and also acts as indispensable tool for the identification and selection of meat (Lawrie & Ledward 2006; Adeyemi & Sazili 2014). The higher redness value observed in the LTL muscle from goats supplemented with 4 and 8% BCPO could be due to the higher antioxidant content in their muscle which enhanced oxidative stability. Redness is the most significant color parameter for the assessment of lipid oxidation and the oxidation of myoglobin to

metmyoglobin (Lynch *et al.* 1999; Zakrys *et al.* 2008; Adeyemi *et al.* 2015b). The decrease in redness as post mortem storage progressed could be due to the increase in myoglobin oxidation. This lends credence to the decrease in the concentration of total carotenoids and tocopherols. This finding is consistent with that of Sabow *et al.* (2015b) who observed a decline in the redness of Longissimus lumborum muscle from goats during a 14 day chill storage. Similarly, Adeyemi *et al.* (2015b) observed a decrease in the redness of Semimembranosus muscle in goats during an 8 day chill storage. Neither diet nor post mortem ageing influenced L* and b* of the LTL muscle from goats.

Diet had no effect on the proximate composition of the LTL muscles from goats. Similarly, bulls (Oliveira *et al.* 2012) and goats (Najafi *et al.* 2012) fed different dietary oils did not show differences in chemical composition compared to those fed control diets. In contrast, Zinn (1989) observed that steers fed 4 and 8% animal-vegetable fat blend had higher intramuscular fat compared to the control steers. The similarity in intramuscular fat observed in the current study could be due to the similarity in the metabolizable energy of the diets. Feed energy supplied in excess of the requirement for muscle deposition can increase the concentration of intramuscular fat in muscle (Wood *et al.* 2008).

Antioxidant status

Living cells can maintain the balance between antioxidants and pro-oxidants and the pool of antioxidants required for cellular protection can be sustained via diet (Thomas 1995). However, such a mechanism is not possible in food systems such as post mortem muscle, because it cannot repair itself (Maraschiello *et al.* 1999). Thus, storage of meat necessitates the enhancement of antioxidant status to attenuate lipid oxidation. Dietary supplementation of antioxidants is an effective means of enhancing the oxidative stability of raw and cooked meat (Renerre *et al.* 1999). Dietary BCPO increased the concentration of total carotenoids, α and δ -tocopherol in the LTL muscle from goats. This observation mirrored the dietary antioxidant contents. This finding could be due to the increase in the fat content of the diets. Both carotenoid and tocopherol are fat soluble compounds; thus, the presence of fat would enhance their absorption and deposition. The increase in the tocopherol content of the LTL muscle following dietary supplementation of BCPO is in tandem with the findings of Jensen *et al.* (1998) who observed an increase in vitamin E content of Longissimus muscle in pigs fed rapeseed oil compared to those fed control diet. Similarly, pigs fed 5 and 10% canola oil had higher vitamin E content in muscle and liver compared to those fed control diets (Soler-Velasquez *et al.* 1998).

Notwithstanding the increase in the concentration of n-3 FA, dietary BCPO reduced the TBARS value in LTL

muscle from goats. This observation could be due to the increase in the concentration of total carotenoids and tocopherols. The observed reduction in lipid oxidation is akin to the findings of Karami *et al.* (2011) who observed a lower TBARS value in Longissimus muscle of goats fed vitamin E, *Adrographis paniculata* and tumeric compared to those fed the control diet.

Dietary BCPO had no effect on the SOD, CAT and GPX activities in LTL muscle from goats. This finding could be related to the level of antioxidant compounds in the muscle. Increase in the activities of antioxidant enzymes during ageing could be due to feedback mechanisms in response to lipid oxidation (Rodriguez-Martinez & Ruiz-Torres 1992). Contrary to the present observation, Renerre *et al.* (1999) found that the Pectoralis major muscle of turkey fed soy oil and rapeseed oil had significantly higher CAT and GPX activities but similar SOD activity compared to those fed tallow. Luostarinen *et al.* (1997) observed higher SOD activity in rats fed 7% fish oil compared to those fed 7% butter. Venkatraman and Pinnavaia (1998) reported that the tissue of rats fed fish oil had higher CAT activity compared to those fed safflower or beef fat. Also, Lemaitre *et al.* (1997) observed that n-3 PUFA enhanced GPX activity. The increase in the antioxidant enzyme activities observed in the aforementioned studies was attributed to enhanced enzyme synthesis in response to oxidative stress caused by high concentrations of unsaturated FAs. Thus, the higher tocopherols and total carotenoids in the LTL muscle from goats fed 4 and 8% BCPO substantiates their inability to increase antioxidant enzyme activities despite the increase in the proportion of n-3 PUFA.

There was a significant decrease in the concentration of tocopherols and total carotenoids as storage continued. The decrease corresponds to the increase in the TBARS value as post mortem storage progressed. This observation is consistent with that of Adeyemi *et al.* (2015b) who observed a decrease in the concentration of tocopherol and total carotenoids and an increase in TBARS value during an 8 day post mortem ageing of Semimembranosus muscle from goats. Similarly, Sabow *et al.* (2015a) observed an increase in TBARS value during a 14 day chill storage of Longissimus lumborum muscle from goats.

Post mortem chill storage did not affect the antioxidant enzyme activities in the LTL muscle from goats. The stability of GPX and CAT is consistent with the findings of Renerre *et al.* (1996) who observed that the CAT and GPX activities in beef observed on day 1 was similar to those observed on day 8 post mortem. The stability of SOD activity is also consistent with the finding of Renerre *et al.* (1999) who observed a non-significant decrease in the SOD activity in Pectoralis major muscle of turkey as storage progressed. DeVore and Greene (1982) reported that the GPX activity of beef Semitendinosus measured at 2 h post mortem was similar to those observed at 2 days post mortem. Contrary to

the present observation, Renner *et al.* (1999) observed a significant decrease in GPX and CAT activities in the Pectoralis major muscle of turkey as storage progressed. DeVore and Greene (1982) posited that the decrease in GPX activity in meat is a function of the rejuvenation of glutathione (GSH) with GSH reductase in the presence of nicotinamide adenine dinucleotide glycohydrolase.

Free thiol and carbonyl are important parameters for accessing protein oxidation in tissues (Lund *et al.* 2008). Dietary BCPO did not affect the free thiol and carbonyl contents in the LTL muscle from goats. Similarly, Lund *et al.* (2008) observed that dietary soybean oil (2%) compared with the control diet had no effect on the free thiol group and carbonyl content of porcine Quadriceps femoris subjected to a chill storage for 7 days. The implication of this finding is that the increase in the concentration of tocopherols and total carotenoids in the LTL muscle from oil-supplemented goats was insufficient to curb protein oxidation. The decrease in the free thiol content and the increase in carbonyl content over storage are consistent with the findings of Lund *et al.* (2008). Similarly, Sabow *et al.* (2016) observed that the concentration of free thiol decreased during a 14-day chill storage of chevon.

Conclusion

The results of this study demonstrate that dietary blend of 80% canola seed oil and 20% palm oil enhanced the proportion of n-3 FA in the LTL muscle from goats. The oil blend also improved redness and lipid oxidative stability of the LTL muscle. However, the muscle cholesterol, nutrient composition, water-holding capacity, tenderness and activities of antioxidant enzymes and protein oxidation were not affected. Further studies on the effect of dietary blend of 80% canola seed oil and 20% palm oil on the nutritional and physico-chemical properties of other muscles are suggested.

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