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The effect of fibre source in finishing diets on lamb performance and muscle fatty acid composition

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Abstract

The objective of this study was to evaluate the effect of dietary fibre sources on the performance, carcass characteristics, and muscle fatty acid composition of finishing lambs. Fifty-eight Merino ram lambs were randomly allocated to nutritionally similar treatment diets differing mainly in fibre source: i.e., lucerne hay (LH - 350 g/kg), soybean hulls (SH - 229 g/kg), maize stover (MS - 195 g/kg) and *Eragrostis tef* (ET - 216 g/kg). The animals were slaughtered on day 78. Growth performance and carcass characteristics were determined. Meat samples were taken from the *Musculus longissimus dorsi* for fatty acid determination. Lucerne hay resulted in a higher dry matter intake (1604 g/day), metabolizable energy intake (14.43 MJ/day), average daily gain (315 g/day), cold carcass weight (24.93 kg), and dressing percentage (47.48%). Total n-6 muscle content (7.99%) was low following lucerne hay inclusion compared only to treatment ET (10.01%). Lucerne hay resulted in a higher α-linolenic acid (0.79%) and total n-3 (1.24%) muscle tissue content which resulted in a decrease in the n-6:n-3 ratio (6.42). In the present study it was proven that fibre source in nutritionally-similar diets can influence growth performance and the fatty acid profile of lamb meat.

Keywords: growth, lucerne hay, roughage, sheep, unsaturated
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Introduction

Ruminant meat is perceived as unhealthy, and the possibility exists of having undesirable effects on human health. Coronary heart disease is an example of one risk factor associated with the consumption of high levels of saturated fatty acids (SFA), due to the fact that these fatty acids are correlated with increased serum low-density lipoprotein (LDL) concentrations (Keys, 1997). The replacement of saturated fats with polyunsaturated fatty acids (PUFA) implies that saturated fat causes disease in humans (Forouhi *et al*., 2018). Wood *et al*. (2004) reported that conjugated linoleic acid (CLA:C18:2*cis*-9,*trans*-11) can possibly have important human health advantages, such as a decrease in developing cancer, cardiovascular disease, and diabetes. There is proof that CLA supplementation increases serum high-density lipoprotein (HDL) cholesterol (Zhao *et al*., 2009) and improves blood pressure by increasing adiponectin and endothelial nitric oxide synthase activity (Basak & Duttaroy, 2020). There is also proof that elevated blood CLA content in humans reduces the overall risk of heart failure by preventing the development of cardiac hypertrophy (Wannamethee *et al*., 2018). Human CLA studies focusing on cardiovascular disease risk factors are however inconclusive (Dilzer & Park, 2012), as there are still known and unknown consequences with the consumption thereof (Basak & Duttaroy, 2020). McDonald *et al*. (2011) added that the well-being of humans can benefit from the consumption of PUFAs – especially the beneficial effects exerted by n-3 fatty acids. Ruminant fat, compared to non-ruminant fat, is higher in SFA, but lower in the ratio of PUFA:SFA because dietary unsaturated fatty acids (UFA) undergo extensive hydrogenation in the rumen (French *et al*., 2000). French *et al*. (2000) stated that ruminant fats are amongst the richest natural sources of CLA isomers.

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The recommendation to limit dietary SFA intake has persisted despite mounting evidence to the contrary. It was claimed that saturated fats, along with dietary cholesterol, were the principal causes of cardiovascular disease, whereas more rigorous clinical-trial data in the 1970s and 1980s did not provide support for this hypothesis (Astrup *et al*., 2021). Saturated fatty acids are necessary to assimilate calcium into the skeletal structure, adds to the strength of bones, and contributes to more than 50% of the rigidness and durability of cell membranes in humans (Fallon & Enig, 2001). Even though SFAs can pose some health benefits, an unbalanced consumption is not an ideal situation.

The protection of dietary oil from rumen fermentation and the increase in cereals in feed presented to animals are the two primary methods in animal nutrition that were used to increase the beneficial fatty acid content of ruminant meat (Wood *et al*., 2004). The amount of lipid source included (Zhao *et al*., 2009) and the composition thereof (Manso *et al*., 2009) are also important factors to be considered. Other factors that are applied within the present study are the type of forage (Wood *et al*., 2008) and forage maturity. Forages with a shorter rumen transit time limit the opportunities for microbial biohydrogenation (Wood *et al*., 2008).

There is a vast array of literature available that supports strategies to improve or manipulate the fatty acid content of animal meat, especially through dietary means. There are studies that have focused either on the roughage:concentrate ratio (Kucuk *et al*., 2001; Demirel *et al*., 2006; Marino *et al*., 2006) or source of roughage (French *et al*., 2000; Wood *et al*., 2008) and its effect on ruminant product fatty acid composition. Fibre sources included in diets with comparable neutral detergent fibre (NDF) content (similar NDF:concentrate ratio) and their effect on ruminant meat fatty acid composition is not referred to in the literature. The hypothesis was that a high-quality fibre source (lucerne hay) fed to finishing lambs could decrease the effect of biohydrogenation of UFAs in the rumen and potentially increase their content in lamb meat. The objective of this study was therefore to evaluate the effect of different fibre sources in nutritionally-similar finishing diets on the growth performance, carcass characteristics, and fatty acid composition of lamb muscle tissue.

Materials and methods

A production study was conducted just outside of Bloemfontein (Free State province, South Africa) on the University of the Free State's experimental farm (Paradys) from May to July, 2018. The average minimum and maximum temperature and humidity during the trial ranged from 7.29 °C and 19.20 °C, and 27.39% and 55.90%, respectively. All procedures conducted during this study were approved by the Interfaculty Animal Ethics Committee for Animal Experimentation at the University of the Free State (Animal Experiment No. UFS-AED2018/0004).

Fifty-eight Merino ram lambs (27.63 ± 2.05 kg; mean ± SD) of four months of age (112 ± 14 days; mean ± SD) from a homogeneous group were randomly allocated to four different treatments [n = 15 (lucerne hay); n = 15 (soybean hulls); n = 14 (maize stover); n = 14 (*Eragrostis tef*) – randomised trial design was used and lambs were placed individually (n = repetitions per treatment) in pens (1.404 m^2) in a closed but well-ventilated building. The production study was conducted over a period of 78 days (including a ten-day adaptation period). The treatment diets were fed to the lambs and water were available on an *ad libitum* basis. All animals were vaccinated against pulpy kidney and treated to be free of internal and external parasites.

Commonly accessible feeds used for lamb finishing diets in South Africa were used in this study. Four treatment diets were formulated to be nutritionally comparable [similar total fibre (NDF):concentrate ratio], differing mainly in the fibre source included, i.e., lucerne hay (LH – high quality), soybean hulls (SH – high quality), maize stover (MS – low quality) and *Eragrostis tef* (ET – low quality). Rations were presented to the lambs in pelleted form. The chemical composition of the four fibre sources is presented in Table 1.

Table 1 Analysed chemical composition of the four fibre sources added to the finishing diets of rams

	Primary source of fibre in diet			
	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*
Nutrient composition (g/kg DM)				
Dry Matter	890	900	800	900
Organic matter	910	950	930	928
Ash	90	50	70	72
Crude protein	242	135	50	47
Non-structural carbohydrate	188	227	167	98
Neutral detergent fibre	457	563	700	767
Acid detergent fibre	360	460	440	332
Ether extract	23	26	13	16

The physical and chemical composition of the treatment diets are presented in Table 2. Citrus pulp was used as a filler to formulate for similar NDF content between diets as maize stover, for example, contains a higher NDF content than lucerne hay (Table 1). Soluble non-starch polysaccharides, such as pectins, which are contained in citrus pulp (Bampidis & Robinson, 2006) are not fermented to lactate in the rumen due to the galacturonic acid structure, and therefore provide buffering to the rumen because cellulose digestion is largely unaffected (Van Soest *et al*., 1991). Animals were adapted to each respective treatment diet for 10 days and were presented with their respective experimental diets on an *ad libitum* basis for the remainder of the experimental period (68 days). Water was freely available to the animals during the whole production study. Full stomach live weight of all animals were determined at the onset and end of the production study.

Table 2 Physical and analysed chemical composition of the four composite dietary treatments fed to finishing rams (NDF:concentrate ratio of 25:75)

	Primary source of fibre in diet			
	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*
Raw material (g/kg as is)				
Maize meal	550	550	550	550
Lucerne hay	350	-	-	-
Soybean hulls	-	229	-	-
Maize stover	-	-	195	-
Eragrostis teff hay	-	-	-	216
Soybean oil	0.3	-	2.9	2.4
Citrus pulp	69	177	201	179
Urea	6.6	14.3	20.7	19.1
Limestone	2.5	6.7	7.2	8.5
Ammonium chloride	7.5	7.5	7.5	7.5
Mono-calcium phosphate	7.0	8.5	8.5	9.2
Salt	5.0	5.0	5.0	5.0
Mineral and vitamin premix[1]	2.5	2.5	2.5	2.5
Nutrient composition (g/kg DM)				
Dry Matter	926	924	931	933
Organic matter	923	916	908	906
Ash	78	84	92	94
Crude protein	153	156	151	154
Non-structural carbohydrate	487	493	486	455
Neutral detergent fibre	245	232	240	265
Acid detergent fibre	176	197	189	181
Ether extract	37	35	31	32

[1] Vitamins, minerals, and additives: vitamin A = 3 500 000 IU/ton, vitamin B1 = 2.0 g/ton, cobalt = 1.5 g/ton, ferrous iron = 30.0 g/ton, iodine = 1 g/ton, manganese = 30.0 g/ton, selenium = 0.3 g/ton, zinc = 65.0 g/ton, Salinomycin = 18 mg/ton, ZnBac = 25.0 g/ton, Toxibind = 500.0 g/ton

Treatment diet lipid content and fatty acid composition are represented in Table 3. Palmitoleic (C16:1c9), oleic (C18:1c9; n-9), and vaccenic acid (C18:1t11) of the LH treatment were all lower thank in other treatments. In contrast, the linoleic acid (C18:2c9,12; n-6) and α-linolenic acid (C18:3c9,12,15; n-3) contents of the LH treatment were higher than in other treatments. The difference in the individual fatty acid content of the LH treatment compared to the other treatments was associated with the total MUFA, PUFA, total n-6, total n-3, as well as fatty acid ratio (PUFA:SFA and n-6:n-3). The lower n-6:n-

3 ratio of the LH treatment is due to the higher content of n-3 that lucerne hay contains (Mitchell *et al*., 1991), although it has the highest n-6 content of the different fibre sources (Table 3).

Table 3 The lipid content and fatty acid composition of composite treatment diets fed to finishing ram lambs

	Primary source of fibre in diet			
Fatty acid (% of total fatty acids)	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*
Proximate analysis:				
Feed lipid content (% DM)	3.71	3.50	3.11	3.17
Saturated fatty acids:				
Myristic (C14:0)	0.58	0.37	0.56	0.53
Palmitic (C16:0)	27.39	31.08	30.19	30.77
Stearic (C18:0)	6.20	7.83	7.57	7.79
Monounsaturated fatty acids:				
Palmitoleic (C16:1c9)	0.00	0.86	1.01	1.06
Oleic (C18:1c9; n-9)	23.02	28.13	33.65	33.13
Vaccenic (C18:1t11)	1.87	3.30	3.84	3.66
Polyunsaturated fatty acids:				
Linoleic (C18:2c9,12; n-6)	31.62	21.63	16.05	15.52
α-Linolenic (C18:3c9,12,15; n-3)	3.49	1.20	0.98	0.99
Long-chain polyunsaturated fatty acids:				
Arachidonic (C20:4c5,8,11,14; n-6)	-	-	-	-
EPA (C20:5c5,8,11,14,17; n-3)	0.10	0.00	0.00	0.00
DPA (C22:5c7,10,13,16,19; n-3)	0.04	0.00	0.00	0.11
Total fatty acids:				
Saturated fatty acid	36.20	41.31	40.60	41.49
Monounsaturated fatty acid	25.75	32.93	39.00	38.37
Polyunsaturated fatty acid	38.05	25.76	20.40	20.14
n-6	34.42	24.57	19.41	19.05
n-3	3.63	1.20	0.98	1.10
Fatty acid ratios:				
n-6:n-3	9.48	20.56	19.74	17.41
PUFA:SFA	1.05	0.62	0.50	0.49

All lambs (48.84 ± 3.92 kg live weight; mean ± standard deviation, SD) were slaughtered at the end of the production study. Carcasses were left to cool down to 2–4 ºC, whereafter carcass characteristics were measured. Dressing percentage was calculated using cold carcass weight.

The left side of the carcass was used for evaluation after it was split between the 12th and 13th thoracic vertebra. Fat depth was measured 45 mm and 110 mm from the mid dorsal line with an electronic digital calliper (Omni-Tech). The area of the *Musculus longissimus dorsi* was traced onto transparent film and scanned using a video image analysis system (Soft Imaging System: analysis® 3.0) after it was calibrated with a scale bar.

Meat tenderness was evaluated using the *Musculus longissimus* cut from the left 11th to 13th rib of the same carcass, vacuum sealed, and stored frozen (-20 °C). These samples were later thawed at 4 °C and prepared by direct heat according to an oven-broiling method (AMSA, 2015). *Musculus longissimus* was placed in an electric oven set on “broil” (260 °C) and cooked to an internal temperature of 35 °C, turned over, and finished until the core temperature reached 70 °C. Four cylindrical muscle samples (12.5 mm core diameter) per repetition were sheared perpendicular to fibre direction in the centre of each core using a Warner–Bratzler shear force device, where the resultant value represents the average of the peak force (kg). The crosshead speed was 200 mm/min. Shear force was measured after the cuts were cooled down to room temperature.

The fatty acid content of feed and *Musculus longissimus dorsi* samples were determined. Total lipid from the treatment diets was determined by means of the Soxhet extraction method (No. 920.39), using petroleum ether (40–65°C boiling point) as a solvent (AOAC, 2003). Total lipid from *Musculus longissimus dorsi* samples was quantitatively extracted (Folch *et al*., 1957) using chloroform and methanol in a ratio of 2:1 with butylated hydroxytoluene (0.001%) added to the mixture. The fat extracts were dried overnight in a rotary evaporator oven at 50 °C and stored in a polytope vial under a blanket of nitrogen (–20 °C) before fatty acid analyses were conducted. Total *Musculus longissimus dorsi* lipid content was expressed as a percentage of total muscle weight.

Thirty milligrams of each muscle lipid aliquot were converted to methyl esters by base-catalysed transesterification using sodium methoxide (0.5 M solution in anhydrous methanol) for 2 h at 30 °C (Alfaia *et al*., 2007). Fatty acid methyl esters (FAME) from feed and muscle lipid were quantified using a Varian 430 flame ionization gas chromatograph with a fused silica capillary column (Chrompack CPSIL 88; 100 m length, 0.25 mm ID, 0.2 µm film thicknesses). Analysis was performed using an initial isothermic period of 40 °C for 2 min; temperature was then increased at a rate of 4 °C/minute to 230 °C, whereupon an isothermic period was followed for 10 min with a split ratio of 100:1. Fatty acid methyl esters (1 µl) were injected into the column using a Varian CP 8400 autosampler. Hydrogen functioned as the carrier gas and nitrogen as the makeup gas. The retention time of each FAME peak was used to identify fatty acids by comparing fatty acid standard peaks.

Fatty acids were expressed as the proportion of each individual fatty acid (percentage) compared to the total of all fatty acids present in the sample. Fatty acid data were used to calculate the following ratios of fatty acids: total SFA, total MUFA, total PUFA, PUFA:SFA, Δ^9 desaturase index, total n-6; total n-3; the ratio of total n-6:n-3. Atherogenicity index (AI) was calculated as: AI = (C12:0 + 4 × C14:0 + C16:0)/(MUFA + PUFA) (Chilliard *et al*., 2003).

Statistical analysis

The data was subjected to analysis of variance (ANOVA) using the general linear model procedures of the Statistical Analysis System (SAS) program (SAS, 1999). As post hoc analysis, Tukey's HSD test was used to identify significant differences between treatments and significance was declared at the 5% probability level (P <0.05).

The description of the model used for ANOVA was:

$Yij = \mu + ti + \varepsilon ij$,

where Yij is the individual observation (dependent variable) of the i-th treatment (independent variable) and the j-th random error, µ is the general effect, ti is the effect of the i-th treatment, and εij is the random variation or experimental error. The i-th treatment effect (dietary roughage source) during this study was defined as: i_1 = lucerne hay (LH), i_2 = soybean hulls (SH), i_3 = maize stover (MS), i_4 = *Eragrostis tef* (ET).

Results

The effect of fibre source on ram performance is presented in Table 4. The dry matter intake of lambs that received the LH treatment was higher than in the SH and MS treatments (P <0.05), but no effect was recorded compared to the ET treatment with reference to the LH treatment.

Table 4 The effect of fibre source in finishing diets on intake and performance of Merino ram lambs (mean values)

	Primary source of fibre in diet				*P*-value	SE
	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*		
Dry matter intake (g/sheep/day)	1604^{a}	1472^{b}	1477^{b}	1491^{ab}	0.0153	125.11
Metabolizable energy intake (MJ/sheep/day)	14.43^{a}	12.32^{b}	11.48^{b}	12.25^{b}	<.0001	1.07
Initial weight (kg)	27.89	27.63	27.62	28.83	0.2007	1.76
End weight (kg)	52.49^{a}	49.49^{b}	45.68^{c}	47.69^{bc}	<.0001	3.07
Average daily gain (g/sheep/day)	315^{a}	280^{b}	231^{c}	242^{c}	<.0001	32.54
Feed conversion ratio (kg DM feed intake/kg weight gain)	5.10^{b}	5.30^{b}	6.40^{a}	6.25^{a}	<.0001	0.47
Metabolizable energy intake/kg live weight gain (MJ)[1]	45.90^{b}	44.33^{b}	49.78^{a}	51.35^{a}	<.0001	3.98

^{a,b,c} Within a row, means with a common superscript were not different with probability P = 0.05

[1] Daily metabolizable energy intake (MJ) divided by average daily gain (ADG)

Lambs that received the LH treatment also recorded a higher metabolizable energy intake, final empty stomach weight, and average daily gain (ADG) compared to the rest of the treatments (P <0.0001). Of interest is the substantially increased response of lambs fed the SH treatment compared to MS and ET treatments with respect with final weight and ADG. The LH and SH treatments resulted

in the most efficient feed conversion ratio (FCR) and metabolizable energy (MJ) utilized per kg live weight gained (P <0.0001) by the lambs compared to the MS and ET treatments.

The effect of fibre source on lamb carcass characteristics is presented in Table 5. Cold carcass weight of lambs fed the LH treatment was higher (P <0.0001) than in other treatments. Similar results (P <0.0001) were obtained for metabolizable energy intake and ADG as affected by dietary treatment (Table 4). The LH treatment had the highest dressing percentage and only differed (P <0.05) from MS and ET carcasses.

Table 5 The effect of fibre source in finishing diets on the carcass characteristics and meat tenderness of Merino ram lambs (mean values)

	Primary source of fibre in diet					
	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*	*P*-value	SE
Cold carcass weight (kg)	24.93[a]	23.30[b]	20.89[c]	21.89[bc]	<.0001	1.58
Dressing percentage (%)	47.48[a]	47.08[ab]	45.08[b]	45.74[b]	0.0042	1.53
Shoulder circumference (cm)	78.63[a]	77.40[ab]	75.43[c]	76.68[bc]	<.0001	1.72
Buttock circumference (cm)	65.03[a]	63.63[ac]	61.11[b]	62.32[bc]	<.0001	1.91
Carcass length (cm)	61.40	60.47	59.21	60.04	0.0875	2.28
M. longissimus dorsi width (mm)	65.37	65.54	62.52	65.24	0.0960	3.63
M. longissimus dorsi depth (mm)	30.78[a]	27.69[b]	27.09[bc]	28.53[ac]	0.0012	2.51
M. longissimus dorsi area (mm^2)	1912[a]	1766[ab]	1651[b]	1751[ab]	0.0026	176.67
Fat thickness 45 (mm)	2.90	2.76	2.46	2.41	0.5398	1.05
Fat thickness 110 (mm)	7.63[a]	6.16[ab]	5.71[ab]	4.72[b]	0.0023	1.97
Warner-Bratzler shear force (kg)	2.02	2.12	2.09	2.11	0.8283	0.30

[a,b,c] Within a row, means with a common superscript were not different with probability P = 0.05

Shoulder and buttock circumference of lambs fed the LH treatment were higher (P <0.0001) compared to that of the MS and ET treatments (Table 5). The same parameters measured for MS did not differ from that of the ET treatment. *Musculus longissimus dorsi* depth, *Musculus longissimus dorsi* area, as well as fat thickness measured 110 mm from the mid-dorsal line of the lamb carcasses fed the LH treatment were higher (P <0.05) compared only to treatments SH and MS (depth), MS (area), and ET (110 mm fat thickness) (Table 5). There was a lack of effect of fibre source (P >0.05) on carcass length, *Musculus longissimus dorsi* width, fat thickness measured 45 mm from the mid-dorsal line, as well as *Musculus longissimus dorsi* shear force (tenderness).

The effect of fibre source on the muscle fatty acid composition of Merino ram lamb meat is presented in Table 6. Fibre source had no effect (P >0.05) on the muscle lipid content, nor the myristic, palmitic and stearic acid, or palmitoleic acid content of lamb muscle tissue. Marked effects of fibre source on oleic acid, linoleic acid, α-linolenic acid, and CLA were recorded (Table 6). Despite a lower oleic acid content in the LH diet (Table 3), its muscle content higher than the ET treatment (Table 6). The same effect (P <0.05) occurred with reference to muscle linoleic acid content (LH compared to SH and ET). In contrast, and according to most literature, the higher dietary α-linolenic acid content of the LH diet (Table 3) resulted in an increase (P <0.0001) in muscle α-linolenic acid content compared to the other treatments. Even though an effect was recorded (P = 0.0091) the CLA content of lamb muscle tissue fed the LH treatment was comparable with the other treatments. Muscle vaccenic acid content was not affected (P = 0.5079) by dietary treatment.

Table 6 The effect of different fibre sources in finishing diets on the lipid content and fatty acid composition of Merino ram lamb muscle tissue (mean values)

Fatty acid (% of total fatty acids)	Primary source of fibre in diet				*P*-value	SE
	Lucerne hay	Soybean hulls	Maize stover	*Eragrostis tef*		
Muscle lipid content (%)	3.74	3.50	3.70	3.35	0.4079	0.69
Saturated fatty acids:						
Myristic (C14:0)	2.589	2.77	2.59	2.70	0.6714	0.47
Palmitic (C16:0)	29.95	29.85	29.28	29.42	0.6370	1.63
Stearic (C18:0)	12.88	12.54	13.04	13.45	0.2585	1.22
Monounsaturated fatty acids:						
Palmitoleic (C16:1c9)	1.99	1.98	1.86	1.80	0.3201	0.32
Oleic (C18:1c9; n-9)*	37.86^{a}	36.48^{ab}	37.12^{ab}	35.95^{b}	0.0542	1.92
Vaccenic (C18:1t11)	2.33	2.41	2.43	2.52	0.5079	0.34
Polyunsaturated fatty acids:						
Linoleic (C18:2c9,12; n-6)*	6.38^{bc}	7.70^{a}	7.00^{ac}	7.92^{a}	0.0070	1.27
α-Linolenic (C18:3c9,12,15; n-3)**	0.79^{a}	0.49^{b}	0.41^{c}	0.45^{bc}	<.0001	0.07
Conjugated linoleic acid (C18:2c9,t11; n-6)*	0.24^{ab}	0.21^{b}	0.29^{a}	0.22^{b}	0.0091	0.06
Long-chain polyunsaturated fatty acids:						
Arachidonic (C20:4c5,8,11,14; n-6)*	1.26^{b}	1.46^{ab}	1.52^{ab}	1.76^{a}	0.0275	0.43
Eicosapentaenoic (C20:5c5,8,11,14,17; n-3)**	0.19^{a}	0.09^{b}	0.12^{b}	0.12^{b}	<.0001	0.05
Total fatty acids:						
Saturated	46.49	46.26	46.11	46.73	0.8694	2.09
Monounsaturated	44.27	43.48	44.25	42.47	0.0541	1.94
Polyunsaturated	9.24	10.26	9.64	10.80	0.0789	1.69
Unsaturated	53.51	53.74	53.89	53.27	0.8702	2.09
n-6*	7.99^{b}	9.49^{ab}	8.92^{ab}	10.01^{a}	0.0081	1.58
n-3**	1.24^{a}	0.76^{b}	0.72^{b}	0.79^{b}	<.0001	0.14
Fatty acid ratios:						
n-6/n-3**	6.42^{b}	12.59^{a}	12.55^{a}	12.85^{a}	<.0001	1.56
PUFA:SFA	0.20	0.22	0.21	0.23	0.1969	0.04
Δ^{9} desaturase index	2.98	2.93	2.85	2.71	0.1214	0.32
Atherogenicity index	0.76	0.76	0.74	0.76	0.8734	0.09

^{a,b,c} Within a row, means with a common superscript are not different with probability $P = 0.05$

The significant effect of the LH treatment on the linoleic (decreased) and α-linolenic acid (increased) content of lamb meat was associated with a marked decrease in total muscle n-6 content (compared to only the ET treatment), as well as increased total muscle n-3 content (compared to the other treatments) (Table 6). A substantial decrease in the n-6:n-3 ratio of lamb meat fed the LH treatment, compared to the other treatments, was recorded. Of interest to note is the lack of significant effect of dietary treatment on total SFA, MUFA, and PUFA content of lamb meat.

Discussion

The effects of fibre source on lamb growth, carcass characteristics, and muscle fatty acid composition were evaluated in this study. Lucerne hay is a high-quality roughage source and is primarily used in high-producing animals such as growing lambs (Macdonald *et al*., 2021). Maize stover (as well as rice and wheat straw) are referred to as low quality roughages. An important characteristic of low-quality roughages is their slow rate of degradation in the rumen (Pathak, 2008).

Lucerne hay is one of South Africa's most important fibre sources and its superior nutritional quality makes it one of the most sought-after ruminant animal feed sources. The lower dry matter intake of the lambs receiving the MS treatment compared to that of lucerne hay in the present study can be attributed to the slow digestion rate of a lower quality roughage that limits the intake thereof (ARC, 1980). This is primarily due to the formation of cross-linkages between ferulic acid and arabinoxylans in maize stover cell walls (Sun *et al*., 2018). Lucerne hay has a low mean fibre content and brittle texture and is highly degradable (Scholtz *et al*., 2009). Undi *et al*. (2001) observed a positive response in the dry matter intake of sheep fed maize stover as a sole feed compared to legume pasture mixtures in their study. One factor that could have led to a higher dry matter intake of the LH treatment versus MS treatment is the high palatability of lucerne hay (Carro *et al*., 2012). Carro *et al*. (2012) observed a reduction (P <0.05) in dry matter intake of a grass-based diet (less palatable) compared to a lucerne hay diet fed to sheep and goats. A similar dry matter intake among the SH, MS, and ET treatments was

not expected, as soybean hulls are a by-product from a legume plant. The quality of lucerne hay as a fibre source for ruminants can also be recognized by an increased intake due to its higher cell wall density, its ruminal buffering capacity, and to a certain extent, its fast rate of fermentation (Scholtz, 2001).

The significant effect that lucerne hay had on metabolizable energy intake probably resulted in the increased final empty stomach weight and ADG (growth response) of the affected lambs. Sayed (2009) explained that the higher amount of metabolizable energy available for rumen micro-organisms results in an increase of the synthesis of microbial protein and the amount of protein available to the animal, thus improving growth.

Even though lamb growth efficiency (FCR and metabolizable energy MJ utilized per kg live weight gained) of the lambs fed the SH treatment was comparable to that of the LH treatment, the lower dry matter intake of the SH treatment (1472 g/day) compared to the LH treatment (1604 g/day) probably contributed to the lower metabolizable energy intake and ADG of the SH treatment (as stated earlier).

The plant cell wall is the primary cause of reduced feed intake of ruminants fed forage-based diets. The plant cell wall consists of carbohydrates that eventually need to be broken down by sufficient micro-organisms in the rumen (Van Soest, 1994). With an increase in the cell wall content of a fibre source, the fermentation of cellulosic carbohydrates will take longer and, due to rumen fill, will induce the animal to consume more feed (Kung, 2014). A lower dry matter intake could lead to a lower intake of nutrients that could influence animal response which would explain the less efficient FCR and energy efficiency of lambs fed the ET and MS treatments. The latter is however not consistent regarding the dry matter intake of lambs presented the SH treatment, compared to the LH treatment.

The higher cold carcass weight and dressing percentage of lambs fed the lucerne hay diet can be attributed to a higher metabolizable energy intake which resulted an increased weight gain. The increased shoulder and buttock circumference, *Musculus longissimus dorsi* depth and area, as well as fat thickness measured 110 mm from the mid-dorsal line of lambs fed the LH treatment could also have been influenced by the higher lamb metabolizable energy intake. Carcass and meat quality of animals can be influenced by many environmental factors, but it largely depends on the feeding system used. Pasture-fed lambs have a lower dressing percentage, contain more beneficial fatty acids, and have a leaner carcass, whereas concentrate-fed lambs will have better carcass conformation, a higher n-6:n-3 ratio, and higher growth rates (Zervas & Tsiplakou, 2011). The lack of effect of fibre source on carcass length, *Musculus longissimus dorsi* width, shear force, and fat thickness measured 45 mm from the mid-dorsal line is difficult to explain due to the significant effect of LH treatment on the other carcass characteristics reported here.

Significant effects of fibre source on individual UFA (oleic acid, linoleic acid, and α-linolenic acid) can be ascribed to the fatty acid content of the treatment diets, feed and lipid intake, or lipolytic and hydrogenation activity in the rumen. The markedly lower lamb α-linolenic acid content of all the treatments compared to that of the lucerne hay treatment can be attributed to the lower dietary content thereof as well as lower feed intake. In contrast, the substantially lower linoleic acid content of lamb meat (and higher conjugated linoleic acid) presented in the lucerne hay treatment compared to the *Eragrostis teff* and soybean hulls treatments could be the result of increased linoleic acid hydrogenation due to increased isomerisation (Woods & Fearon, 2009). The rate of UFA hydrogenation is higher with immature (higher quality) compared to more mature or senescent roughage (lower quality) (Gerson *et al*., 1986). Any effect on rumen microbial populations may affect rumen hydrogenation patterns. Biohydrogenation has the ability to decrease the UFA and increase the SFA content of rumen digesta (Wood *et al*., 2008). One of the simplest approaches to effect rumen biohydrogenation is to alter the rumen microflora by reducing rumen pH (Kucuk *et al*., 2001). High concentrate diets decrease rumen biohydrogenation and promote increased unsaturation of carcass fat and milk due to decreased lipolysis resulting from low rumen pH (Jenkins, 1993). A decreased pH is believed to also cause incomplete biohydrogenation, which will result in an increased production of CLA and vaccenic acid (Xu *et al*., 2014), indicating the partial escape of PUFAs from biohydrogenation in the rumen.

In the rumen, CLA is produced by different bacterial species. One way is through the isomerisation of linoleic acid, but it can also be produced through the endogenous synthesis from vaccenic acid via Δ9-desaturase enzymes (Woods & Fearon, 2009). The *cis*-9, *trans*-11-CLA isomer is rapidly hydrogenated to vaccenic acid and less rapidly to stearic acid. This could lead to the build-up of this CLA isomer and vaccenic acid in the rumen digesta (Demeyer & Doreau, 1999). Within muscle tissue, the available vaccenic acid content could be desaturated to *cis*-9, *trans*-11-CLA isomer (Schmid *et al*., 2006). The desaturation of vaccenic acid could result in a lower content thereof within the same tissue. Muscle vaccenic acid content was not affected by dietary treatment in the current study.

Dietary fibre source and the quality thereof can have a marked effect on the fatty acid composition of ruminant meat. The maturity of a fibre source has a direct influence on lipolytic and hydrogenation rates (stated earlier). The rate of *in vitro* lipolysis and biohydrogenation of UFA with immature ryegrass is higher compared to when the pasture is more mature or senescent, hence the concentration of vaccenic acid in the rumen is higher (Gerson *et al*., 1986). Ponnampalam *et al*. (2001) observed that the fatty acid composition of the meat was influenced on a low-quality hay (oat hay) or a higher quality hay (lucerne hay) presented to lambs. The same result is apparent in the current study. The same diets used in the current study affected diet DM digestibility fed to Merino rams. Finishing diet total tract DM digestibility with lucerne hay (0.72) and soybean hulls (0.70) was higher (P <0.0001) compared to that of *Eragrostis* teff (0.68) and maize stover (0.66) (Macdonald *et al*., 2021), indicating that lucerne hay and soybean hulls are higher quality roughage sources. One major factor relating to forage quality is the apparent digestibility of its energy content (Blaxter *et al*., 1961). The metabolizable energy content of the different diets were evaluated to contain 9.49, 8.9, 8.7, and 8.25 MJ ME/kg DM (P <0.0001) for lucerne hay, soybean hulls, *Eragrostis* teff and maize stover treatments, respectively (Macdonald *et al*., 2021).

The decrease in the n-6:n-3 ratio is associated with an increased total muscle n-3 and higher α-linolenic acid content, possibly due to the higher content of n-3 in lucerne hay (Mitchell *et al*., 1991). According to McDonald *et al*. (2011) even though the lipid content of lucerne hay on a dry matter basis is low (1.61%), inclusion at high levels could affect certain dietary fatty acids to a limited extent. Hence the fatty acid composition of a feed source fed to animals has a direct influence on the fatty acid content thereof (Manso *et al*., 2009). In a study conducted by Poulson *et al*. (2004) on beef cattle, it was observed that cattle raised on pasture had a more favourable fatty acid composition in comparison to cattle fed intensively. Forages are generally rich in n-3 fatty acids (Woods & Fearon, 2009). Lambs produced in intensive housing and feeding systems were also proven to have a less favourable fatty acid composition (Webb & O'Neill, 2008). The latter effect depends on the type of intensive feeding regime and feed/lipid composition.

Wood *et al*. (2008) stated that the overall fat content of the animal and muscle tissue has an important impact on the proportionate fatty acid composition thereof because of the different fatty acid compositions of neutral lipid and phospholipid. In ruminant animals, the MUFA (French *et al*., 2000) as well as SFA content increases with increasing carcass fatness (De Smet *et al*., 2004). Dietary fibre source did substantially affect muscle fatty acid composition in the current study and could thus be deemed as a causative effect.

From a consumer's health point of view, the suggested ratio of n-6:n-3 should be below 4.0 in muscle tissue, where the suggested value for the PUFA:SFA ratio is 0.4 or higher (Wood *et al*., 2004). In the present study the ratio of n-6:n-3 was higher than 4.0 as suggested by Wood *et al*. (2004) and the PUFA:SFA ratio was lower than the suggested 0.4 in all four of the treatments (Table 6). This is not ideal but at least lucerne hay seems to have a favourable effect (P <0.05) on the n-6:n-3 ratio of ram lamb meat when fed in their finishing diets.

Conclusions

The results of the present study indicate that fibre source has an influence on finishing ram lamb growth, carcass characteristics, and fatty acid composition of *Musculus longissimus dorsi* fat. Most literature agrees that the fatty acid profile of lamb meat can be manipulated by dietary means, where fibre source supports this effect. Lucerne hay included in lamb finishing diets seem to increase the muscle n-3 content, which has a favourable effect on the n-6:n-3 ratio. Lucerne hay did not however affect the PUFA:SFA ratio. The technique to manipulate carcass characteristics and meat fatty acids of finishing lambs by using different fibre sources differing in quality seems promising and warrants further investigation. The effect of fibre source quality in nutritionally similar diets on rumen fatty acid biohydrogenation and pH needs to be elucidated.

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Authors' contributions

JFM and OBE designed the experiment and carried out the research trial. MDF completed the statistical analyses. JFM and OBE structured the scientific content and drafted the manuscript. AH & AL assisted with the

experimental design and research trial, while all authors provided editorial suggestions and approved the final manuscript. AH also conducted the carcass and fatty acid analysis.

Conflict of interest

The authors wish to declare that there is no conflict of interest.

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