Cholesterol, fatty acids profile and the indices of atherogenicity and thrombogenicity of raw lamb and mutton offal

[1]Pretorius, B*. and [1,2]Schönfeldt, H.C.

[1]Department of Animal and Wildlife Sciences, University of Pretoria, South Africa

[2]Professor and Director: ARUA Centre of Excellence: Food Security, University of Pretoria, South Africa

*Corresponding author: beulah.pretorius@up.ac.za; Tel: +27 12 348

Highlights

•Lamb and mutton liver and lung contain <10% a fat and can be considered as “lean” meat.

•There are more cholesterol lowering fatty acids than cholesterol raising fatty acids.

•Lamb offal contains a higher conjugated linoleic acid content compared to mutton offal.

•The offal studied showed an IT of 2.29–3.65 and the IA varied between of 0.03 and 1.83.

Abstract

Dietary fats may affect blood lipid levels and the development of cardiovascular diseases. Offal, may contribute to food security in marginalised communities and information on the contribution to dietary fat intake is needed to inform dietary guidelines and recommendations and consumers. This study aimed to describe the fatty acid profile, cholesterol content and indexes of lipid quality. The fatty acid profile and cholesterol were determined by gas chromatography coupled with flame ionisation detection (GC-FID). To evaluate lipid quality the indices of atherogenicity (IA) and thrombogenicity (IT) were calculated. Offal products can contribute beneficial fatty acids to the diet, not only in terms of essential fatty acids such as linoleic (C18:2n-6) and alpha linolenic (C18:3n-3) acids, but also the polyunsaturated fatty acids, arachidonic (C20:4n-6) and eicosapentaenoic (C20:5n3) acids. The offal studied in the present work showed a P/S ratio of 0.04-0.12 and the n-6/n-3 ratio varied between 3.9 and 12.5.

Keywords: liver, tongue, lipid quality; saturated fatty acids, unsaturated fatty acids

1. Introduction

Offal (or organ meats), as part of the ”fifth quarter” of a carcass, has not been included in the past in dietary guidelines and recommendations, irrespective of the potential contribution towards food and nutrition security (Bester, Schönfeldt, Pretorius, & Hall, 2018). Recent studies by Van Heerden and Morey (2014) and Bester, et al. (2018) confirmed that significant amounts of protein, iron and zinc could be found in some offal, which compared positively with beef and lamb muscle meat cuts. Researchers have recommended offal (organ meats) as good, low cost, nutritious food products and that could potentially contribute positively to food and nutrition security in a country.

However, recent concerns by both consumers and researchers has focused on meat's lipid and fatty acid (FA) content and its wide-ranging implications on health (Vermeulen, Schönfeldt, & Pretorius, 2015). Both the positive and negative effects of dietary lipids in human nutrition has been widely discussed in the past, and increasingly so in recent times with the rise in the popularity of diets high in fat and low-in carbohydrates (Ambrosini, Johns, Northstone, & Jebb, 2015; Kim, 2020). When dietary intake and food sources of fatty acids and cholesterol are considered, it is often perceived that animal sources of food, including red meat, often contribute to a significant proportion of cholesterol and total and saturated fatty acid in the diet (Astrup et al., 2020). Nevertheless, red meat can also provide two essential fatty acids, linoleic acid (C18:2) and linolenic acid (C18:3), in notable quantities (McNeill, Harris, Field, & Van Elswyk, 2012; Hall, Schönfeldt, & Pretorius, 2016). According to previous research reports, omega−3 polyunsaturated fatty acids and conjugated linoleic acid can play a major role in preventing chronic conditions such as coronary heart disease and some cancers. This has resulted in increased interest in the quality of the dietary lipid source as a major contributing factor of long-term health and wellbeing (Koba & Yanagita, 2014; Saini & Keum, 2018; den Hartigh, 2019). Experts agreed that in people with inadequate total energy intake, such as seen in many developing countries, dietary fat is an important macronutrient that increase energy intake to more adequate amounts (FAO, 2010). It is believed that offal is an affordable, alternative nutrient dense animal source food. It is known that offal has a high lipid content (Van Heerden & Morey, 2014; Bester, Schönfeldt, Pretorius, & Hall, 2018), but the fatty acid and cholesterol content is seldomly reported on.

Considering the importance of both macro- and micronutrients in our diet and a precise knowledge of the food composition and quality, this data will provide useful information to consumers, formulators of dietary recommendations and nutrition scientists alike, on health and nutritional value of lamb and mutton offal products.

The aim of this study was to characterize the fatty acids profile and cholesterol content of raw lamb and mutton offal and relate this data to the associated health lipid indices in order to add information on their nutritional quality. Although not the focus of the article, protein, moisture and ash content is also reported to provide a more complete nutrient profile.

2. Materials and methods

2.1. Sampling and preparation

Three samples, from three different animals, of each raw A2 lamb and C2 mutton offal (i.e. hearts, livers, lungs, tongues, stomachs and intestines). Animals were on veld for the first few months of their lives (grass fed and extensive conditions) and then rounded off in the feedlot (intensive conditions) with a total mixed formulated ration. Samples were procured directly from two abattoirs in the Pretoria and Bronkhorstspruit areas, Gauteng, South Africa. This was deemed the best method of sample procurement to ensure that samples were correctly classified as per the Agricultural Products Standards Act (Department of Agriculture, Forestry and Fisheries, 2015). A2 lamb and C2 mutton referred to in this article describes the carcass' age and fatness. Age class ''A" refers to a young animal with no permanent incisors (< 9 months) whereas, age class ''C" refers to an animal with more than six permanent incisors (> 4 years). A fatness code (subcutaneous fat thickness) of ''2" refers to a ''lean" animal (1-4 mm subcutaneous fat).

All lamb and mutton offal samples were washed, scrubbed and cleaned with water to remove all remaining manure and stomach contents, as would be done by the consumer at household level. All the samples were dissected and inedible fractions removed. Thereafter the samples were packed and coded before being transported in chilled containers to the laboratory. At the laboratory the samples were cubed, mixed, minced and freeze-dried. Subsequently the freeze-dried sample were divided into subsamples and sent for

nutritional analyses. at (moisture, protein, ash and total fat analyses), ARC-Irene Analytical Services, Agricultural Research Council (cholesterol analysis) and the Department of Microbial, Biochemical and Food Biotechnology, University of the Free State (fatty acid analysis).

2.2. Nutrient analyses

2.2.1. Proximate analyses

Moisture, ash, nitrogen and total fat analyses were performed in duplicate at NutriLab Analytical Laboratory at the University of Pretoria, South Africa. Because of the scope of the paper, only fat values are discussed.

Moisture was gravimetrically determined the samples by determining the loss in weight of the sample after it had been dried in an oven at 105±1°C for 16 hours. Weight loss is used to calculate dry matter content (AOAC, 2005). Determination of fat percentage was done by continuous extraction with petroleum ether using a Soxhlet apparatus. The extracted fat is recovered, weighed and the fat content calculated. (AOAC, 2000). Nitrogen was determined by the Kjeldahl method. Protein was calculated as nitrogen multiplied by 6.25 (Jones factor) (AOAC, 2000). Ash (inorganic matter) was determined by a dry ashing method in a muffle furnace maintaining temperatures of between 550 °C and 600 °C (AOAC, 2000).

2.2.2. Determination of cholesterol

Cholesterol was determined using the method as reported by Fletouris, et al. (1998) and optimised and validated by the laboratory. Total fat is extracted from the sample using, Soxhlet extraction. The extract is dried, saponified with a ethanol-potassium hydroxide mixture, extracted again with hexane and analysed on a gas chromatograph equipped with a 1.5 m x 3 mm id glass column, packed with 5 % SP 2401 on 100/120 mesh (Supelcoport®) and flame ionization detector. Nitrogen is used as carrier gas with injector temperature of 280 °C and detector temperature of 290 °C. The oven temperature is isothermic at 270 °C. Cholesterol (Sigma-Aldrich) and Stigmasterol (Sigma-Aldrich) dissolved in n-Heptane was used as standard and internal standard respectively.

The cholesterol concentration is expressed as mg/100 g product.

2.2.3. Determination of fatty acids

Lipid from the meat sample was extracted according to the Folch-method (Folch, Lees, & Sloane-Stanley, 1957). A 20 mg aliquot was converted to methyl ester by base-catalysed transesterification in order to avoid conjugated linoleic acid (CLA) isomerisation (Alfaia, et al., 2007). Fatty acid methyl esters (FAMEs) from fat were quantified using a Varian® 430-Gas Chromatograph equipped with a fused silica capillary column, (i.e. Chrompack® CP-Sil 88 (100 m length, 0.25 mm internal diameter and 0.2 µm film thickness)and flame ionization detector. Hydrogen, at 45 psi, functioned as the carrier gas, while nitrogen was employed as the makeup gas. Fatty acid methyl esters in the samples were identified by comparing the retention times of the FAME peaks from samples with those of standards obtained from the Supelco® 37 Component Fame Mix (47885-U, Sigma-Aldrich). Nonadecanoic acid (C19:0) was used as internal standard. CLA standards were obtained from Matreya Inc. (Pleasant Gap, Unites States). These standards included: cis-9, trans-11 and trans10, cis-12-18:2 isomers. The peak identified as CLA was for the specific isomer C18:2c9t11(n–6) (also called rumenic acid), which accounts for 72 to 94% of total CLA in foods from ruminant animals (Mulvihill, 2001; Bhattacharya, Banu, Rahman, Causey, & Fernandes, 2006).

Fatty acids were expressed as the gram (g) of each individual fatty acid per 100g edible portion of the sample using a fatty acid conversion factor of 0.953 **(FAO/INFOODS, 2012)..**

2.2.4. Indexes of lipid quality

From the analytical data on the fatty-acid composition, the following combinations and ratios were calculated: total saturated fatty acids (SFA), total monounsaturated fatty acids (MUFA), total polyunsaturated fatty acids (PUFA), total omega-6 fatty acids, total omega-3 fatty acids, PUFA/SFA ratio and omega-6/omega-3 ratio. To evaluate lipid quality the Index of Atherogenicity (IA) and Index of Thrombogenicity (IT) were calculated.

i) The Index of Atherogenicity indicates the relationship between the sum of the main saturated fatty acids and that of the main classes of unsaturated fatty acids. IA were calculated using the following formula proposed by Ulbricht and Southgate (1991):

$$IA = \frac{[C12{:}\,0 + 4(C14{:}\,0) + C16{:}\,0]}{\sum(MUFA + PUFA)}$$

ii) Index of Thrombogenicity (IT) is defined as the relationship between the pro-thrombogenic (saturated) and the anti-thrombogenic fatty acids (MUFAs, PUFAs – n6 and PUFAs – n3). The thrombogenic index was calculated using the following formula proposed by Ulbricht and Southgate (1991):

$$IT = \frac{C14{:}\,0 + C16{:}\,0 + C18{:}\,0}{0.5x\sum MUFA + 0.5x\sum PUFA(n-6) + 3x\sum PUFA\,(n-3) + \frac{\sum(n-3)}{\sum(n-6)}}$$

2.3. Statistical analyses

All data collected were subjected to an appropriate analysis of variance. Fisher's least significant difference (LSD) was calculated to compare treatment means of significant effects at the 5% probability level (Snedecor & Cochran, 1989). Data were analysed using the statistical program GenStat®.

3. Results and discussion

3.1. Proximate and cholesterol content

The moisture content of the offal ranged from approximately 64% in lamb and mutton tongue and intestines to 80% in lamb and mutton lung. The fat content of the offal (g/100g) ranged from approximately 2.4% in lamb and mutton lung to 28.4% in mutton intestines (Table 1a and 1b). No significant differences ($p \geq 0.05$) were found for either the moisture or the fat content between lamb and mutton offal cuts. Both lamb and mutton liver and lung had a fat content < 10 g/100g and can be considered as “lean” meat according to the Food Based Dietary Guidelines (Schönfeldt, Pretorius, & Hall, 2013). The cholesterol level varies in individual offal cuts. Table 1a and 1b displays the cholesterol level of selected offal cuts and illustrates differences between individual cuts. Cholesterol was highest in mutton tongue (1791 mg/100g) and lowest in lamb lung (11.67 mg/100g).

Table 1a: Proximate, Cholesterol, Fatty Acids Profile and Atherogenic Index of raw lamb and mutton offal (heart, liver, lung and tongue)

Sample Description	Unit	Heart			Liver			Lung			Tongue		
		Lamb	Mutton	p-value	Lamb	Mutton	p-value	Lamb	Mutton	p-value	Lamb	Mutton	p-value
Proximates and Cholesterol													
Moisture	g/100g	71.12±0.36	70.05±4.43	0.79	67.02±3.82	69.830±1.13	0.33	78.58±0.78	79.34±0.67	0.271	64.71±3.16	64.12±3.68	0.84
Ash	g/100g	0.87±0.05	1.10±0.16	0.01	1.40±0.11	1.86±0.60	0.12	1.05±0.04	1.09±0.17	0.60	0.83±0.07	0.61±0.09	0.00
Protein	g/100g	17.24±1.26	16.30±1.42	0.25	18.50±0.50	19.94±1.13	0.03	17.31±0.73	16.75±0.61	0.17	16.00±1.35	12.96±0.74	0.00
Fat	g/100g	11.81±0.42	12.70±3.41	0.78	8.94±5.90	4.33±0.79	0.31	2.53±0.91	2.38±0.16	0.808	17.45±2.70	21.71±3.95	0.207
Cholesterol	mg/100g	590±101.4	786±210.7	0.40	152±170.1	26±5.5	0.33	12±9.3	17±1.5	0.45	809±383.5	1791±495.9	0.06
Fatty Acid Profile													
C12:0	g/100g	0.026±0.02	0.015±0.02	0.62	0.014±0.02	nd	0.31	0.002±0.00	nd	0.18	0.024±0.00	0.007±0.01	0.02
C14:0	g/100g	2.500±1.42	0.325±0.04	0.12	0.759±0.32	0.040±0.02	0.06	1.556±0.46	0.043±0.01	0.03	1.426±0.83	0.327±0.05	0.15
C14:1c9	g/100g	0.009±0.02	nd	0.42	nd	nd	-	nd	nd	-	0.035±0.01	0.007±0.01	0.06
C15:0	g/100g	0.630±0.08	0.090±0.02	0.00	0.391±0.13	0.030±0.01	0.04	0.738±0.07	0.020±0.00	0.00	0.304±0.17	0.107±0.05	0.17
C16:0	g/100g	1.630±0.84	3.030±1.03	0.26	2.185±0.76	1.050±0.30	0.11	4.175±1.08	0.730±0.09	0.03	1.776±1.88	4.577±0.84	0.11
C16:1c9	g/100g	nd	0.140±0.01	0.30	nd	0.040±0.01	0.02	nd	0.020±0.00	-	0.107±0.19	0.187±0.00	0.54
C17:0	g/100g	nd	0.305±0.16	0.23	nd	0.110±0.01	-	nd	0.047±0.01	0.01	0.088±0.15	0.377±0.18	0.11
C18:0	g/100g	0.061±0.03	4.475±2.02	0.20	0.029±0.03	1.407±0.16	0.00	0.014±0.01	0.577±0.11	0.01	1.043±1.76	4.210±0.38	0.08
C18:1t9	g/100g	0.006±0.02	0.235±0.20	0.47	0.008±0.00	0.057±0.07	0.36	0.005±0.00	0.033±0.03	0.19	0.015±0.01	0.037±0.01	0.07
C18:1c9	g/100g	0.009±0.02	2.650±0.14	0.02	nd	1.000±0.27	0.02	nd	0.597±0.04	0.00	2.212±3.74	9.603±2.26	0.06
C18:1c7	g/100g	0.013±0.00	0.180±0.03	0.08	0.007±0.00	0.080±0.04	0.07	0.016±0.00	0.043±0.01	0.05	0.062±0.08	nd	0.33
C19:0	g/100g	0.060±0.03	0.035±0.02	0.34	0.082±0.03	0.023±0.01	0.06	0.119±0.03	0.010±0.00	0.02	0.024±0.00	0.040±0.00	0.03
C18:2t9,12 (n-6)	g/100g	0.002±0.00	0.010±0.00	0.01	0.001±0.00	0.003±0.01	0.60	nd	nd	-	0.018±0.03	0.057±0.01	0.10
C18:2c9,12 (n-6)	g/100g	0.115±0.04	0.440±0.03	0.00	0.114±0.05	0.203±0.06	0.11	0.053±0.00	0.060±0.01	0.37	0.244±0.31	0.737±0.27	0.11
C20:0	g/100g	0.100±0.06	0.030±0.01	0.15	0.046±0.02	0.010±0.00	0.07	0.366.±0.02	0.013±0.01	0.01	0.024±0.00	0.030±0.00	0.07
C18:3c9,12,15 (n-3)	g/100g	0.0015±0.00	0.050±0.01	0.13	0.006±0.01	0.020±0.01	0.14	0.001±0.00	0.010±0.00	0.00	0.040±0.07	0.233±0.03	0.00

Sample Description	Unit	Heart			Liver			Lung			Tongue		
		Lamb	Mutton	p-value	Lamb	Mutton	p-value	Lamb	Mutton	p-value	Lamb	Mutton	p-value
C18:2c9,t11 (CLA) (n-6)	g/100g	0.035±0.02	0.025±0.01	0.56	nd	0.003±0.01	0.42	0.008±0.01	nd	0.42	0.147±0.16	0.097±0.03	0.65
C21:0	g/100g	0.002±0.00	0.005±0.01	0.61	nd	nd	-	0.003±0.00	nd	0.04	0.002±0.00	0.010±0.00	0.03
C22:1c13	g/100g	nd	nd	-	nd	0.003±0.01	0.46	nd	nd	-	nd	nd	-
C20:3c8,11,14 (n-6)	g/100g	nd	0.005±0.01	0.51	nd	0.003±0.01	0.43	nd	0.010±0.00	-	nd	nd	-
C20:4c5,8,11,14 (n-6)	g/100g	0.024±0.01	0.040±0.06	0.76	0.024±0.01	0.033±0.04	0.74	0.024±0.01	0.030±0.01	0.52	0.028±0.02	0.040±0.01	0.41
C23:0	g/100g	nd	nd	-	nd	0.007±0.01	0.19	0.001±0.00	nd	-	nd	nd	-
C24:0	g/100g	0.001±0.00	nd	-	nd	0.003±0.01	0.47	0.001±0.00	0.010±0.00	-	nd	nd	-
C20:5c5,8,11,14, 17 (n-3)	g/100g	nd	nd	-	nd	0.007±0.01	0.42	nd	nd	-	nd	0.003±0.01	0.42
SFA	g/100g	7.447±0.39	8.324±3.24	0.77	5.170±3.01	2.677±0.50	0.29	1.507±0.53	1.453±0.14	0.88	8.100±1.40	9.683±1.23	0.22
MUFA	g/100g	3.093±0.76	3.204±0.03	0.82	2.870±2.38	1.178±0.37	0.34	0.790±0.32	0.700±0.02	0.67	7.570±1.16	9.839±2.29	0.22
PUFA	g/100g	0.713±0.17	0.576±0.04	0.30	0.477±0.24	0.273±0.12	0.28	0.107±0.02	0.116±0.01	0.57	0.960±0.04	1.169±0.31	0.36
Omega-6	g/100g	0.600±0.11	0.520±0.02	0.35	0.427±0.20	0.247±0.10	0.27	0.093±0.02	0.104±0.01	0.49	0.837±0.03	0.931±0.30	0.65
Omega-3	g/100g	0.113±0.06	0.056±0.02	0.25	0.050±0.03	0.025±0.02	0.33	0.013±0.01	0.012±0.00	0.85	0.120±0.02	0.239±0.03	0.00
PUFA/SFA		0.093±0.03	0.075±0.02	0.45	0.097±0.01	0.110±0.07	0.77	0.073±0.01	0.080±0.01	0.50	0.123±0.02	0.120±0.02	0.83
n6/n3		6.830±4.33	9.660±2.28	0.41	8.750±1.73	12.500±5.83	0.38	8.063±2.22	9.773±4.28	0.58	7.173±1.00	3.890±1.05	0.02
IA		0.029±0.10	1.147±0.23	0.74	0.813±0.09	0.823±0.16	0.93	1.103±0.11	1.087±0.11	0.86	0.717±0.02	0.543±0.06	0.02
IT		2.924±0.16	3.648±1.30	0.57	2.723±0.42	2.847±0.59	0.78	2.287±0.07	2.444±0.30	0.46	1.621±0.09	1.445±0.12	0.12

nd = not detected. Results are reported as “not detected” for values less than the limit of detection for the specific analyses.

Table 1b: Proximate, Fat, Cholesterol, Fatty Acids Profile and Atherogenic Index of raw lamb and mutton offal (intestines and stomach)

Sample Description	Unit	Intestines			Stomach		
		Lamb	Mutton	p-value	Lamb	Mutton	p-value
Proximates							
Moisture	g/100g	65.34±8.00	64.51±6.84	0.90	73.94±2.41	70.99±4.34	0.38
Ash	g/100g	0.46±0.19	0.29±0.15	0.15	0.43±0.02	0.40±0.08	0.38
Protein	g/100g	7.01±1.41	6.96±1.29	0.95	10.00±0.91	10.26±1.07	0.66
Fat	g/100g	26.53±8.26	28.37±9.1	0.81	15.69±1.95	18.47±4.42	0.40
Fatty Acid Profile							
C12:0	g/100g	0.088±0.03	0.017±0.01	0.05	0.090±0.05	0.040±0.04	0.26
C14:0	g/100g	1.427±0.22	0.583±0.15	0.01	3.353±1.39	0.780±0.48	0.07
C14:1c9	g/100g	0.049±0.03	nd	0.09	0.030±0.04	0.007±0.01.	0.38
C15:0	g/100g	0.564±0.11	0.237±0.04	0.02	0.659±0.10	0.127±0.04	0.01
C16:0	g/100g	2.042±1.58	5.817±1.07	0.03	0.350± 0.30	4.980±1.46	0.03
C16:1c9	g/100g	nd	0.277±0.08	0.02	nd	0.157±0.03	0.01
C17:0	g/100g	nd	0.703±0.28	0.05	nd	0.400±0.12	0.03
C18:0	g/100g	0.083±0.02	9.510±4.39	0.07	0.133±0.03	5.413±0.95	0.01
C18:1t9	g/100g	0.164±0.062	0.5667±0.48	0.28	0.165±0.13	0.413±0.34	0.33
C18:1c9	g/100g	0.0410±0.03	7.643±2.84	0.04	0.029±0.03	4.677±1.43	0.03
C18:1c7	g/100g	0.013±0.002	0.547±0.24	0.06	nd	nd	-
C19:0	g/100g	0.072±0.02	0.107±0.06	0.40	0.071±0.00	0.047±0.01	0.06
C18:2t9,12 (n-6)	g/100g	0.001±0.00	0.040±0.03	0.16	0.001±0.00	0.020±0.010	0.08
C18:2c9,12 (n-6)	g/100g	0.046±0.01	0.607±0.27	0.07	0.035±0.03	0.350±0.14	0.06
C20:0	g/100g	0.064±0.05	0.090±0.05	0.54	0.068±0.01	0.037±0.01	0.04
C18:3c9,12,15 (n-3)	g/100g	0.012±0.00	0.170±0.12	0.16	0.020±0.02	0.093±0.04	0.07
C18:2c9,t11 (CLA) (n-6)	g/100g	0.080±0.03	0.060±0.03	0.40	0.111±0.02	0.043±0.02	0.01
C21:0	g/100g	nd	0.017±0.01	0.13	nd	nd	-
C22:1c13	g/100g	nd	nd	-	nd	nd	-
C20:3c8,11,14 (n-6)	g/100g	nd	0.023±0.02	0.12	nd	nd	-
C20:4c5,8,11,14 (n-6)	g/100g	0.002±0.00	0.023±0.02	0.14	0.002±0.00	0.013±0.01	0.07
C23:0	g/100g	nd	nd	-	nd	nd	-
C24:0	g/100g	nd	nd	-	nd	nd	-
C20:5c5,8,11,14,17 (n-3)	g/100g	nd	nd	-	nd	nd	-
SFA	g/100g	15.559±3.93	17.078±5.73	0.73	9.989±1.89	11.830±2.65	0.39
MUFA	g/100g	8.903±3.63	9.035±2.89	0.96	4.510±0.19	5.253±1.55	0.49
PUFA	g/100g	0.823±0.35	0.922±0.44	0.78	0.457±0.17	0.522±0.17	0.67
Omega-6	g/100g	0.723±0.33	0.753±0.33	0.92	0.370±0.16	0.426±0.13	0.68
Omega-3	g/100g	0.100±0.02	0.169±0.12	0.44	0.087±0.02	0.096±0.04	0.71
PUFA/SFA		0.053±0.01	0.053±0.01	1.00	0.047±0.03	0.043±0.01	0.84
n6/n3		6.95±2.30	5.123±1.63	0.33	4.410±2.08	4.647±1.07	0.87
AthIndex		1.29±0.12	0.853±0.15	0.02	1.833±0.88	1.423±0.54	0.54
IT		2.868±0.47	2.897±0.52	0.95	3.165±0.58	3.385±0.41	0.62

nd = not detected. Results are reported as “not detected” for values less than the limit of detection for the specific analyses.

3.2. Fatty acid profile

Figure 1 shows the percentages of saturated (SFA), monounsaturated (MUFA), and polyunsaturated (PUFA) fatty acids in various selected lamb and mutton offal cuts. Between 45–70% of the fatty acids in the offal fat are saturated. The SFA ranged from 47-48% in lamb (8.100 g/100g) and mutton (9.683 g/100g) tongue to 66-67% in lamb heart (7.447 g.100g) and stomach (9.989 g/100g) and mutton heart (8.324 g/100g) and stomach (11.830 g/100g). Lamb and mutton tongue have the lowest percentage SFA and highest MUFA. MUFA accounts for around 25–50% of the fat. It ranged from 27% in lamb (3.093 g/100g) and mutton (3.204 g/100g) heart to 47% in lamb (7.570 g/100g) and mutton (9.839 g/100g) tongue with mutton offal samples generally shows a numerically higher MUFA content than lamb samples. The polyunsaturated fatty acid (PUFA) varied between 3-7% depending on the offal cut. PUFA ranged from 3% in mutton stomach (0.522 g/100g) to 7% in lamb liver (0.477 g/100g). The offal studied in the present work showed a P/S ratio of 0.04-0.05 for the lamb and mutton stomach and intestines, while the other offal cuts have a P/S ratio of 0.07-0.12. The P/S ratio of beef is typically about 0.1 and decreases with an increase of fatness of the meat (FAO, 2010). In relation to the P/S ratio, a value above 0.4 is preferable (Wood, et al., 2004).

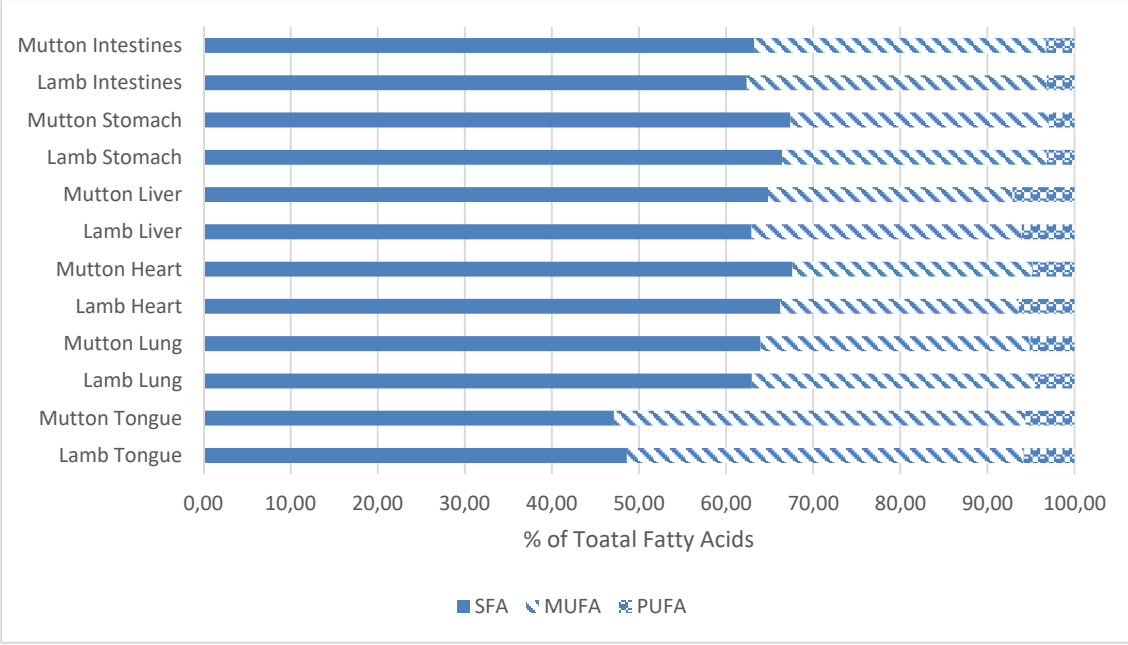

Figure 1: Fatty acid ratio's (SFA, MUFA and PUFA) for various raw lamb and mutton offal cuts.

Table 1a and 1b also shows the fatty acid profile (means of the analyses ± standard deviation) of each lamb and mutton offal cut. Although fats are grouped according to their number of double bonds, and recommendations are formulated for intake based on these groupings, individual fatty acids may have unique biological properties and health effects. Offal contains various individual fatty acids, of which the specific profile varies depending on factors such as animal age, breed, feeding regime and the proportions of lean meat and fat.

Pentadecanoic acid (C15:0) were significantly higher ($p<0.05$) in lamb offal than in mutton offal. Pentadecanoic acid is a biological marker of long term milk intake (Risérus & Marklund, 2017). The reverse was found for stearic acid (C18:0) and *cis*-oleic acid (C18:1c9) where mutton had a significantly higher content than lamb offal cuts. Although stearic acid is a long chain saturated fatty acid, stearic acid has been shown to have a neutral effect on blood total and low-density-lipoprotein (LDL) cholesterol levels and implies to not increase the risk for cardiovascular disease. Mutton offal has a significantly higher concentration oleic acid

than lamb offal. The highest concentration was found in mutton intestines (7.64 g/100g), with the lowest concentration in lamb liver and lung (not detected). Oleic acid is generally considered a heart-healthy fatty acid and is the major MUFA in Western diets (FAO, 2010; Vahmani, et al., 2020). Two specific fatty acids are considered as essential: linoleic acid (LA C18:2 n-6) and alpha-linolenic acid (ALA C18:3 n-3) as these fatty acids cannot be synthesised by the human body and must be obtained from the diet. Although arachidonic acid (AA C20:4 n-6) can be converted from LA, the conversion efficiency is low and therefore AA is considered as conditionally essential. In relation to these essential fatty acids, mutton offal showed a higher percentage compared to lamb offal. LA ranged from 0.035 g/100g in lamb stomach to 0.607 g/100g in mutton intestines, ALA ranged from 0.001 g/100g in lamb lung to 0.233 g/100g in mutton tongue and AA ranged from 0.002 g/100g in lamb intestines and stomach to 0.040 g/100g in mutton heart and tongue.

Food products from ruminants are the principal dietary source of CLA. CLA are produced by microbial fermentation of PUFAs and isomerization of linoleic acid in the rumens of ruminants. The potential benefits of C18:2c9t11 include lowering of total blood cholesterol content (low density lipoprotein (LDL) and high density lipoprotein (HDL)), anti-carcinogenic, anti-diabetic, anti-obesogenic and immunomodulation effects (Koba & Yanagita, 2014; den Hartigh, 2019). In relation to CLA, lamb offal showed a higher content compared to mutton offal, with lamb (0.147 g/100g) and mutton (0.097 g/100g) tongue reporting the highest concentration. Feeding practices and animal related factors such as breed and age of the animal appears to affect CLA content in meat from ruminants (Khanal & Olson, 2004).

3.3. Indexes of lipid quality and health impacts

In Table 2, the cholesterol raising and cholesterol-lowering fatty acids found in South African lamb and mutton offal are presented in terms of the scientific evidence summarised in the Expert Consultation on Fats and Fatty Acids in Human Nutrition (FAO, 2010). The offal cuts in this study contain numerically higher percentage cholesterol lowering fatty acids than cholesterol raising fatty acids. In this study, two individual lipid quality indexes were investigated: 1) Index of Atherogenicity (IA); and 2) Index of Thrombogenicity (IT). These indexes take into account the different effects that fatty acids might have on human health and in particular on the probability of increasing the incidence of atherosclerotic and/or thrombus formation.- Generally lamb offal has a higher IA than mutton with lamb stomach showing the highest value (1.833). The lowest IA was observed in lamb heart (0.029). Saturated fatty acids are considered to favours the adhesion of lipids to cells of the immunological and circulatory system (pro-atherogenic) and unsaturated fatty acids are considered to inhibit the aggregation of plaque and diminishing the levels of esterified fatty acid, cholesterol, and phospholipids, thereby decrease the appearance of micro- and macro-coronary diseases (anti-atherogenic) (FAO, 2010). There was no significant difference between IT for lamb and mutton offal. It must be noted that although stearic acid (C18:0) has a neutral effect on LDL cholesterol and proposed to not have an effect on CVD, it was included in the calculation for IT.

Recently special attention has also been paid to the ratio of n–3/n–6 fatty acids as a very high intake of n–6 acids has been recognized to be less desirable. n−3 PUFAs provide significant protection against cancer, cardiovascular disease and other chronic and metabolic diseases such as diabetes, obesity and osteoporosis. Therefore, a balanced ratio of omega-6/omega-3 fatty acids is important for health and in the prevention of coronary heart disease and possibly other chronic diseases (Simopoulus, 2010; Saini & Keum, 2018). n−3 FAs is usually inadequate because of their limited sources. The n-6/n-3 ratio varied between 3.9 for mutton tongue to 12.5 for mutton liver. It is important to have a low n–6/n–3 ratio in the diet to reduce negative prothrombotic effects caused by increased n–6 linoleic acid concentration in the diet. Currently the desired ratio is still a point of discussion. Simopoulus (2010) suggested that a ratio of 1:1 to 2:1 omega-6/omega-3 fatty acids should be the target ratio for health and earlier in an article by Moreira, *et al.* a desirable ratio n-6/n-3 (n-6 to n-3 fatty acids) of five (5) was suggested (Moreira, Visentainer, de Souza, & Matsushita, 2001).

However, the Expert Consultation on Fats and Fatty Acids in Human Nutrition (FAO, 2010) reported that there is no rational for a specific recommendation for n-6 to n-3 ratio if intakes of individual n-6 and n-3 fatty acids are within guidelines. A low ratio of -6/-3 fatty acids is more desirable in reducing the risk of many of the chronic diseases. Ruminant meats and oily fish are the only significant sources of preformed C20 and C22 PUFA in the diet (Enser, et al., 1998; Wyness, et al., 2011). Although human beings have the metabolic capacity to synthesize C20 and C22 fatty acids from the n-6 or n-3 precursors of linoleic and α-linolenic acid respectively, an increase in the consumption of C20 and C22 n-3 polyunsaturated fatty acids could overcome the perceived imbalance in the ratio of n-6:n-3 polyunsaturated fatty acids in modern diets. Increasing the n- 3 fatty acid content of animal feed is a promising and sustainable way to improve the dietetic value of meat and offal without forcing consumers to change their eating habits. The type and quality of the lipid supplementation in the diet is of special importance (Saini & Keum, 2018).

Table 2: Fatty acid content of South African untrimmed lamb and mutton affal in relation to effect on plasma cholesterol levels

Fatty Acids	**Content (% of total fatty acids)**	
	Lamb	**Mutton**
LDL-cholesterol raising	**31.30**	**27.87**
Lauric acid C12:0	0.25	0.06
Myristic acid C14:0	3.93	2.15
Palmitic acid C16:0	27.12	25.66
Cholesterol neutral	**26.15**	**30.62**
Stearic acid C18:0	26.51	30.62
LDL-cholesterol lowering	**34.2**	**33.30**
Oleic acid C18:1	29.88	29.13
Linoleic acid (Omega 6) C18:2	3.25	3.08
Alpha-linolenic acid (Omega 3) C18:3	0.62	0.60
Arachidonic acid C20:4	0.45	0.49

3.4. International significance

Table 3 shows the comparison of total fat (g/100g), fatty acid profile (g/100g), ratio between polyunsaturated and saturated fatty acids (P/S ratio) and cholesterol (mg/100g) concentration in three offal cuts as reported in various international food composition tables. When compared to these international studies, South African heart and liver have generally a higher fat and cholesterol content than reported for the other countries. Fat content and the resultant SFA, MFA and PUFA values reported for South African tongue are in the same range than the values reported for the other countries. The animal (genetics), feeding regime (grain-fed or pasture-grazed), meat cut and fat trimming practices influence the total fat content of meat cuts (Enser et al., 1998, FAO, 2010, Bester et al., 2018). It is hypothesised that trimming can explain the higher fat and fatty acid content for South African heart and liver, while the values reported for the tongue are in the same range as the international studies. Offal cuts for this study were not trimmed from adipose fat around the organs before sample preparation and the tongue generally needs less trimming before consumption.

Table 3: Comparison of total fat (g/100g), fatty acid profile (g/100g), P/S ratio and cholesterol (mg/100g) concentration of three raw lamb offal cuts as reported in different food composition tables

Nutrient	Country	Offal cut		
		Heart	Liver	Tongue
Total Fat (g/100 g)	South Africa	11.81	8.94	17.45
	New Zealand	3.70	4.90	18.60
	Australia	5.60	7.50	15.5
	France	5.68	5.45	10.9
	United Kingdom	6.80	6.20	14.6
	United States of America	5.68	5.02	17.17
SFA	South Africa	7.447	5.170	8.10
	New Zealand	1.03	1.52	6.06
	Australia	2.34	2.22	6.02
	France	2.25	1.45	3.52
	United Kingdom	2.10	1.70	-
	United States of America	2.25	1.94	6.63
MUFA	South Africa	3.093	2.870	7.57
	New Zealand	0.52	0.79	6.40
	Australia	1.47	2.04	7.08
	France	1.60	0.85	4.99
	United Kingdom	1.70	1.80	-
	United States of America	1.6	1.05	8.46
PUFA	South Africa	0.71	0.477	0.96
	New Zealand	0.64	0.91	0.66
	Australia	0.62	1.26	0.89
	France	0.55	1.44	0.43
	United Kingdom	0.50	0.90	-
	United States of America	0.55	0.75	1.06
P/S ratio	South Africa	0.093	0.09	0.12
	New Zealand	0.63	0.60	0.17
	Australia	0.26	0.57	0.15
	France	0.24	0.99	0.12
	United Kingdom	0.24	0.53	-
	United States of America	0.24	0.39	0.16
Cholesterol (mg/100g)	South Africa	590	152	809
	New Zealand	119	386	88
	Australia	132	389	132
	France	135	371	-
	United Kingdom	140	430	180
	United States of America	135	371	156

- *Means values were not reported in the tables*

Note: South African data reported in this study
New Zealand - (Purchas & Wilkinson, 2013)
Australia – (Food Standards Australia, 2011)
France - (ANSES, 2010)
United Kingdom - (Public Health England, 2015)
United States of America - (USDA, 2018)

The World Health Organisation (WHO) recommends total fat intake not to exceed 30% of total energy intake to avoid unhealthy weight gain. Adults and children should consume a maximum of 10% of their daily energy intake in the form of saturated fat such as meat and butter and less than 1% from trans fats to reduce the risk of heart disease (WHO, 2018). Although no conclusive association can be drawn between animal fat and cardiovascular disesease, public awareness on obesity and cardiovascular disease have increased our concerns in minimizing the consumption of saturated fats and trans fats.

4. Conclusion

Offal is a valuable food of high nutritional content, in which the proportion of fat is frequently overestimated. The fatty acid ratios of offal cuts vary substantially, depending on the cut, the age of the animal and trimming practices. Offal contains a low concentration of beneficial unsaturated fatty acids (UFA), including conjugated linoleic acids (CLA, (C18:2c9t11)), α-linolenic (ALA, 18:3n–3) and oleic acids (C18:1c9) which could be improved through feeding. Offal products can contribute consistently to the diets of consumers that regularly consume it, not only in terms of essential fatty acids such as linoleic acid (C18:2n-6) and alpha linolenic acid (C18:3n-3), but also arachidonic acid (C20:4n-6) and eicosapentaenoic acid (C20:5n3) polyunsaturated fatty acids. Expanding and updating information in country-specific food composition databases on the fatty acid composition of food, especially as measured with improved methods and advances in analytical instrumentation, is essential for studying the relationship between fat and fatty acid intake and health and disease.

5. Limitation of the study

Due to financial constraints, the sample size per age group were small. Fatness code were selected according to the fatness that is mostly preferred by the consumer.

CRediT authorship contribution statement

Beulah Pretorius: Project administration, Visualization, Methodology, Formal analysis, Writing - review & editing.

Hettie Schönfeldt: Conceptualization, Supervision, Funding acquisition, Writing - review

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