THE EFFECTS OF MAIZE COBS AND SUPPLEMENTAL RUMEN-PROTECTED CONJUGATED LINOLEIC ACID (CLA) ON PRODUCTION EFFICIENCY AND MEAT QUALITY CHARACTERISTICS OF SOUTH AFRICAN MUTTON MERINOS

FRANCOIS JACQUES ROUSSEAU

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Supervisor: Prof. L.C. Hoffman
Co-supervisor: Dr. A.V. Ferreira

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DECLARATION

I, the undersigned, hereby declare that the work contained in this thesis is my own original work and that I have not previously in its entirety or in part submitted it at any other university for a degree.

Signature: Date: 10 Maart 2006

SUMMARY

The objective of this investigation was to determine the effect of incremental inclusion of maize cobs, as well as supplemental rumen-protected conjugated linoleic acid (CLA) on the production efficiency and meat quality characteristics of South African Mutton Merino lambs. Prior to this trial, an *in situ* rumen degradability trial was conducted to determine the dry matter, protein and fibre degradability of maize cobs.

Five ruminally cannulated Dohne Merino wethers were used to compare ruminal degradability of lucerne and oat hay with that of maize cobs, using the *in situ* nylon bag technique. The samples were incubated in the rumen for varying time intervals. Both post-incubated and original samples were analysed for dry matter (DM), crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF). The percentage disappearance at each incubation time was calculated from the proportion remaining after rumen incubation. Results obtained clearly established that, on average, the degradability of maize cobs is comparable with that of oat hay.

Twenty four South African Mutton Merino (SAMM) lambs were used in a feedlot finishing study to evaluate the effect of maize cobs at incremental inclusion levels (4, 8 and 12%) on individual feed intake, average daily gain (ADG) and feed conversion efficiency (FCE). The apparent digestibilities of the three diets were also determined. The three pelleted diets were formulated, on an isonitrogenous and isoenergetic basis. The lambs were slaughtered after the trial had finished and the *longissimus dorsi* and *biceps femoris* muscles were removed from each carcass for the determination of proximate chemical composition and physical quality characteristics. Neither ADG nor FCE of the lambs was influenced by dietary treatment. Lambs fed the diet containing 4% maize cobs had the higher nitrogen retention, while diet did not affect energy retention. The proximate chemical composition of both muscles was not significantly affected by diet. Diet only had a significant effect on the cooking loss and the colour measurements of the *M. longissimus dorsi.*

Sixteen South African Mutton Merino (SAMM) lambs were used in a second feedlot finishing study to evaluate the effect of feeding a diet containing supplemental rumen-protected CLA. The procedure followed was the same as in the first feedlot finishing study, except for an additional sensory and fatty acid analysis. No significant differences occurred in the ADG, FCE and dressing percentage of the lambs. Both energy and nitrogen retention of the lambs were not affected by dietary treatment. The proximate chemical composition of both muscles

was similar, but diet had a significant effect on the cooking loss and the colour measurements of the *M. longissimus dorsi.* Diet had no significant effect on any of the five sensory attributes measured. Fatty acid composition was significantly affected by dietary CLA. Palmitic (C16:0), stearic (C18:0) and oleic (18:1n-9) acid were the major fatty acids in both muscles, as well as in the three adipose tissue types. The rumen-protected CLA increased the CLA (C18:2n-6) contents of both muscles, although the increase was only significant for the *M. longissimus dorsi* and not for the *biceps femoris* muscle.

OPSOMMING

Die doel van die ondersoek was om die invloed van toenemende insluitingsvlakke van mieliestronke, asook aanvullende rumen-beskermde gekonjugeerde linoleïensuur (CLA), op die produksie doeltreffendheid en vleis kwaliteitseienskappe van Suid-Afrikaanse Vleismerino (SAVM) lammers, te bepaal. Voorafgaande dié proef, is 'n *in situ* degradeerbaarheidstudie uitgevoer om die droë materiaal-, proteïen- en vesel degradeerbaarheid van mieliestronke te bepaal.

Vyf Dohne Merino hamels met rumenkannulas is gebruik om, met behulp van die *in situ* nylon sakkie tegniek, die degradeerbaarheid van lusern en hawerhooi met dié van mieliestronke te vergelyk. Sakkies met die monsters is by verskillende tydsintervalle in die rumen geïnkubeer. Monsters is voor en na inkubering ontleed vir droë materiaal (DM), ruproteïen (RP), neutraal bestande vesel (NBV), asook suur bestande vesel (SBV). Die persentasie verdwyning is by elke inkubasie tydperk bereken vanaf die oorblywende proporsie van die monster na inkubering in die rumen. Dit is bevind dat die gemiddelde degradeerbaarheid van mieliestronke vergelykbaar is met die degradeerbaarheid van hawerhooi.

Vier en twintig Suid-Afrikaanse Vleismerino (SAVM) lammers is vir 'n voerkraalafrondings studie gebruik. Die effek van toenemende insluitingsvlakke (4, 8 en 12%) van mieliestronke op individuele voerinname, gemiddelde daaglikse toename (GDT) en voeromsettings-doeltreffendheid (VOD) is gemonitor. Die skynbare verteerbaarheid van die rantsoene is ook bepaal. Die drie diëte was geformuleer op 'n gelyke stikstof en energie basis. Na afloop van die proef is die lammers geslag en die *longissimus dorsi*, asook *biceps femoris* spiere van elke karkas is verwyder vir die bepaling van chemiese samestelling en fisiese kwaliteits eienskappe. Dieet het geen betekenisvolle effek op GDT of VOD van die lammers gehad nie. Die dieet met 'n mieliestronk insluitingsvlak van 4% het aanleiding gegee tot lammers met die hoogste stikstofretensie, terwyl energieretensie nie deur dieet beïnvloed is nie. Die chemiese samestelling van beide spiere is nie betekenisvol deur die dieet beïnvloed, maar wel slegs die kookverlies en kleur van die *M. longissimus dorsi.*

Sestien Suid-Afrikaanse Vleismerino (SAVM) lammers is vir 'n tweede voerkraalafrondings studie gebruik om die effek van 'n dieet wat aangevul is met 'n rumen-beskermde CLA, te evalueer. Dieselfde proefprosedure as tydens die eerste afrondingsproef is gevolg, behalwe vir die addisionele/bykomende sensoriese, sowel as vetsuur analises. Geen betekenisvolle

verskille het voorgekom vir GDT, VOD en uitslag persentasie van die lammers nie. Beide energie en stikstofretensies is ook nie deur die dieet beïnvloed nie. Chemiese samestelling van beide spiere was soortgelyk, maar dieet het wel 'n betekenisvolle effek op kookverlies en kleur van die *M. longissimus dorsi* gehad. Dieet het geen betekenisvolle effek op enige van die vyf sensoriese eienskappe van die vleis teweeg gebring nie. Vetsuursamestelling van die vleis was betekenisvol beïnvloed as gevolg van die CLA aanvulling in die dieet. Palmitiensuur (C16:0), steariensuur (C18:0) en oleïensuur (18:1n-9) het die grootste proporsie van die vetsure in beide spiere uitgemaak, asook in al drie die vetdepots. Die rumen-beskermde CLA het die CLA (C18:2n-6) inhoud van beide die spier tipes verhoog, alhoewel die verhoging slegs betekenisvol vir die *M. longissimus dorsi* was.

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NOTES

Language and style used in this thesis are in accordance with the requirements of the *South African Journal of Animal Science.* This dissertation represents a compilation of manuscripts where each chapter is an individual entity and some repetition between chapters has, therefore, been unavoidable.

Chapter 1

General introduction

1. Introduction

This project was conducted to obtain more information on the *in situ* rumen degradability of maize cobs, as well as the effect of incremental inclusion of maize cobs in feedlot finishing diets on the production efficiency and meat quality characteristics of South African Mutton Merino (SAMM) lambs. A second independent study was undertaken to determine whether rumen-protected conjugated linoleic acid (CLA) has an effect on production efficiency, proximate chemical composition and physical meat quality characteristics, as well as on fatty acid composition and sensory attributes of South African Mutton Merino (SAMM) lambs.

Maize cobs

Due to the unprecedented increase in the cost of conventional ingredients used in compounding livestock feed, intensive investigations into the use of agricultural by-products was necessitated. Because of the high priority of food crop production over that of animal feedstuffs, low-quality roughages (< 7% crude protein), especially crop residues such as cereal straws, are the predominant energy sources available to ruminants for a considerable part of the year. According to the most recent predictions, the commercial production of maize in South Africa for the year 2006 is estimated at 6 238 000 metric ton (Van Zyl, 2006). Kossila (1984, as cited by Tuah *et al.*, 1996) stated that for every kilogram of maize grain, three kilogram of fibrous by-products and residues are produced. This indicates that residues (cobs and stovers) from the harvesting of maize are one of the most abundant crop residues produced annually in the summer rainfall area of South Africa. Crop residues in this particular area already fulfil a strategic role in fodder flow programs of ruminants and can be used to reduce feed costs (Snyman & Joubert, 2002).

Ruminants fed on crop residues and by-products add value to resources which are largely wasted in the absence of a ruminant component in the system. The primary nutrient sources in these residues are the structural carbohydrates in the cell walls, i.e. cellulose, hemicellulose, pectin and lignin. In contrast to humans and monogastric animals, ruminants have the ability to digest these carbohydrates in the rumen through microbial fermentation. Energy in the form of volatile fatty acids (VFA's), of which butyrate, propionate and acetate are the most predominant, can then be utilized (McDonald *et al.*, 1995). Livestock farmers in

the northern parts of Nigeria (Onwuka *et al.,* 1997), Ghana and other West African countries (Tuah *et al.,* 1996) utilize significant quantities of crop residues while their stock return manure to the soil. In Ghana, maize have been grown on plots that had been grazed by cattle for more than ten years, without applying fertilizers during the cultivation of the maize (Tuah *et al.,* 1996).

The fact that these low-quality roughages are generally not palatable, fibrous and often deficient in nitrogen (N), minerals (macro- and trace minerals) and vitamins, often cause them to be insufficient in providing an adequate amount of digestible energy to meet the maintenance requirements of grazing ruminants (Dias-da-Silva, 1986). However, these residues are relatively inexpensive, readily available and considering the growing population of the world with its consequent increasing demand for food, the challenge is to produce more food on a decreasing area of agricultural land. This means an increased efficiency of production processes, and therefore these low-quality roughages need to be re-examined for maximum utilization of its energy for livestock production.

Research has been done in recent years in Ghana to increase the efficiency of utilization of maize cobs (Tuah *et al.,* 1996). Tuah & Ørskov (1989) reported that treating maize cobs with sodium hydroxide will increase the dry matter disappearance in the rumen, thus enhancing its nutritive value. They have also found that anhydrous ammonia treatment increased, although less effectively than sodium hydroxide treatment, the dry matter disappearance of maize cobs.

Meat quality

The past four decades has been characterized by a decline in consumption of lamb in the United States from more than 1.36 kg to a level of 0.37 kg per person annually on a boneless, retail weight basis (Duckett, 2003). Consumption patterns indicate that Americans continue to consume greater amounts of meat in their diet; however the consumption of lamb continues to decline (Duckett, 2003). The declined lamb and mutton consumption might be due to the fact that the last couple of years have been characterized by an increase in consumer interest in the nutritional aspects of health, which resulted in the development of health directives by governments for some food components, especially fats (Simopoulos, 2001). The pressure on sales has caused a reappraisal of the factors which influence the appeal of meat to consumers, which all comes down to the quality of the meat. According to Wood *et al.* (1999), the freedom of meat from microbial hazards, prevention of animal exploitation, sensory appeal of meat, and perceived healthiness, especially in relation to the amount and type of fat; is some of the factors determining meat quality.

Lamb and mutton contain high concentrations of saturated fatty acids, so much so that its polyunsaturated:saturated fatty acid ratio (P:S ratio) is lower than the recommended minimum value of 0.45 given by Warris (2000) for human diets. The excessive consumption of food with a high proportion of saturated fatty acids (SFA) is a major predisposing factor to the risk of coronary heart diseases (CHD), hypertension, stroke, diabetes and obesity in humans, which has led to a worldwide decline in red meat consumption (Webb *et al.,* 1994b; Moloney *et al.,* 2001). The relationships between dietary fat and incidences of diseases associated with the modern life or lifestyle, especially CHD (Kritchevsky, 1998) and various cancers (Wood *et al.,* 2003), are well established. The low P:S ratio of lamb and mutton is a consequence of the extensive biohydrogenation of ingested PUFA by the rumen microorganisms, leading to the formation of *trans*-monounsaturated and SFA, which are then incorporated into the lipids in the muscle (Jenkins, 1993).

The degree of saturation of animal fats is influenced by the fatty acid composition (Webb & Casey, 1995). Accordingly, the quality of fat is determined by the fatty acid composition, which affects the palatability and shelf life (Rhee, 1992). Webb *et al.* (1994a) reported that a shift in fatty acid composition can be induced by means of dietary manipulation, which will subsequently enhance the nutritional quality of lamb meat and fat quality. Dietary manipulation strategies are also available that minimise biohydrogenation of ingested PUFA in the rumen (Rowe *et al.,* 1999; Chikunya *et al.,* 2004).

2. *In situ* degradability of feedstuffs

Ruminant animals generally consume large quantities of forage in their diet. The ability to formulate a diet for sheep and cattle according to their requirements depends to a great extent on the accuracy with which the quality and quantity of forages offered and consumed can be estimated (Valentin *et al.,* 1999). Animal performance (milk production and growth) can be improved through the ability to accurately determine the nutritive value and potential digestibility of feedstuffs, and thus improve estimation of requirements for supplementary feeds (Valentin *et al.,* 1999).

A variety of methods exist by which degradability or digestibility of feedstuffs can be measured. *In vitro* techniques, as in the early technique developed by Tilley & Terry (1963) or in the gas production method (Menke *et al.,* 1979), are routine evaluation methods of rumen fermentation. An alternative method, which has become increasingly popular as a means to quantify the rumen degradable and undegradable fractions of feedstuffs, is the *in*

situ technique (Nocek & English, 1986). This technique incorporates estimates of the kinetics of degradation in the reticulo-rumen, which was achieved by measuring the rumen degradation of feed placed in nylon bags incubated in the rumen of ruminally cannulated animals (Ørskov & McDonald, 1979). This *in situ* technique also allows a number of feeds to be assessed simultaneously, whereas conventional *in vivo* digestibility measurements are limited in that they are restricted to assessing one feed or a combination of feeds in the form of a diet at a time (Sauvant *et al.,* 1985). This technique is used as the standard method for determination of protein degradation in the protein evaluation systems in the USA, UK and Nordic countries (Van der Honing & Alderman, 1988), since the separation of feed protein into ruminally degradable (RDP) and undegradable (UDP) protein fractions has been emphasized (Broderick *et al.,* 1988).

However, the *in situ* technique also has some limitations. The rate of disappearance from the nylon bag rather than the actual degradation is essentially measured by this technique. Furthermore, feedstuffs are confined in a bag and are not subjected to mastication and rumination by the animal. Several researchers (Kennedy *et al.,* 1984; Nocek, 1985; Nocek & Grant, 1987) have demonstrated that bacterial contamination may also cause considerable error, particularly with feedstuffs low in protein. Susmel *et al.* (1989) found that the main dietary factors affecting the disappearance of feedstuffs are intake level, forage to concentrate ratio, diet composition and frequency of feeding.

Although some information is available on the nutritive value of low-grade roughages in other countries, these values are not necessarily applicable to crop residues produced and harvested under South African conditions. Little information is available on the degradability of nutrients and effective degradability values for maize cobs. Ørskov (1989) highlighted the importance of ruminal degradability of a feedstuff for the evaluation of its nutritive value.

3. Production efficiency of sheep in a feedlot

Growth of an animal can be defined as an increase in body weight until mature size is reached, which is due to an increase in cell size and numbers resulting in protein deposition. Changes in body conformation and form, and the direct coordination of all diverse processes until maturity is reached, is defined as development (Lawrie, 1985). This growth curve relating live weight to age has an S-shape, consisting of a short initial phase when live weight increases little with increasing age; followed by a phase of explosive growth and finally a phase when growth rate has reached a plateau (Brody, 1927, as cited by Lawrie, 1985).

Santos-Silva *et al.* (2002) made the statement that changing the production system with the focus on increasing animal productivity and economic results is desirable, provided that meat quality and consumer acceptance are maintained. The supplementation of lambs with commercial concentrates in a feedlot system is becoming a common practice in the endeavour to obtain better quality products. In meat production systems, a small increase in slaughter weight of lambs, may result in higher productivity, and will give more flexibility to the production system (Sañudo *et al.,* 1996). Carcasses can be produced with fatness levels increased to more adequate levels, ensuring better carcass appearance and protection against cold-shortening during storage (Lawrie, 1985; Santos-Silva *et al.,* 2002).

Feedlot animals fed concentrate-based diets have higher average daily gains (ADG) in comparison with lambs on pastures and thus, if slaughtered at a constant age, feedlot animals would have obtained higher body weights (Priolo *et al.,* 2002; Santos-Silva *et al.,* 2002). These authors also reported that carcasses from feedlot lambs were heavier and had a higher dressing percentage than lambs on pasture. The higher dressing percentage might be explained by the fact that dressing percentage depends on the forage content of the diet, and therefore on the weight of the digestive tract (Brosh *et al.,* 1995). Lambs in a feedlot with similar ADG, receiving a high energy diet will achieve a more efficient feed conversion efficiency (FCE) than lambs fed a low or medium energy diet (Malik *et al.,* 1996). According to Guertin *et al.* (1995), an animal will continue to eat until its intake capacity for energy is reached and metabolic factors limit its voluntary feed intake, thus lambs on a high energy diet need to eat less than lambs on a low energy diet to elicit the same growth response. Zinn & Plascencia (1996), on the other hand, reported that energy intake by feedlot steers increased linearly with increasing dietary energy density. Haddad & Husein (2004) stated that it has been shown that high energy diets improve growth rates, as well as growth efficiency in different sheep breeds.

The inclusion of dietary fat or oil in feedlot diets to increase the energy content of the diet is a commonly used practice. Johnson & McClure (1973) reported that feeding high levels of fat to ruminants can cause palatability problems, as well as an adverse effect on production by decreasing fibre digestibility. A decrease in fibre digestibility is the consequence of modifications in the ruminal microbial ecosystem, as it has been demonstrated that lipids have a negative effect on bacterial growth, especially cellulolytic strains (Chalupa *et al.,* 1986; Doreau & Chilliard, 1997). Another effect of fat supplementation is the decrease in protozoal population, which contributes to cellulolysis. Both these effects of fat supplementation is more pronounced with polyunsaturated fatty acids than with saturated fatty acids (Doreau & Chilliard, 1997). Zinn (1989) observed a linear decrease in feed intake and empty body gain,

when fat was added to diets of finishing steers. In practice, therefore, fats are usually limited to 5% in the diets of ruminants (Murphy *et al.,* 1990, as cited by Brand *et al.,* 2001).

Numerous attempts have been made to limit the extent of lipid ruminal hydrogenation and the disturbances of fibre digestion. This means not only the protection of lipids against microbial attack, but also the protection of microbes against the negative effect of lipids (Doreau & Chilliard, 1997).

According to Casey & Webb (1995) and Santos-Silva *et al.* (2004) the feeding of pelleted diets increases the average daily gain (ADG) and decreases the feeding period as opposed to feeding diets in the loose form. One explanation given by them for this tendency is that wethers fed pelleted diets cannot select specific feed components from the diet, and therefore achieve higher intakes and feed conversion ratios, compared to those fed in a loose form. It is well established that a reduction in forage particle size, by for example pelleting the feed, increases voluntary intake (Sauvant, 2000, as cited by Santos-Silva *et al.,* 2004) by a reduction in rumination time and in the retention time of food in the reticulo-rumen (Faichney, 1995, as cited by Santos-Silva *et al.,* 2004).

Quite often lambs/sheep are raised on poor quality forage, resulting in higher mortality rates, slower growth rates and reduced reconception of the ewes in the following mating season. In such conditions lambs may have to be weaned earlier and finished in feedlots (Nolte & Ferreira, 2004). Results obtained by Malik *et al.* (1996) suggest that with older and heavier lambs the FCE and ADG declines with increasing age. They have also suggested that it may be beneficial to market lambs at 5 months when the growth rate has peaked, rather than marketing them at 7 months when growth has significantly slowed down. A faster turnover from the feedlot and possibly higher profits can be achieved by the early weaning and finishing of lambs in a feedlot.

4. Physical characteristics of sheep meat

Colour

Meat purchasing decisions are influenced by colour more than any other quality factor because consumers use discolouration as an indicator of freshness and wholesomeness (Jeremiah *et al.,* 1972). Carpenter *et al.* (2001) noted a positive association between colour preference and purchasing intent, with consumers discriminating against beef or mutton that

is not red (i.e., meat that is purple or brown). Colour is the visual characteristic of meat that imparts the first impression and it can be measured subjectively and objectively.

Myoglobin is the principle protein responsible for meat colour, although other heme proteins such as blood pigment hemoglobin and cytochrome C also play a role in beef, lamb, pork and poultry colour. According to Schmidt (2002), lamb and mutton contain less myoglobin than beef. Lamb and mutton are therefore generally lighter in colour than beef, but this colour darkens with increasing animal age. The transformation of myoglobin to oxymyoglobin occurs due to oxygenation, when myoglobin is exposed to oxygen and is characterized by the development of a bright red colour (Lawrie, 1985). After prolonged atmosphere exposure, the ferrous myoglobin is oxidised to ferric iron, forming brown metmyoglobin and causing discolouration (Livingston & Brown, 1982). Metmyoglobin formation depends on numerous factors including oxygen partial pressure, temperature, pH, meat's reducing activity, and in some cases, microbial growth (Mancini & Hunt, 2005).

Currently, many options are available for instrumental colour analysis. According to Stevenson *et al.* (1989) the expression of colour by the coordinates L*, a* and b* of the CIELab colour space (Commission Internationale de l'Eclairage, 1976) are appropriate measures of colour. The L* represents lightness in meat colour on a scale from 0 to 100, where 0 corresponds to pure black and 100 corresponds to pure white. A positive a* value indicates redness with redness increasing as the number gets further from 0. A negative a* value indicates greenness with greenness increasing as the value gets further away from 0. A positive b* value indicates yellowness and a negative b* value represents blueness (Poulson *et al.*, 2004). The a* and b* values are used to calculate hue angle (h_{ab}) and chroma value (C*) according to the following equations: hue angle (°) = $\tan^{-1}$ (b*/a*), and the chroma value = $[(a^*)^2 + (b^*)^2]^{1/2}$ (Commission Internationale de l'Eclairage, 1976). The value of chroma is zero at the centre of the chromaticity diagram and increases according to the distance from the centre, being dull near the centre and becoming more vivid away from the centre. Hue angle is defined as starting at the positive a* axis and is expressed in degrees. The closer the hue angle is to 90°, 180° and 270° the more the colour corresponds to yellow, green and blue, respectively.

According to Honikel (1998), there are three sources responsible for colour variation in meat. Firstly, the myoglobin content is dependent on primary production factors such as species, breed, age and nutritional status; secondly, the preslaughter period, slaughter process and subsequent processing affect colour by influencing the rate and extent of pH and temperature decline; thirdly, the colour and colour changes occurring during handling and storage.

According to Lawrie (1985) and Velasco *et al.* (2004), with more stress factors prior to slaughter, the ultimate pH (pH_u) will be higher causing the meat to be darker (Dark, firm and dry, DFD).

pH

It is a well-known fact that the quality of meat is greatly affected by the development of pH during the first 24 h after exsanquination (Andersen *et al.,* 1999), as well as the ultimate pH (Hoffman *et al.,* 2003). This is due to the anaerobic breakdown of the glycogen reserves in the muscles by glycolysis resulting in the production of lactic acid and a subsequent decline in pH (Lawrie, 1985). The quantity of glycogen, which remains in the muscle at death, determines the ultimate pH (pH_u) value. The conversion of glycogen to lactic acid will continue until a pH is reached when the enzymes affecting the breakdown becomes inactivated, unless when exercise or stress immediately pre-slaughter has appreciably diminished the reserves of glycogen in the muscle, leading to a higher pH_u (Lawrie, 1985; Martínez-Cerezo *et al.,* 2005). In rested, fed animals the pH_u is about 5.4-5.5 (Lawrie, 1985; Devine *et al.,* 1993).

Ante-mortem stress to animals has been a cause of major concern to farmers for many years because of its negative effect on meat quality. Various stress factors have been mentioned as responsible for glycogen depletion: time and manner of transportation of animals from the farm to the abattoir; diet restrictions; mixing of animals of different species; lairage time; climatic factors; pathological conditions and genetic factors to list but a few (Silva *et al.,* 1999). It has been proven that any situation which provokes a substantial depletion of muscle glycogen reserves prior to slaughter, will give rise to meat with a higher ultimate pH, and according to Devine *et al.* (1993) an ultimate pH value greater than 5.8 is regarded as undesirable. Bacteriological stability (Lawrie, 1985) and shelf life (Viljoen *et al.,* 2002) of chilled meat is adversely affected by an ultimate pH value above 6.0. Dark, firm and dry meat (DFD) is classified as having a pH_u above 6.2, whereas intermediate DFD meat has a pH_u that ranges from 5.8 to 6.2 (Wiklund *et al.,* 1995). Meat with high pH_u also has a greater water-holding capacity (Bouton *et al.,* 1973), tends to produce abnormal flavours and is associated with a higher rate of tenderisation (Watanabe *et al.,* 1996) or with a better ultimate tenderness (Bouton *et al.,* 1973).

Conflicting reports regarding the relationship between pH and tenderness (WBSF) are found in the literature. Devine *et al.* (1993) reported an increase in shear force of lamb with higher pH values in the range of 5.4–6.0, whereas Purchas (1990) and Jeremiah *et al.* (1991) found a curvilinear relationship between pH and tenderness, with a minimum tenderness between pH

5.8 and 6.2. Then again, Safari *et al.* (2001) found no relationship between pH and shear force value and cooking loss in six diverse lamb genotypes.

5. Chemical composition of sheep meat

Proximate composition

The nutritive value, appearance, processability, shelf life and palatability of meat are affected by both the quantity and quality of fat in the meat. For this reason fat is an important determinant of meat quality and the degree of saturation of the fat contributes substantially to the sensory properties of meat, as well as to the health concern to consumers (Webb *et al.*, 1994a).

In ruminants, lipids are mainly accumulated as triacylglycerols in adipocytes, located in subcutaneous, inter- and intramuscular adipose tissue, and abdominal fat depots. In literature, emphasis is normally laid on intramuscular fat, since it is irreversibly connected with meat and cannot be removed prior to human consumption, as is the case for visible fat, such as subcutaneous fat (Raes *et al.*, 2004a).

With increasing animal weight, the proportion of subcutaneous fat increases more than intermuscular and intramuscular fat tissue. According to Demeyer & Doreau (1999), marbling fat (intramuscular fat) starts developing only after kidney, intermuscular and subcutaneous fat has been formed. The percentage of fat in meat varies widely, depending on the species, breed, sex, nutritional state and the part of the carcass being investigated (Jeremiah *et al.*, 1997). Breed, as a source of variation of the amount of fat and meat quality, is a complex factor, as the results will vary depending on the comparison criterion used: equal live or carcass weight, equal age, equal degree of maturity or equal percentage of adult live weight (Sañudo *et al.*, 1998). Female animals generally produce carcasses with the highest fat content, which is distributed mainly in the front and ventral regions of the carcass (Bennet *et al.*, 1991), followed by wethers and then males which produces the leanest carcasses (Field *et al.*, 1990).

The composition of muscle varies with increasing animal age, regardless of species and sex. Lawrie (1985) states that there is a general increase in most parameters other than moisture but the rates of increment vary in different muscles. He also stated that the increase in intramuscular fat and myoglobin content of muscle is evident. Also evident is the lesser increase of total nitrogen and a decrease in moisture with age. The decrease in the moisture

content of the carcass with increasing age can be explained by the existence of a reverse relationship between fat and moisture content of the carcass (Dransfield *et al.,* 1990; Martínez-Cerezo *et al.,* 2005). Thus muscles with a high fat content are characterized by a low moisture content.

Intramuscular fat refers to the fatty acids present in the intramuscular adipose tissue and in the muscle fibres. The intramuscular adipose tissue is comprised of fat cells, isolated or in clusters, along the fibres and in the interfascicular area and contains mainly triacylglycerols, while the lipids of the fibres are cytosolic droplets of triacylglycerols, phospholipids and cholesterol. In the muscle, the phospholipid content is relatively constant and less influenced by species, breed, nutrition, and age (Raes *et al.,* 2004a). Phospholipids are characterized by a high polyunsaturated fatty acid (PUFA) content (20–50% of total fatty acids in the phospholipids), mainly represented by long chain fatty acids with 18, 20 and 22 carbons and two to six double bonds (Raes *et al.,* 2004a). In contrast to phospholipids, the largest part of triacylglycerol fatty acids consists of saturated fatty acids (SFA) and mono-unsaturated fatty acids (MUFA) (Sinclair & O'Dea, 1990).

Fatty acid composition and cholesterol content

Dietary cholesterol is an important issue to public health due to its relationship with the incidence of atherosclerosis. It is a general accepted fact that excessive consumption of fat, above a persons caloric needs is an important risk factor in cardiovascular heart diseases (CHD), hypertension, stroke, diabetes and obesity. Currently, consumers are very conscious of their dietary intake of high-fat animal food that contains saturated fatty acids and cholesterol, which in turn, elevate serum cholesterol (Flynn *et al.*, 1985).

Fatty acids are the most important lipid fraction of meat (Russo *et al.,* 1999). The fat type or fatty acid composition of meat is an important diet or health concern to consumers. It also is a primary factor that determines product shelf-life or storage stability, flavour and fat tissue firmness (hardness) (Webb & Casey, 1995). Ruminant fat tissue is naturally firmer than that of pigs, because the fatty acids profile is more saturated. The effect of fatty acids on firmness is due to the different melting points of the fatty acids in the meat. As the degree of unsaturation increases, melting points decline (Wood *et al.,* 2003). In lambs, especially ram lambs, soft fat develops in animals fed grain-based (concentrate) diets. This is due not only to a reduced concentration of C18:0 but also to an increased deposition of medium to long-chain (C10:0-C17:0) branched fatty acids formed from methylmalonate, a metabolite of propionate (Busboom *et al.,* 1981). A survey conducted by Enser *et al.* (1996) illustrated the difference in fatty acid composition and content between beef, lamb and pork (Table 1).

Table 1 Fat and fatty acid composition of beef, lamb and pork loin steaks purchased from four supermarkets (Enser *et al.*, 1996)

Fatty acid	Beef	Lamb	Pork
Whole steak			
Fat content (%)[1]	15.6	30.2	21.1
Muscle[2]			
16:0	25.0	22.2	23.2
18:0	13.4	18.1	12.2
18:1n9	36.1	32.5	32.8
18:2n6	2.4	2.7	14.2
18:3n-3	0.70	1.37	0.95
20:4n-6	0.63	0.64	2.21
20:5n-3	0.28	0.45	0.31
22:6n-3	0.05	0.15	0.39
Total fatty acids[3]	3.8	4.9	2.2
P:S	0.11	0.15	0.58
n-6:n-3	2.11	1.32	7.22
Fat[2]			
16:0	26.1	21.9	23.9
18:0	12.2	2.6	12.8
18:1n9	35.3	28.7	35.8
18:2n6	1.1	1.3	14.3
18:3n-3	0.49	0.97	1.43
C20-22n-3 PUFA[4]	ND	ND	0.36

[1]% of steak; [2]% of total fatty acids; [3]% of muscle; [4]% of fat tissue; ND, not detectable

The last few years have been characterized by an increase in consumer interest in the nutritional aspects of health, which has resulted in the development of specific health directives by governments for some food components (Simopoulos, 2001), including fats. There is a perception among consumers, and often the medical profession, that red meat is a high-fat food with a high proportion of saturated fatty acids (SFA). These fatty acids are considered to increase the risk of coronary heart diseases (CHD), which led to a worldwide decline in red meat consumption (Moloney *et al.*, 2001). The relationships between dietary fat and incidence of diseases associated with modern life or lifestyle, especially CHD (Kritchevsky, 1998) and various cancers (Wood *et al.*, 2003), are well established. In the United Kingdom, the Department of Health recommended that fat intake should not exceed 30% of total energy intake, with a figure of 10% of energy intake for saturated fatty acids (Wood *et al.*, 2003), and energy intake from mono-unsaturated fatty acids (MUFA) and

polyunsaturated fatty acids (PUFA) should be approximately 16% and 7%, respectively (Moloney *et al.*, 2001). At the same time, the recommended ratio of PUFA to SFA (P:S ratio) should be increased to 0.45 or higher (Warris, 2000). The ratio of n-6:n-3 PUFA is also a risk factor in cancers and CHD, especially the formation of blood clots leading to a heart attack (Enser, 2001). According to nutritional guidelines, a n-6:n-3 ratio of less than 4 is recommended (Enser *et al.*, 1998). According to Simopoulos (2001) the current n-6:n-3 ratio in the Western Europe and the USA is between 15 and 20.

Moloney *et al.* (2001) noted that lean beef, with an intramuscular fat content of 5% or less, contained approximately 47%, 42%, and 4% of total fatty acids as saturated fatty acids (SFA), mono-unsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), respectively. Stearic acid (C18:0) represents about 30% of the total SFA. It has been shown that stearic acid (C18:0) is considered to be neutral in its effect on serum cholesterol levels, and is easily converted into oleic acid (C18:1n-9) which, from a dietetic point of view, is one of the desirable fatty acids (Rhee, 1992; Grundy, 1997). It is generally accepted that plasma cholesterol concentration is influenced by the fatty acid composition of dietary fat. It is a well-known fact that C16:0 increases the lipid levels in blood, and thus increases total serum cholesterol levels, whereas C18:1n-9 lowers lipaemia by reducing both the low-density lipoprotein (LDL) cholesterol and triglycerides content of the blood, without reducing the high-density lipoprotein (HDL) cholesterol (Grundy, 1997; Russo *et al.*, 1999).

One of the most important strategies to increase the PUFA content and to alter the fatty acid composition of meat to be more compatible with consumer requirements is through increasing the dietary supply of n-3 PUFA. Feeding grass or linseed as a rich source of α-linolenic acid (C18:3n-3) or fish oil or fishmeal as a source of the long-chain PUFA eicosapentaenoic acid (EPA, C20:5n-3) and docosahexaenoic acid (DHA, C22:6n-3) increases the levels of these PUFA in the meat (Choi *et al.*, 2000; Moloney *et al.*, 2001).

According to literature, increasing the dietary PUFA have had little impact on the P:S ratio of red meat derived from ruminants (Choi *et al.*, 2000; Moloney *et al.*, 2001; Scollan *et al.*, 2001). The intramuscular fatty acid composition of the monogastric animals, and in particular the triacylglycerols are a reflection of the dietary fatty acids, while in ruminants the biohydrogenation in the rumen (i.e. saturation of the dietary unsaturated fatty acids) is responsible for the smaller variations in intramuscular fatty acid composition (Harfoot & Hazlewood, 1988; Byers & Schelling, 1993). This biohydrogenation in the rumen results in low amounts of dietary PUFA relative to SFA to bypass the rumen (Demeyer & Doreau, 1999). Phospholipid composition is less influenced by diet, as they are constituents of cell

membranes. Encapsulation of emulsified oils with formaldehyde-treated proteins provide effective protection against microbial hydrogenation of dietary fatty acids (Scott & Ashes, 1993), thus making the PUFA available for digestion and absorption in the small intestine.

Although some dietary PUFA in linseed and other plant oils escape rumen biohydrogenation, a high proportion (> 90%) is hydrogenated leading to high values for saturated fatty acids in ruminant meat (Scollan *et al.,* 2001). To achieve higher values for the P:S ratio, two options are available. The first option is to feed a predominantly cereal (concentrate) diet in which rumen biohydrogenation is less effective, while the second option is to "protect" the dietary oil using a procedure such as formaldehyde treatment of dietary protein which protects the oil within a matrix structure (Scott *et al.,* 1971).

The main essential fatty acids of the n-3 and n-6 group are α-linolenic acid (C18:3n-3) and linoleic acid (C18:2n-6). These two fatty acids are desaturated and are lengthened in the human body to more potent and bioactive long chain fatty acids, such as arachidonic acid (C20:4n-6), EPA and DHA (Horrobin, 1992). Both n-3 and n-6 PUFA's are essential for normal early human development. Arachidonic acid is an essential nutrient for infant growth (Maurage *et al.,* 1998), whereas DHA is present in very high concentrations in the grey brain matter, in the retina, and is a prerequisite for normal cognitive and visual development (Sangiovanni & Chew, 2005). Arachidonic acid and DHA appear in the highest concentrations in the adult human brain, which contains 50-60% of its dry weight as lipids, and 35% of the lipid content is accounted for by PUFA's. Dietary PUFA intake will thus determine cell membrane phospholipid content and fluidity of synaptic membranes, and to a large extent how they transduce neurotransmitter signals (Horrobin & Bennett, 1999). These essential PUFA's can also be linked with: attention deficit hyperactivity disorder and learning stress, depression, multiple sclerosis, Alzheimer's Disease, cardiovascular disease, obesity, bone health, skin conditions, cancer and immunity (Anonymous, 2005).

6. Sensory attributes of sheep meat

The acceptability of red meat after purchase is determined almost exclusively by the satisfaction derived from its consumption (Jeremiah *et al.,* 1991). The increasing importance of lamb eating quality to retailers and consumers and consistency of tenderness has been highlighted in numerous studies (Safari *et al.,* 2001). The three main components of the palatability of meat include tenderness, juiciness and flavour. The combination of these attributes determines the overall eating satisfaction (Koohmaraie *et al.,* 2003). Consumer

surveys have shown that tenderness is considered the most important component of meat quality (Young *et al.,* 1993; Wood *et al.,* 1999). According to Stone & Sidel (1993), sensory analysis is defined as a scientific method used to evoke, measure, analyse and interpret the responses to products as perceived through the senses of sight, smell, touch, taste and hearing.

Tenderness

Koohmaraie *et al.* (1990) and Safari *et al.* (2001) reported that the most important contributing sensory attribute to eating quality is tenderness, with flavour and juiciness also contributing significantly, although to a lesser extent. Consumer surveys have also shown that tenderness is considered the most important component of meat quality (Young *et al.,* 1993; Wood *et al.,* 1999). After consumption, meat tenderness is the property which will lead to overall acceptability of the product by the consumer. Tenderness corresponds to the ease of mastication during meat consumption (Tornberg *et al.,* 1985). Consumers even stated that, when meat is tough, it is unacceptable (Wood *et al.,* 1999). In general, the more tender the meat, the more rapidly juices are released by chewing and the less residue remain in the mouth after chewing (Tshabalala *et al.,* 2003).

Meat tenderness is assessed by both sensory and instrumental methods. Because of the high cost of sensory panel testing and the fact that it is labour- and time-consuming, muscle pH and Warner Bratzler shear force are instrumental measurements that are often used to indicate eating quality (Safari *et al.,* 2001). Results obtained by Devine *et al.* (1993) and Safari *et al.* (2001) indicated a significant ($P < 0.001$) correlation of -0.7 between shear force values and taste panel tenderness rating.

Species is an important quality differentiating criterion, as the meat from each species has its own characteristics. These characteristics vary a great deal more between red meat (beef, lamb, etc.) and white meat (pork, calf, chicken, etc.) than within each of those groups, especially when meat is consumed cold (Sañudo *et al.,* 1998). It seems clear that sheep meat is more tender than beef or pork (52.63, 82.91 and 78.40 N, respectively, in New Zealand retail meat), but there is a great variability between animals, around 50% (Bickerstaffe *et al.,* 1997, as cited by Sañudo *et al.,* 1998). Dransfield *et al.* (1990) found, with respect to texture, males to be tougher than wethers because collagen accretion might be stimulated by testosterone (Miller *et al.,* 1990). Devine *et al.* (1993) suggest that the effect of age on the shear force and tenderness of lamb is relatively small, but, young animals would, in principle, be more tender as they possess a more soluble collagen. The connective tissue (collagen) content of muscle becomes less soluble with increasing animal age, causing meat to be less

tender (Lawrie, 1985). Sañudo *et al.* (1996), on the other hand, suggested that shear force and tenderness are significantly affected by carcass weight.

Juiciness

Koohmaraie *et al.* (1990) and Safari *et al.* (2001) reported that juiciness is also an important contributing sensory attribute to eating quality. Meat juiciness is firstly dependent on the inherent water-binding capacity (the ability of meat to retain its natural moisture content) and secondly on the marbling fat content of the muscle (Offer & Trinick, 1983). Forrest *et al.* (1975) stated that meat juices contain many of the important flavour components and assist in the process of fragmenting and softening of meat during mastication.

Dryden & Marchello (1970) reported that juiciness is related to both the capacity of the muscle to release its constitutive water (initial juiciness) and the infiltrated fat content (sustained juiciness). The melted lipid and water in the muscle constitutes a broth that, when retained in the meat, is released upon chewing. This broth may also stimulate the flow of saliva, and thus improve the meat's apparent juiciness (Forrest *et al.*, 1975). However, the major contributor to the sensation of juiciness is the water remaining in the cooked meat (Forrest *et al.*, 1975). Safari *et al.* (2001) noted that a negative correlation exists between cooking loss and juiciness, which will in turn affect the eating quality and acceptability of the meat.

The meat from young animals with a lower intramuscular fat content (marbling fat) is characterized with less juiciness than meat from mature animals, which are well marbled. Meat from these young animals give a watery sensation on first chewing but a final impression of dryness, whilst in animals of a greater weight and age, a greater sustained juiciness would be experienced due to their greater level of fatness (Sañudo *et al.*, 1996). Owens & Gardner (1999) indicated that juiciness was negatively correlated to moisture content of the *longissimus* muscle, and positively correlated to the fat content of this muscle. In accordance with the latter authors, Mandell *et al.* (1997, as cited by Moloney *et al.*, 2001) reported that *longissimus* steaks with higher fat content were more juicy than steaks containing a lower fat content. According to Camfield *et al.* (1997) myristic (C14:0), palmitic (C16:0) and margaric (C17:0) acid concentrations of the *longissimus* muscle were negatively related to juiciness.

Flavour and aroma

Aroma and flavour are important components of eating quality of all foods, and are sometimes used as determining criteria in the acceptance or rejection of the product, especially in lamb and mutton meats (Mottram, 1992). Meat flavour is the result of the excitation of two physiological senses: taste and smell. However, the sensation of overall mouthfeel during mastication can also play a role (Geay *et al.*, 2001). According to Owens & Gardner (1999) and Tshabalala *et al.* (2003) the overall flavour intensity of mutton is positively related to fat content of the meat. Crouse *et al.* (1981) found that meat of older lambs had superior flavour scores than meat of younger animals.

In general, aroma compounds of meat are more soluble in the fat content of the meat and tend to be retained for longer in the fat matrix (De Roos, 1997). Therefore, in more fatty meat, the aroma compounds will then be released and perceived as flavour during mastication (Tshabalala *et al.*, 2003). On the contrary, in lean meat the aroma compounds become volatile faster and are released together with the water vapour (aroma perception).

The characteristic mutton aroma is primarily associated with the volatile acidic components and branched-chain fatty acids (Webb *et al.*, 1994a). The meaty flavours of cooked meat can be divided into two groups, those formed from lipid oxidation, particularly polyunsaturated fatty acids (PUFA), and those resulting from reactions between carbohydrates and proteins, and between their breakdown products (Maillard reaction) (Mottram, 1998).

7. Ruminant meat as a source of conjugated linoleic acid (CLA)

Conjugated linoleic acid (CLA) is the name for a group of fatty acids that occurs naturally in ruminant meats, such as beef and lamb, and also in milk. However because CLA is a fatty acid, the making of "fat free" and "reduced fat" dairy products results in decreased amounts of this compound in foods (Rainer & Heiss, 2004). CLA has been shown to reduce the risk of mammary tumours in animal studies and because of these findings; the American Dietetic Association has endorsed lean beef and lamb as functional foods (ADA, 1999). Ha *et al.* (1987) were the first group to identify CLA as a potential anticancer factor when they isolated four isomers of CLA from extracts of fried ground beef. They then used synthetic isomers of CLA and showed that they inhibited the induction of skin tumours in mice by carcinogens.

The acronym CLA is a collective term used to describe a mixture of positional and geometric isomers of linoleic acid (C18:2 *cis*-9, *cis*-12) (Fig. 1). CLA has the same chain length as

linoleic acid (C18:2n-6), but in CLA the double bonds are conjugated, rather than methylene-separated as they are in linoleic acid. Conjugated bonds are separated by one single carbon bond rather than two or more bonds. The bonds in CLA can be either in a *trans* (*t*) or *cis* (*c*) configuration.

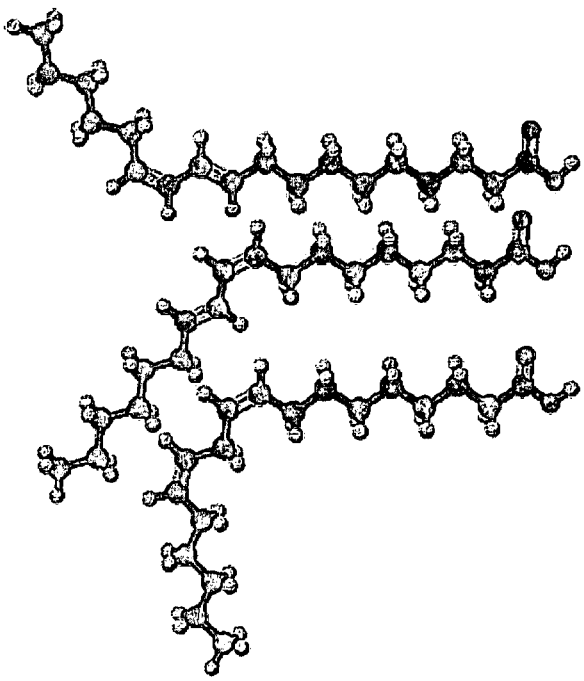

Figure 1 Structures of *trans*-10, *cis*-12 CLA (upper panel); *cis*-9, *trans*-11 CLA (middle panel); and linoleic acid (lower panel) (Pariza *et al.*, 2001)

The anaerobic ruminant microorganism, *Butyrivibrio fibrisolvens*, is considered the main producer of CLA (Kepler & Tove, 1967). CLA isomers are intermediates in the biohydrogenation of linoleic acid (C18:2 *cis*-9, *cis*-12) to *trans*-vaccenic acid (C18:1 *trans*-11) and stearic acid (Fig. 2) (Kelly *et al.*, 1998; Mir *et al.*, 2000). Kinetic studies of rumen biohydrogenation of linoleic acid to stearic acid have shown that the C18:2 *cis*-9, *trans*-11 isomer of CLA is a transient intermediate, whereas C18:1 *trans*-11 is the intermediate that accumulates (Harfoot & Hazlewood, 1988). Factors regulating the synthesis of CLA in the rumen are poorly understood. It was thought that the CLA isomers pass to the small intestine where they are absorbed together with the other fatty acids of dietary origin, re-esterified and ultimately circulated to all parts of the animal. Griinari *et al.* (2000) suggested that the above view is too simplistic and that a portion of CLA may originate from endogenous synthesis. The dietary addition of plant oils containing α-linolenic acid also increase the CLA content of ruminant fat, and intermediates in its pathway of biohydrogenation include C18:1 *trans*-11 but not CLA (Griinari *et al.*, 2000). Results obtained by Griinari *et al.* (2000) clearly demonstrated that the oxidative reaction catalyzed by Δ^9-desaturase for the endogenous

synthesis of CLA, exist. In growing ruminants, adipose tissue was the major site for Δ^9-desaturase, and mammary glands the major tissue site in lactating ruminants (St. John *et al.*, 1991). This knowledge could then be used to manipulate successfully the concentrations of CLA in meat.

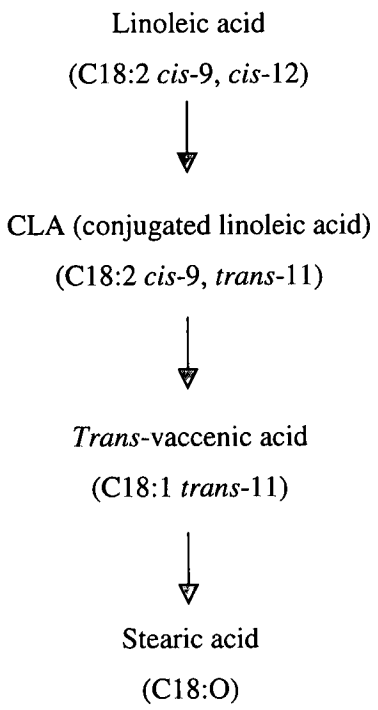

Figure 2 Pathway of biohydrogenation of linoleic acid to stearic acid by rumen microorganisms (Kelly *et al.*, 1998).

Many positional (7,9-; 8,10-; 9,11-; 10,12- or 11,13-) and geometrical (*cis*, *cis*-; *cis*, *trans*-; *trans*, *trans*- or *trans*, *cis*-) isomers of CLA are known to exist. The C18:2 *cis*-9, *trans*-11 isomer is sometimes called rumenic acid (Kramer *et al.*, 1998) and is the predominant isomer of CLA. It has been shown to account for at least 60% of total CLA in beef (O'Shea *et al.*, 1998). Other researchers have estimated that the *cis*-9, *trans*-11 isomer accounts for 80% (Chin *et al.*, 1994) and 90% (Fritsche & Fritsche, 1998) of total CLA in beef. These differences may be accounted for by variances in ruminal biohydrogenation, methods of analysis, diet of the animal, cut of meat, seasonal variation or genetic influences. The enzyme responsible for the formation of this isomer was isolated from *B. fibrisolvens* and identified as linoleate *cis*-12, *trans*-11-isomerase (Kepler & Tove, 1967). Other CLA isomers found in beef include *trans*-9, *trans*-11; *trans*-10, *cis*-12; *trans*-10, *trans*-12 (Ha *et al.*, 1987), and *cis*-9, *cis*-11 and *trans*-9, *cis*-11 (Fritsche & Fritsche, 1998).

CLA and human health

CLA may have many possible beneficial influences in human health, based upon evidence from cell culture and animal studies. Research has highlighted the anticarcinogenic (Ha *et al.*, 1987; Ip *et al.*, 1994), antiatherogenic (Lee *et al.*, 1994) and immune-modulating (Liu & Belury, 1997) properties of CLA. CLA has also been shown to increase growth efficiency (Chin *et al.*, 1994; Wiegand *et al.*, 2001) and reduce body fat while increasing lean body mass in growing animals (Park *et al.*, 1997; Park *et al.*, 1999a; Azain *et al.*, 2000). Anti-diabetic effects of CLA isomers have also been reported (Houseknecht *et al.*, 1998). Probably the greatest area of interest at the present time is in the anticancer properties of CLA (Belury, 1995; Kritchevsky, 2000; Belury, 2002; Wahle *et al.*, 2004).

Despite the evidence that CLA induces changes in body composition in animals, studies using human subjects are limited and contradictory (Rainer & Heiss, 2004). The question whether CLA is safe, is one that is often been asked. Berven *et al.* (2000) conducted a trial on humans and observed no adverse effects among 30 people receiving 3.4 g CLA/day for 12 weeks. This level is approximately 10 times higher than the current estimated intake.

Different isomers of CLA appear to be responsible for its differing biological effects. For instance, in a study conducted by Park *et al.* (1999a), preparations of CLA supplement were enriched with either the *cis*-9, *trans*-11 or the *trans*-10, *cis*-12 isomers, and their effects on the body composition in mice were compared. The supplemented mice had reduced body fat mass and increased body protein, ash and water when compared to the non supplemented mice. The observed changes in the body composition resulted from feeding the *trans*-10, *cis*-12 isomer and not the *cis*-9, *trans*-11 isomer (Park *et al.*, 1999a). The *trans*-10 ,*cis*-12 isomer has been shown to reduce lipoprotein lipase activity, intracellular triacylglycerol and glycerol concentrations, and enhance glycerol release into the medium in cultured adipocytes (Park *et al.*, 1999a). Similarly, when the supplement was withdrawn from the diet of the mice, the *trans*-10, *cis*-12 isomer cleared significantly faster in skeletal muscle than the *cis*-9, *trans*-11 isomer (Park *et al.*, 1999b). The *trans*-10, *cis*-12 isomer appears to accumulate less in the tissues and appears to play a role in adiposity. The *cis*-9, *trans*-11 isomer may accumulate in the tissues, and may affect tissue development and alter gene expression, thus playing an anticarcinogenic role.

As interest in CLA is relatively new, all the studies mentioned above, which have shown health benefits of CLA, have been conducted in either cell culture or animal models. One human study has shown that CLA supplementation (3 g/day for 8 weeks) significantly decreased plasma triglyceride concentrations in healthy volunteers when compared to

supplementing with a control fatty acid (linoleic acid) (Noone *et al.*, 2001). These authors attribute these observations to the *trans*-10, *cis*-12 isomer.

CLA content of meat

The CLA content of meat has been analyzed in many countries all over the world, but some studies shows that the CLA content of meat varies from country to country. For instance, Australian beef has a higher *cis*-9, *trans*-11 CLA content (7.6 mg/g fat) (Fogerty *et al.*, 1988) than American beef (1.7–2.7 mg/g fat) (Shantha *et al.*, 1994). Some of the variances may be due to specificity differences in the methods of CLA analysis, or disparities in the animals, such as their diet. For instance, the higher CLA content in Australian beef is probably due to the likelihood that the animals are fed on grass, which is high in polyunsaturates. Grass-fed beef has been shown to contain higher quantities of CLA than grain-fed beef (Shantha *et al.* 1994).

As already stated, CLA is formed naturally in the rumen, thus ruminant meats (lamb and beef) have a higher CLA content than monogastrics (pork and chicken) (Chin *et al.*, 1992). Comparisons of different meats show that lamb contains the highest quantity of CLA (Table 2). For example, measurement of total CLA/g fat show that lamb contains 5.8 mg/g fat, whereas beef contains 2.9–4.3 mg/g fat and veal contains 2.7 mg/g fat (Chin *et al.*, 1992). Monogastric meats such as chicken and pork, contain 0.9 mg/g fat and 0.6 mg/g fat, respectively (Chin *et al.*, 1992). It is not yet established whether the CLA found in monogastric meats is a consequence of diet or is perhaps due to the conversion of linoleic acid to CLA by bacterial flora (Chin *et al.*, 1994).

Levels of CLA are much higher in meat containing greater amounts of fat (*cis*-9, *trans*-11: 960–1310 mg/100 g meat) than in lean meat (*cis*-9, *trans*-11: 6–43 mg/100 g meat) (Fogerty *et al.*, 1988). As expected, monogastric fat contains much lower levels of CLA than ruminant fat, with levels of *cis*-9, *trans*-11 CLA in chicken fat ranging from 120–130 mg/100 g meat (Fogerty *et al.*, 1988). There is no difference in the CLA content of intermuscular and subcutaneous fat (Fritsche & Fritsche, 1998).

Table 2 Conjugated linoleic acid content of various foods (Chin *et al.*, 1992)

Food	mg/g fat	Food	mg/g fat
Dairy Products		Meats/Fish	
Homogenized milk	5.5	Fresh ground beef	4.3
2% milk	4.1	Veal	2.7
Butter fat	6.1	Lamb	5.8
Condensed milk	7.0	Pork	0.6
Cultured buttermilk	5.4	Chicken	0.9
Butter	4.7	Fresh ground turkey	2.6
Sour cream	4.6	Salmon	0.3
Ice cream	3.6	Egg yolk	0.6
Low-fat yoghurt	4.4		
Custard style yoghurt	4.8	Vegetable Oils	
Plain yoghurt	4.8	Safflower oil	0.7
Frozen yoghurt	2.8	Sunflower oil	0.4
Medium cheddar	4.1		
American processed	5.0		

Factors influencing the CLA content of meat

Feeding trials have focused mainly on changing the CLA content of milk, whereas altering the CLA content of muscle does not appear to have been so widely studied. As CLA is produced naturally in ruminant animals from dietary linoleic acid, elevating dietary intakes of polyunsaturated fatty acids would be expected to result in increases in CLA content of meat. Grass is high in polyunsaturates and top round beef from grass-fed cattle has a 1.5-fold higher CLA concentration compared with that from grain-fed cattle (Shantha *et al.*, 1994). Supplementing the diet of cattle with n-3 fatty acids (linseed) causes a significant 2–3-fold increase in the CLA content of beef when compared with a diet supplemented with saturated fatty acids (Enser *et al.*, 1999). These authors propose that the observed increase in CLA occurs as a result of the n-3 fatty acids inhibiting the conversion of CLA to *trans* vaccenic acid. According to Kelly *et al.* (1998) the CLA content of ruminants fats can be increased by formulating diets that contain more linoleic acid, as the hydrogenation pathway of linolenic acid do not involve CLA as intermediate. Fritsche & Fritsche (1998) found no difference in the CLA concentrations or isomer distribution between the samples of fat from bulls and bullocks, thus suggesting that the CLA content of meat is not influenced by hormonal status. Age, diet and breed of animal, as well as seasonal variation, all appear to influence the CLA content of ruminant meat.

8. The South African Mutton Merino

The South African Mutton Merino (SAMM) is a dual-purpose (mutton and wool) sheep breed that was developed by selection from the German Merino (Neser *et al.*, 2000). The primary selection objective in the SAMM is meat production, while wool plays a secondary role. The SAMM is a strong, well muscled polled (hornless) sheep with an excellent conformation and balance (South African Mutton Merino Breeders' Society, 2001), and is known for its high growth rate and capability to produce a slaughter lamb at an early age with good meat quality attributes (Cloete *et al.*, 2004).

In terms of growth rate, the SAMM is rated the most successful mutton breed in South Africa. A feed conversion efficiency (FCE) of 3.9 has been achieved in finishing lambs, with an achieved optimum between 25-42 kg live weight (South African Mutton Merino Breeders' Society, 2001). Under extensive conditions lambs can obtain an average live weight of 35 kg at 100 days of age. Under intensive conditions, rams can weigh up to 56 kg in the same period (South African Mutton Merino Breeders' Society, 2001). Du Plessis & De Wet (1981) found that 61% of nitrogen retained in the body was utilized for body protein synthesis in SAMM lambs, in comparison with the 19% in Merino lambs. This explains the 53% faster growth rate for SAMM lambs at 100 days of age (Brand & Franck, 2000). Fat deposition in the SAMM lambs occur at a much later stage of life, therefore heavier carcasses, up to 27-28 kg, can be produced and still obtain the best grading possible (South African Mutton Merino Breeders' Society, 2001).

The SAMM has good mothering abilities, with an approximately 30% higher milk production 17 days post partum compared to Merino ewes. Ewes can produce as much as 4.8 ℓ milk per day. The breed is also known for its high fertility, with lambing percentages of 171% (Brand & Franck, 2000). This percentage is slightly higher than the 150%, generally accepted as lambing percentage for this breed (Brand & De Villiers, 1989, as cited by Brand & Franck, 2000). According to Brand & Franck (2000) birth weight of SAMM lambs is lower compared to that of Merino lambs, due to the higher rate of multiple births.

This breed is popular in feedlot practises, due to its ability to efficiently convert feed into body weight and adaptability to a wide variety of environmental conditions (Neser *et al.*, 2000). It also has the ability to utilize low quality roughage and is non-selective in its grazing habits (South African Mutton Merino Breeders' Society, 2001).

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Chapter 2

In situ dry matter, crude protein, and fibre degradation kinetics of selected roughages and maize cobs

Abstract

Ruminal degradability of lucerne hay, oat hay and three maize cob treatments (normal, coarse and fine) was compared using the *in situ* nylon bag technique. Samples of lucerne hay, oat hay, and the three maize cob treatments were ground through a 2 mm screen and five grams DM of each of the milled samples were then weighed into marked dacron bags. The lucerne hay and oat hay were used to compare the three maize cob treatments with. The samples were incubated in the rumen of five ruminally cannulated Dohne Merino wethers for varying time intervals of 0, 2, 4, 8, 16, 24, 48 and 72 h. Chemical analysis measured from both post-incubation and original samples included dry matter (DM), crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF). The percentage disappearance at each incubation time was calculated from the proportion remaining after rumen incubation. Results obtained from this study clearly established that, on average, the DM, NDF and ADF degradability of maize cobs, especially that of the normal maize cobs, is comparable with that of oat hay.

Key words: degradability; percentage disappearance; maize cobs; dry matter; crude protein; neutral detergent fibre; acid detergent fibre

Introduction

The unprecedented increase in the cost of conventional ingredients used in compounding livestock feed has necessitated intensive investigations into the use of agricultural by-products. Ruminants fed on crop residues and by-products add value to resources which are largely wasted in the absence of a ruminant component in the system. Livestock farmers in northern Nigeria utilize significant quantities of crop residue while their stocks return manure to the soil (Onwuka *et al.*, 1997). According to the work done by Negi *et al.* (1988), low-grade roughages contain a significant proportion of rumen-degradable nitrogen (RDN) that contribute to the nutrition of ruminal microflora.

Crop residues in the summer rainfall areas of South Africa fulfil a strategic role in the fodder flow program of ruminants and can be used to reduce feed costs. Livestock farmers, particularly in the eastern part of this region, rely heavily on crop residues during winter when roughage is scarce and the nutritive value of natural veldt has declined to a very low level (Snyman & Joubert, 2002). The total area of South Africa that has been planted with maize during the 2005/2006 production season, by commercial farmers, was about 1 547 850 hectare. According to the most recent predictions, the commercial production of maize in South Africa for the year 2006 is estimated at 6 238 000 metric ton (Van Zyl, 2006).

It is thus deducible from these production figures that residues (cobs and stover) from the harvesting of maize are one of the most abundant crop residues in this summer rainfall area. However, insufficient information regarding their nutritive value results in inefficient utilization thereof by ruminants and thus sub-optimal animal production. Also, maize residues, as well as whole plant maize residues, is of great value when fed in milled form during times of feed scarcity.

Although some information is available on the nutritive value of low-grade roughages in other countries, these values are not necessarily applicable to crop residues produced and harvested under South African conditions. Ørskov (1989) highlighted the importance of ruminal degradability of a feedstuff for the evaluation of its nutritive value. This study was, therefore, conducted to assess the rumen degradability of maize cobs in sheep under South African conditions.

Materials and methods

Animals

Five ruminally cannulated Dohne Merino wethers were used in the *in sacco* trial. The wethers were housed individually in an enclosed but adequately ventilated shed with a wooden slatted floor. The animals had free access to drinking water. A pelleted complete sheep finisher (manufactured by Meadow Feed Mills, P.O. Box 262, 7620, Paarl, South Africa) was fed *ad libitum* to all the animals. Chemical composition of the pelleted diet is presented in Table 1.

Sample preparation

Maize cobs (without grain) were milled in a hammer mill at a feed manufacturing plant (Senwesko Feeds·Voere, P.O. Box 52, 9520, Viljoenskroon, South Africa). Afterwards, a

sample of the milled maize cobs was sieved through a 4 mm screen. The part of the sample that remained on the screen was used as the coarse maize cob treatment and the part that passed through the screen was used as the fine maize cob treatment. The normal maize cob treatment was cobs that were only milled in the hammer mill and therefore consisted of particles smaller and larger than 4 mm.

Table 1 Chemical composition of the pelleted complete sheep finisher on a dry matter basis

Chemical composition	Content
Organic matter (%)	91.78
Ether extract (%)	3.28
Crude Protein (%)	15.16
Acid detergent fibre (%)	11.37
Neutral detergent fibre (%)	20.69

Marked dacron bags (23 x 10 cm, 53 µm pore size) were dried in an oven at 60°C for 24 h, cooled in a dessicator and weighed. Samples of lucerne hay, oat hay, and the three maize cob treatments were ground in a Wiley mill through a 2 mm screen. A sub-sample of each feedstuff was taken for dry matter (DM) determination. Five grams DM of each of the milled samples were then weighed into the bags. The lucerne hay and oat hay were used to compare the three maize cob treatments with.

After the bags were closed with a nylon string, they were incubated in the rumen for 0, 2, 4, 8, 16, 24, 48 and 72 h. Incubation times longer than 24 h would not leave enough residue for all the chemical analyses and therefore duplicate bags were prepared for the 48 and 72 h incubation times.

The sealed bags were individually attached to the cannula plug with a piece of nylon string. The free length between the plug of the cannula and the bag was 25 cm (Mehrez & Ørskov, 1977). The bags were inserted into the rumen in reverse order, starting with the 72 h incubation samples at 08:00 on the first day. The rest of the bags were inserted on appropriate time intervals over the following days to allow the relevant incubation times. All the bags were removed at 08:00 on the fourth day. The advantages of the reverse order procedure have been discussed by Nocek (1985) and Huntington & Givens (1995).

After the bags were removed from the rumen they were placed into buckets of ice water and then rinsed under running cold tap water to stop microbial activity. The bags were then washed in cold water in a twin-tub washing machine for ten minutes using the gentle cycle.

Water was drained after five minutes of washing and the bags were then washed in fresh water for another five minutes. Bags containing feed samples but which were not submitted to ruminal incubation (0 h) were washed like the other bags to determine the soluble fraction. Bags were then dried in a forced draught oven at 65°C to a constant mass as described by Nocek (1985) and Janicki & Stallings (1988). At the end of the drying period bags were cooled in a dessicator and weighed to calculate the residual DM. Residues were then removed from the bags and stored for further analysis. The contents of the duplicate bags (48 and 72 h) were composited for analysis.

Chemical analysis

Samples of the initial sample and residues from the bags were analysed for dry matter (DM), crude protein (CP), neutral detergent fibre (NDF) and acid detergent fibre (ADF). The samples were analysed for nitrogen, according to the combustion method (Method 990.03, AOAC, 2002) with a Leco FP-428 Nitrogen and Protein Analyzer (Leco Corporation, 3000 Lakeview Avenue, St. Joseph, MI 49085-2396). NDF and ADF were determined with an ANKOM^{220} Fiber Analyzer (ANKOM Technologies, Fairport, NY).

The percentage disappearance at each incubation time of DM, CP, NDF and ADF were calculated using the following equation:

$$\text{Percentage disappearance} = \frac{(\text{g before incubation - g after incubation})}{\text{g before incubation}} * 100$$

Statistical analysis

Ruminal degradability of lucerne hay, oat hay and three maize cob treatments (normal, coarse and fine) was compared using the *in situ* nylon bag technique. The percentage disappearance of DM, CP, NDF and ADF at each incubation time was obtained. Data were analysed by using PROC ANOVA and PROC GLM (General Linear Model procedures) of SAS (2000) which employs the use of Least Square Means for each parameter. Significance was declared at $P \leq 0.05$. There were no significant differences between the animals used in the trial as pertaining to the rate of disappearance of the various constituents and the data was therefore tested only for the main effect of roughage type per time.

Results and discussion

Chemical analysis of the feedstuffs is presented in Table 2. The dry matter (DM) of the three maize cob treatments ranged from 99.01 to 99.57%, crude protein (CP) from 3.14 to 3.54%, neutral detergent fibre (NDF) from 77.62 to 79.79%, and acid detergent fibre (ADF) from 39.98 to 40.13%. Aregheore (1995) reported the following values for maize cobs: 96.00% DM, 4.64% CP, 76.08% NDF, and 49.92% ADF. Variation such as these in nutrient composition could be attributed to variety of maize, stage of maturity at harvest time, soil type, weather conditions and management practices such as level of fertilization (Turgut & Yanar, 2004). According to Von Keyserlingk *et al.* (1996) these factors may influence rumen degradability characteristics as well. A CP value of 3.30% is given by Adeyemi & Familade (2003) for maize cobs, which corresponds to the values in the current study. Although no statistical analysis of feedstuff nutrient composition was conducted, the following trends were apparent. The three maize cob treatments had the highest NDF and ADF content between the treatments, and lucerne hay had the highest amount of CP content. The CP and NDF content of lucerne hay correspond to the values given by the NRC (1996) for late vegetative lucerne hay (21.70 and 39.00% respectively). The CP content of the oat hay was much lower and the NDF content higher than the tabular values from the NRC (1996) for oat hay (9.50 and 63.00% respectively).

Table 2 Chemical composition of feedstuffs used for the *in situ* experiment (on a dry matter basis)

Chemical component	Lucerne hay	Oat hay	NMC[1]	CMC[2]	FMC[3]
Dry matter (%)	89.70	90.73	99.01	99.57	99.14
Crude protein	20.67	5.64	3.16	3.54	3.14
Neutral detergent fibre	37.58	65.74	79.72	77.62	79.79
Acid detergent fibre	28.32	39.33	40.13	39.98	40.00

[1]NMC – normal maize cobs.

[2]CMC – coarse maize cobs.

[3]FMC – fine maize cobs.

The mean values for DM, CP, NDF and ADF disappearance (%) of the individual feedstuffs at different rumen incubation times are presented in Table 3 and the profiles illustrated in Figures 1-4.

Table 3 Comparison of the mean disappearance of DM, CP, NDF and ADF from *in situ* nylon bags containing different types of roughage at different times of ruminal incubation

Feedstuffs	Rumen incubation time (h)							
	Initial	Cumulative additional[1]						
	0	2	4	8	16	24	48	72
Dry matter (DM) disappearance (%)								
Lucerne hay	31.17	6.45^{a}	13.41^{a}	20.13^{a}	25.67^{a}	29.08^{a}	32.44^{a}	35.54^{a}
Oat hay	25.05	3.23^{b}	3.39^{bc}	4.68^{bc}	4.59^{b}	5.69^{b}	7.80^{b}	9.29^{b}
NMC[2]	7.55	5.53^{a}	5.67^{b}	4.70^{bc}	4.40^{b}	6.92^{b}	11.71^{b}	11.44^{b}
CMC[3]	11.89	-1.56^{c}	-0.53^{c}	-0.56^{c}	-0.63^{b}	1.57^{b}	6.55^{b}	6.85^{b}
FMC[4]	7.73	2.85^{b}	4.66^{b}	4.92^{b}	5.03^{b}	5.07^{b}	7.03^{b}	9.96^{b}
SEM		0.645	1.173	1.606	2.058	2.286	2.542	2.919
Crude protein (CP) disappearance (%)								
Lucerne hay	43.84	2.35^{ab}	10.24^{a}	20.07^{a}	25.97^{a}	32.64^{a}	37.98^{a}	40.71^{a}
Oat hay	57.90	4.30^{a}	4.18^{a}	3.39^{b}	2.78^{b}	3.51^{b}	1.94^{b}	-5.57^{b}
NMC	54.65	0.92^{ab}	-4.91^{b}	-11.49^{c}	-19.64^{c}	-27.26^{c}	-41.51^{c}	-50.19^{c}
CMC	68.56	-4.19^{b}	-10.11^{b}	-15.20^{c}	-21.03^{c}	-27.19^{c}	-50.34^{c}	-48.89^{c}
FMC	50.41	-3.58^{b}	-8.99^{b}	-15.46^{c}	-29.43^{c}	-40.19^{c}	-48.26^{c}	-48.37^{c}
SEM		1.135	2.013	3.126	4.576	6.319	7.655	8.434
Neutral detergent fibre (NDF) disappearance (%)								
Lucerne hay	2.69	-3.58^{c}	5.61^{ab}	9.15^{a}	16.43^{a}	18.90^{a}	24.52^{a}	28.35^{a}
Oat hay	6.80	0.24^{bc}	0.23^{c}	2.07^{c}	1.52^{b}	2.55^{c}	6.33^{b}	8.41^{b}
NMC	-5.26	9.07^{a}	8.26^{a}	7.15^{ab}	6.81^{b}	12.90^{ab}	17.39^{ab}	18.09^{ab}
CMC	-1.88	-0.06^{bc}	0.74^{c}	1.36^{c}	2.08^{b}	4.75^{bc}	12.65^{ab}	11.87^{ab}
FMC	-0.55	1.61^{b}	3.04^{bc}	3.94^{bc}	4.53^{b}	4.84^{bc}	7.83^{b}	11.46^{ab}
SEM		1.032	0.802	0.812	1.453	1.816	2.406	2.836
Acid detergent fibre (ADF) disappearance (%)								
Lucerne hay	1.24	4.82^{bc}	10.09^{a}	10.69^{a}	17.34^{a}	22.41^{a}	25.29^{a}	30.51^{a}
Oat hay	2.67	6.96^{abc}	6.34^{ab}	7.85^{ab}	7.10^{bc}	7.81^{b}	10.28^{a}	13.49^{a}
NMC	-14.40	11.15^{a}	10.44^{a}	7.58^{ab}	8.55^{bc}	14.30^{ab}	19.23^{a}	18.73^{a}
CMC	-12.17	2.64^{c}	2.86^{b}	3.87^{b}	3.00^{c}	6.55^{b}	13.88^{a}	13.10^{a}
FMC	-11.14	7.94^{ab}	8.26^{ab}	8.59^{ab}	10.87^{ab}	10.05^{b}	12.52^{a}	16.37^{a}
SEM		0.918	0.969	0.945	1.433	1.736	2.418	2.957

^{a,b,c} Means in the same column, within chemical component, with different superscripts differ ($P \leq 0.05$); Each value is the mean of five determinations;

[1]Mean disappearance (%) of DM, CP, NDF and ADF were corrected with the zero hour disappearance;

[2]Normal maize cobs; [3]Coarce maize cobs; [4]Fine maize cobs.

Because of high variation observed in the disappearance (%) of the five feedstuffs at zero hour (0 h) incubation time, all other disappearance values were corrected with their appropriate initial values for DM, CP, NDF and ADF. DM disappearance (%), which is highly correlated with NDF degradability (Parys *et al.,* 2000, as cited by Akbar *et al.,* 2002), increased with time and at 72 h of ruminal incubation was the highest in lucerne hay (35.54), followed by NMC (11.44), FMC (9.96), oat hay (9.29) and CMC (6.85). At the last four incubation times there were no significant differences in disappearance between the oat hay, normal, coarse or fine maize cobs. In the current study at the 4 and 8 h rumen incubation times, the DM disappearance values for lucerne hay (44.58 and 51.30%, respectively) was higher than the values reported by Turgut & Yanar (2004) (38.4 and 47.1%, respectively). After 16 h the disappearance (%) was similar to the 56.7% reported by these authors at the same incubation time.

According to Nocek & Grant (1987) the soluble and 53 µm filterable DM components are negatively correlated to the concentration of fibrous components. Their results suggest that the rate of ruminal DM digestibility is related to the amount of soluble and 53 µm filterable DM, as well as the fibrous components of forage. The major differences in disappearance rates from dacron bags due to particle size would be established in the first hour of incubation (Kirkpatrick & Kennelly, 1987). Our 0 h data basically support this conclusion.

The results clearly demonstrate that lucerne hay is more rumen degradable than the other four treatments. The low percentage DM and CP disappearance of coarse maize cobs could be due to the formation of Maillard reaction products during the milling process (Batajoo & Shaver, 1998). The coarse maize cobs predominantly consisted of larger particles than the other two maize cob treatments and therefore generated more heat during the milling process. Maillard reaction products have been found to be resistant to digestion *in vivo* (McDonald *et al.,* 1995).

In general, the extent of CP disappearance (%) increased with time in the lucerne hay, and decreased in time in the three maize cob treatments. There were no differences ($P > 0.05$) in the rate of CP disappearance between the maize cob treatments during the rumen incubation times. At 2 h of rumen incubation, a slight disappearance of CP was apparent in normal maize cobs, but coarse and fine maize cobs had a negative disappearance of CP. The negative CP disappearance would seem to indicate that some ruminal bacteria had adhered to roughage particles and were not removed by washing and rinsing (Nocek, 1988). At 4 h of rumen incubation all three the maize cob treatments showed a negative CP disappearance. The extent of negativity went on increasing up to 48 h for coarse and fine maize cobs, and up to 72 h for normal maize cobs. Beyond the 48 h period the extent of negative disappearance

decreased for the coarse and fine maize cobs. Negi *et al.* (1988) suggested that this decrease in extent of negative disappearance is due to the degradation of roughage or absorbed CP. They demonstrated that degradability of CP remained negative up to 12 h of incubation for wheat straw and up to 24 h for rice straw and grass hay. The same was found by Erasmus *et al.* (1990) when CP disappearance of maize fodder, Midmar ryegrass and Smutsfinger grass hay were less after 1 or 2 h of rumen incubation than at 0 h (10-min rinsing).

Several researchers (Kennedy *et al.,* 1984; Nocek, 1985; Nocek & Grant, 1987) have demonstrated that bacterial contamination increased curvilinearly with time of ruminal incubation. According to Nocek (1988), these data suggest that bacteria continually attach to particles up to a particular time of ruminal exposure. Thereafter, bacterial attachment appears to be a function of attachment site availability, i.e., attachment sites are degraded, or substrate availability is limited. The observed increase in CP content per gram DM of roughage represents the sum total of two opposing trends, i.e. decreasing roughage CP and increasing microbial CP contamination (Negi *et al.,* 1988). Concentrate ingredients generally contain little microbial contamination and it seems apparent that low protein roughages and coarse feedstuffs should be corrected for microbial contamination. Nocek & Grant (1987) showed reduced digestion, lag times, less indigestible residue, and faster CP digestion rates when roughages were corrected for bacterial CP contamination. Lindberg (1981) reported high correlations between ruminal CP and NDF disappearance from nylon bags for hays, and suggests that nitrogenous compounds in natural feedstuffs are protected by fibrous structures. With protein-rich feeds, however, the microbial error seems small (Varvikko, 1986).

The CP disappearance (%) values of 54.08 and 63.91 at 4 and 8 h of rumen incubation for lucerne hay in this study was similar to the findings reported by Turgut & Yanar (2004) (54.8 and 64.1%, respectively). They reported an 87.4% CP disappearance for lucerne hay after 72 h, which was higher than the 84.55% in this study. The CP disappearance values of normal maize cobs in the present study after 2, 4 and 8 h incubation periods (55.57, 49.74 and 43.16%, respectively) were higher than the findings of Erasmus *et al.* (1990) for maize stover (36.4, 41.3 and 42.6%, respectively). They also reported a 52.6% CP disappearance for teff hay after 48 h of rumen incubation, whereas a similar disappearance for coarse maize cobs was achieved at 8 h. After 2 h, a disappearance value of 46.0 and 46.2% was reported by Erasmus *et al.* (1990) for kikuyu pasture and sorghum hay respectively. These values were lower than the 55.57% disappearance for normal maize cobs at the same incubation time.

NDF and ADF disappearance (%) followed the same trend, as can be seen in Fig. 3 and 4. At 72 h of ruminal incubation, NDF disappearance (%) were the highest in lucerne hay, followed

by normal, coarse, fine maize cobs and oat hay. In the case of ADF disappearance at the same incubation time, the highest disappearance value was reported for lucerne hay, followed by normal, fine, oat hay and the coarse maize cobs. At 16 h incubation time there were no significant difference in NDF disappearance between the oat hay and the three maize cob treatments, but they differed significantly from lucerne hay. After 48 and 72 h of rumen incubation there were no significant differences in ADF disappearance between the treatments.

In situ degradability estimates are affected by factors such as feed particle size (Nocek, 1985), bag surface area ratio (Mehrez & Ørskov, 1977), sample size, origin of feed, and bag material and pore size (Nocek, 1985), washing procedures (Huntington & Givens, 1997b), sampling schedules (Nocek, 1985; Huntington & Givens, 1997a) and bag mobility (Huntington & Givens, 1997a). These factors may partially explain differences in kinetic measurements between our study and others in the literature.

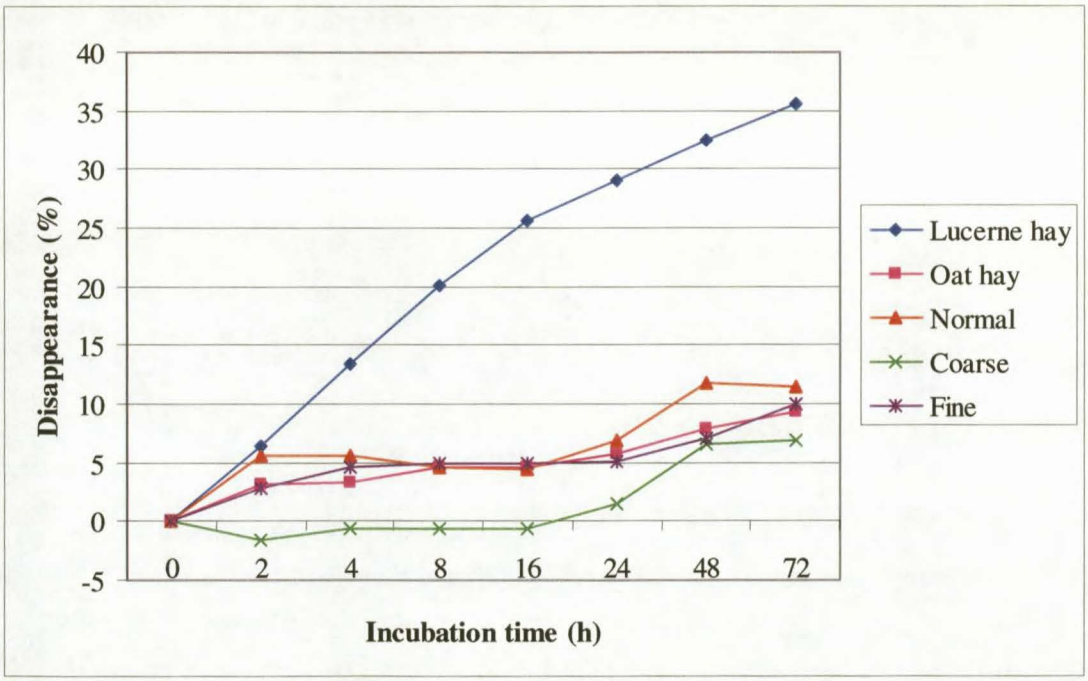

Figure 1 Mean ruminal dry matter disappearance of the five treatments

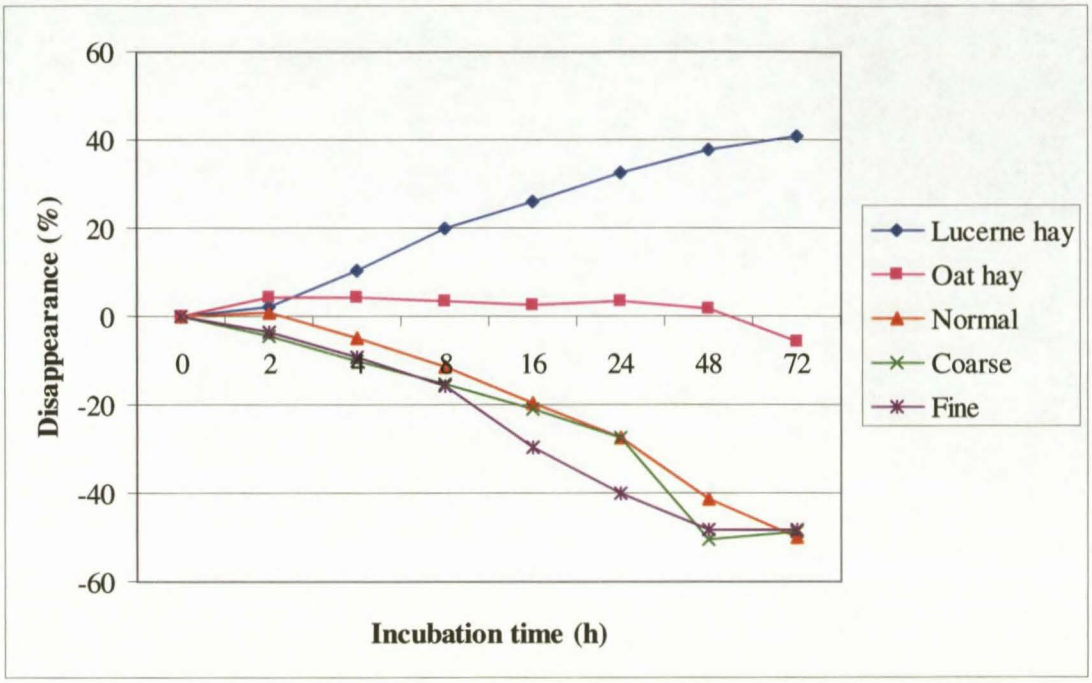

Figure 2 Mean ruminal crude protein disappearance of the five treatments

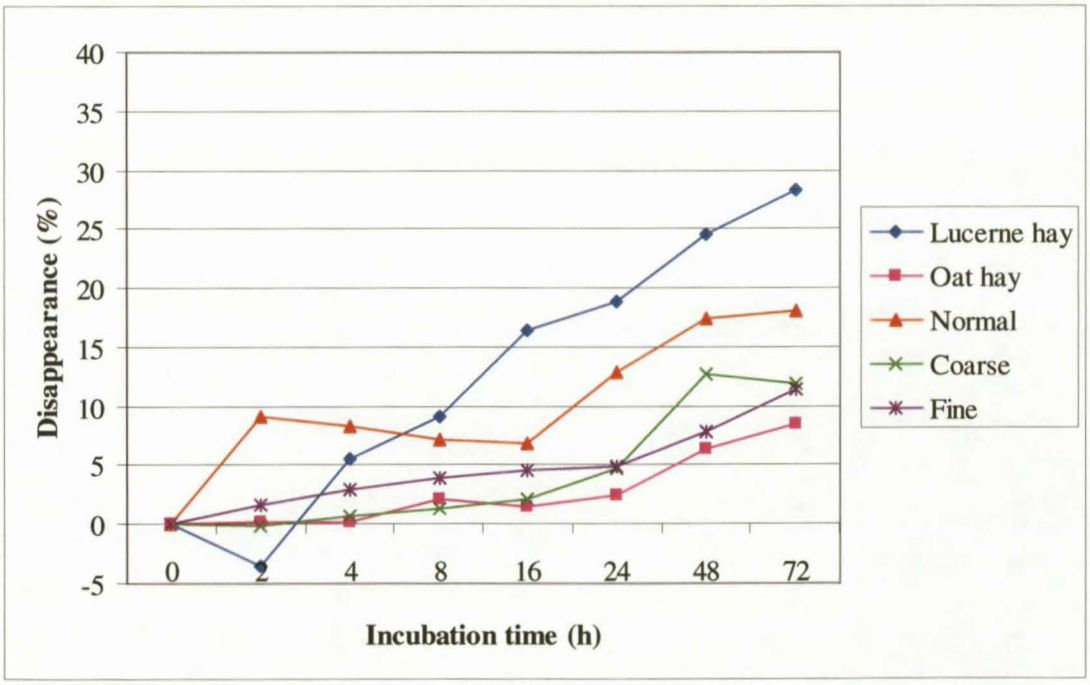

Figure 3 Mean ruminal neutral detergent fibre disappearance of the five treatments

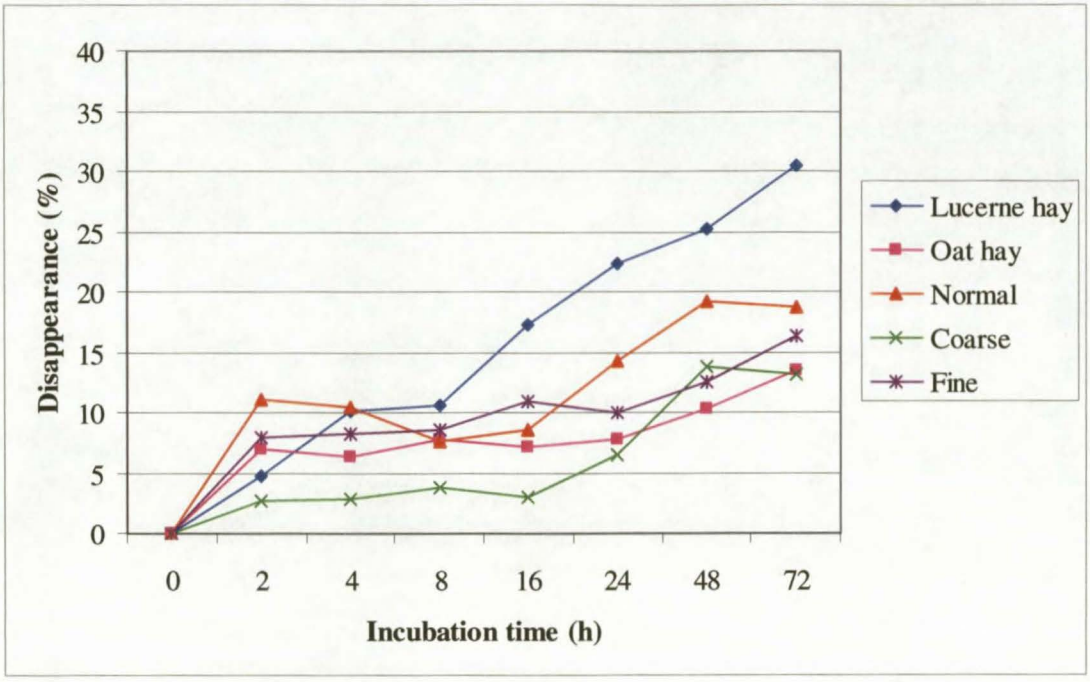

Figure 4 Mean ruminal acid detergent fibre disappearance of the five treatments

Conclusion

Results obtained from this study clearly established that, except at the 2 h incubation time, there were no difference ($P > 0.05$) between normal maize cobs and oat hay for DM disappearance (%). Normal maize cobs and oat hay did not differ ($P > 0.05$) in NDF disappearance (%) at the 16, 48 and 72 h incubation times. It is evident that in the case of ADF disappearance (%) there were no significant difference between normal maize cobs and oat hay at all the incubation times. Except at the 2 h incubation time, normal maize cobs differed ($P \leq 0.05$) from oat hay for CP disappearance (%), as the three maize cob treatments elicited a negative CP disappearance.

It can be concluded that on average, the degradability of milled maize cobs, especially of maize cobs consisting of particles smaller and larger than 4 mm, normal maize cobs, is comparable with that of oat hay. Maize cobs, therefore, can be used as a source of roughage for inclusion in diets of finishing lambs.

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Chapter 3

Effect of maize cobs as roughage source in feedlot finishing diets on production efficiency of South African Mutton Merino lambs

Abstract

Maize cobs were evaluated as a roughage source in the finishing diets of 24 South African Mutton Merino (SAMM) lambs. Three diets were formulated on an isonitrogenous and isoenergetic basis (15.50% crude protein (CP) and a metabolizable energy content of 10.64 MJ/kg on a dry matter (DM) basis) that contained maize cobs at a rate of 4, 8 and 12% (Diet 1, 2 and 3, respectively). The lambs were fed *ad libitum* for 24 days from an average 34.9 ± 2.64 kg to 44.1 ± 3.04 kg body weight. Individual feed consumption, utilisation, growth and digestion efficiency were investigated. No differences occurred in the growth performance and feed conversion efficiency of the lambs consuming the three different diets. Lambs fed the diet that contained 4% maize cobs had the higher nitrogen retention, while the energy retention was not affected by diet. It is concluded that an increase in the inclusion of maize cobs up to a level of 12% in lamb feedlot finishing diets has no adverse affect on their production performance.

Key words: maize cobs; feed efficiency; South African Mutton Merino lambs; feedlot

Introduction

In Africa, a significant part of the total wealth of poor families is comprised of sheep and goats (Peacock, 1996), which are also their primary source of meat and milk. These flocks are raised under a wide variety of extreme ecological areas and are able to survive and produce in environments too harsh for cattle (El Khidir *et al.,* 1998), effectively utilizing large parts of hostile agricultural land. In many tropical areas of Africa, for example Ghana, there is a problem of inadequate feed supply for animals during the dry seasons (Tuah & Ørskov, 1989). Agricultural wastes and by-products, such as maize cobs, can be fed to ruminants during these seasons to offset the detrimental effects of inadequate nutrition on reproduction and growth (Tuah & Ørskov, 1989). Under such circumstances, with limited feed and water resources, the efficient utilization of available nutrients is essential.

Raising lambs under extreme conditions with inadequate natural grazing, or when available forage is of poor quality, results in higher mortality rates and slower growth rates. This poor performance is mainly due to a nitrogen and energy deficiency in the rumen, which has an inhibitory effect on substrate fermentation and impairs microbial protein synthesis (Chakeredza *et al.,* 2001). In such conditions lambs may have to be weaned earlier and finished in a feedlot (Santos-Silva *et al.,* 2002). By changing the production system and to make use of feedlots, the focus changes to increasing animal productivity and economic results, and to obtain better quality meat (Santos-Silva *et al.,* 2002). In meat production systems, a small increase in slaughter weight of lambs, may result in higher productivity, and will give more flexibility to the production system (Sañudo *et al.,* 1996). The economy of a feedlot requires that animals be slaughtered at a targeted weight or after a certain number of days in the feedlot (Sheridan, 2001).

The South African Mutton Merino (SAMM) is a dual-purpose (mutton and wool) sheep breed and was selected for the study due to its high growth rate and capability to produce a slaughter lamb at an early age with good meat quality attributes (Cloete *et al.,* 2004). It has a high fertility, good conformation, good feed conversion ratio and adaptability and is therefore popular in feedlot production systems (Neser *et al.,* 2000).

There is little information available on the effect of maize cobs on production efficiency of lambs. Crop residues (cobs and stovers) from the harvesting of maize are one of the most abundant crop residues produced annually in the summer rainfall area of South Africa, therefore this study was conducted to determine the effect of maize cobs in feedlot finishing diets on the feed intake, growth parameters and digestion of SAMM lambs.

Materials and methods

Animals and feeding trial

Twenty four South African Mutton Merino (SAMM) intact male lambs were used in this investigation. All the lambs were weaned and shorn prior to entering the feedlot at an age of approximately 6 months. All the animals were dewormed with a broad-spectrum drench to eliminate all internal parasites on their arrival at the University of Stellenbosch experimental farm, Welgevallen. The average initial body weight (BW) was 34.9 ± 2.64 kg. The lambs were randomly allocated into individual pens (1 m x 2 m) in an enclosed but adequately ventilated shed with a wooden slatted floor. Lambs were divided in three groups of equal

Table 1 Physical and chemical composition of the three diets fed to the SAMM lambs

	Maize cobs inclusion level		
	4%	8%	12%
	Diet 1	Diet 2	Diet 3
Physical composition[1]			
Maize meal	21.80	23.00	28.78
Lucerne hay	17.00	17.00	17.00
Citrus pulp	4.00	4.00	2.50
Maize cobs	4.00	8.00	12.00
Hominy Chop	24.74	25.44	25.00
Sunflower hulls	2.80	0.00	0.00
Wheat bran	12.40	9.10	0.00
Full-fat soybeans	2.00	2.00	3.00
Ammonium chloride	0.50	0.50	0.50
Limestone	1.30	1.40	1.30
Mono calcium phosphate	0.00	0.00	0.16
Salt	0.60	0.60	0.60
Urea	0.60	0.70	0.90
Molasses	7.50	7.50	7.50
Premix B[3]	0.25	0.25	0.25
Acid Buf[4]	0.50	0.50	0.50
Avatec[5]	0.02	0.02	0.02
Chemical composition[2]			
Organic matter (%)	92.43	93.05	93.13
Ether extract (%)	4.01	4.29	4.07
Crude Protein (%)	15.15	15.70	15.63
Acid detergent fibre (%)	17.89	16.21	16.10
Neutral detergent fibre (%)	32.89	30.66	30.42
Gross energy (MJ/kg)	18.13	18.29	18.20
Metabolizable energy (MJ/kg)[6]	10.64	10.66	10.62

[1]On an air dry basis; [2]Analysed values on a DM basis; [3]A standard mineral (macro and micro) and vitamin supplement; formulated and supplied by BASF Animal Nutrition (P.O. Box 1783, 1620, Kempton Park, South Africa); [4]A rumen buffer; supplied by Nutec Southern Africa (234 Royston Road, Willowton, Pietermaritzburg, KwaZulu-Natal, South Africa); [5]A growth promoter, lasalocid sodium; supplied by Instavet Import and Export Pty (Ltd) (P.O. Box 346, 2163, Kya Sand, South Africa); [6]Based on the nutritive value of feeds according to laboratory determined values by Senwesko Feeds.

initial weights. Each group was randomly assigned to one of three treatments. The experimental diets contained maize cobs at a rate of 4, 8 and 12% (Diet 1, 2 and 3, respectively; Table 1). Maize cobs used in this study refers to the normal maize cobs used in the *in situ* degradability study in Chapter 2.

The pelleted diets were formulated according to the NRC (1985) recommendations for lambs, on an isonitrogenous and isoenergetic basis. The lambs had *ad libitum* access to the feed and fresh water. They were fed twice daily, for 24 days, at 08:00 and 16:00 and weighed weekly to determine the average daily gain (ADG) for each lamb. Feed refusals were collected daily in the morning before the provision of new feed and water, pooled and weighed weekly to calculate voluntary feed intake, as well as feed conversion efficiency (FCE) (kg feed/kg BW gain) for each lamb.

Digestibility trial

At the end of the feeding trial, eighteen of the lambs were used to determine the nutrient digestibility and nitrogen (N) as well as energy retention of the three diets. For the duration of the digestibility trial, the other six lambs were maintained on the diet. Faeces were collected twice daily, for 7 days, at 10:00 and 16:00 whilst the urine was collected with the morning feeding. Twenty mℓ of urine preservative (80 g potassium dichromate and 20 g mercuric chloride dissolved in 1 ℓ distilled water) were added each morning to the urine collection jugs to prevent volatilization of ammonia from the urine. Daily sub-samples of the faeces (10%) and urine (5%) from each lamb were pooled over the whole period, prior to chemical analysis.

Methane gas production (MJ/day) was calculated as 8% of the gross energy intake (McDonald *et al.*, 1995). Nitrogen retention was corrected both for endogenous urinary N (EUN) and metabolic faecal N (MFN) according to McDonald *et al.* (1988) as follows:

EUN (g) = 0.18 g N/kg $BW^{0.75}$/day

MFN (g) = 5 g N/kg dry matter intake

N retention (g N/kg $BW^{0.75}$/day) = $\{N_{intake} - (N_{faeces} - MFN) - (N_{urine} - EUN)\}/BW^{0.75}$/days

At the end of the 7-day digestibility trial, the twenty four SAMM lambs were slaughtered at a commercial abattoir using standard South African procedures. The fasted (16 hrs) average live weight (BW) of the lambs were 42.3 ± 4.22 kg as determined prior to slaughter. After being electrically stunned (4 seconds at 200 volts) the lambs were exsanguinated and the carcasses suspended by the Achilles tendon to bleed. After evisceration, the weight of the

warm carcass of each lamb was recorded. The carcasses were then suspended in a cooler at 2°C and weighed after 48 h to determine the moisture loss in the cooler. Dressing percentage was calculated from the cold carcass weight as a percentage of live BW. No electrical stimulation of the carcasses was applied.

Chemical analysis

The faeces were dried at 60°C for 96 h, air-equilibrated and weighed. The feed and faeces were ground through an 1 mm screen and analysed for dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fibre (NDF), acid detergent fibre (ADF) and gross energy (GE) content (AOAC, 2002). The urine samples were analysed for N and gross energy content (AOAC, 2002). All nitrogen analyses were performed according to the combustion method (Method 990.03, AOAC, 2002) with a Leco FP-428 Nitrogen and Protein Analyzer (Leco Corporation, 3000 Lakeview Avenue, St. Joseph, MI 49085-2396). NDF and ADF were determined with an ANKOM^{220} Fiber Analyzer (ANKOM Technologies, Fairport, NY, USA) and GE by adiabatic oxygen bomb calorimetry (CP500 calorimeter).

Statistical analysis

The effect of the incremental inclusion of maize cobs as a roughage source in feedlot finishing diets of SAMM lambs on production efficiency were analysed by using PROC ANOVA and PROC GLM of SAS (2000). Least Square Means were calculated for all effects. Significance was declared at $P \leq 0.05$.

Results and discussion

The growth parameters of the SAMM lambs are presented in Table 2. Feed intake, ADG and FCE were not affected ($P > 0.05$) by dietary treatment. Sheridan *et al.* (2003) observed results similar to that recorded for this investigation as pertaining to ADG and FCE (0.330 kg/day and 5.56 kg feed/kg BW gain, respectively) for SAMM lambs receiving a similar diet at an equivalent initial live weight. Brand *et al.* (2001), on the other hand, found a lower ADG, but a more efficient FCE (0.267 kg/day and 4.80 kg feed/kg BW gain, respectively) for SAMM lambs in a feedlot receiving a diet (15.60% CP) that had been supplemented with 6% canola. The fact that no significant differences occurred in any of the growth parameters may be ascribed to the isonitrogenous and isoenergetic state of the three experimental diets (Table 1).

In terms of cold carcass weight and dressing percentage, no significant differences were observed for the lambs on the three different diets. The values for cold carcass weight and dressing percentage in the present study are, however, higher than that (19.42 ± 0.777 kg and 47.02 ± 0.647%, respectively) reported by Sheridan *et al.* (2003) for SAMM lambs weighing 42.20 ± 1.013 kg at slaughter. The dressing percentage (41%) given by Brand *et al.* (2001) for SAMM lambs was also lower than the dressing percentage found in the current study. The differences that occurred in dressing percentage between these two authors and the current study may be due to a difference in the level of gut fill at the time of recording the final body weights (Kirton *et al.*, 1995). Skin weight may also affect dressing percentage when the live weights are recorded (Cloete *et al.*, 2004). If the lambs are shorn prior to entering the feedlot or slaughtering, it will lead to a lower skin weight and thus a higher dressing percentage. As sheep approaches maturity and fat is accumulated in the carcass, the dressing percentage will again increase (Warmington & Kirton, 1990).

Table 2 Growth parameters, average feed intake and conversion efficiency of South African Mutton Merino lambs receiving the three different feedlot diets (LSMean ± SE)

	Maize cobs inclusion level		
	4%	8%	12%
	Diet 1	Diet 2	Diet 3
Initial body weight (kg)	34.56 ± 0.993	35.00 ± 1.090	35.13 ± 0.817
Final body weight (kg)	43.69 ± 1.206	44.06 ± 1.120	44.44 ± 1.020
Body weight gain (kg)	9.13 ± 0.532	9.06 ± 0.427	9.31 ± 0.432
Cumulative feed intake (kg)	47.45 ± 1.750	47.95 ± 1.481	47.73 ± 1.454
Daily feed intake (kg)	1.98 ± 0.073	2.00 ± 0.062	1.99 ± 0.061
ADG (kg/day)	0.380 ± 0.022	0.378 ± 0.018	0.388 ± 0.018
FCE (kg feed/kg BW gain)	5.29 ± 0.301	5.38 ± 0.299	5.16 ± 0.114
Body weight at slaughter (kg)	41.94 ± 1.361	42.31 ± 1.839	42.56 ± 1.428
Warm carcass weight (kg)	22.16 ± 0.676	22.31 ± 0.755	22.23 ± 0.663
Cold carcass weight (kg)	21.86 ± 0.681	21.89 ± 0.751	21.83 ± 0.641
Moisture loss (%)	1.37 ± 0.290	1.91 ± 0.198	1.79 ± 0.162
Dressing percentage (%)	52.17 ± 0.493	51.91 ± 0.774	51.35 ± 0.676

Table 3 shows the apparent digestibility of the different components of the three diets. There were no differences ($P > 0.05$) between treatments for the apparent digestibility examined. Although no significant difference were detected in DM and OM digestibility with each incremental inclusion of maize cobs, these digestibilities tended to be the highest in Diet 2 (74.00 and 75.40% respectively) and the lowest in Diet 1 (72.42 and 73.67% respectively). A

possible reason for the lower digestibility of DM and OM in Diet 1 may be due to the higher dry matter (g/day) and energy intake (MJ/day) during the digestibility trial period. Higher feed intakes usually lead to increased passage rates and reduced rumen retention times, resulting in decreased digestibility (Ørskov & McDonald, 1979).

There were no significant differences in ADF, nor NDF digestibility between the three diets. Nor did the three diets differ ($P > 0.05$) in crude fat digestibility. This indicates that the lambs digested the ADF, NDF and crude fat fractions of the three diets with similar efficiency and suggests that the composition of the microbial population of the lambs was relatively similar when fed the three different diets (Nolte & Ferreira, 2004). The mean ADF and NDF digestibility values (39.52 and 45.79% respectively) obtained in this study were lower than the corresponding values (52.66 and 62.46% respectively) observed by Sheridan *et al.* (2003) for SAMM lambs. Nangole *et al.* (1983) reported an ADF digestibility value of 53.0%, which is also higher than that obtained in the current study, for a diet consisting of 87.5% maize cobs fed to Romney Marsh wethers. Although rumen pH was not measured in this investigation, Haddad & Husein (2004) found that ADF and NDF digestibility is adversely affected by low ruminal pH levels. Therefore, the low ADF and NDF digestibility observed in the current study might be attributed to low ruminal pH levels. Haddad *et al.* (1995) stated that a ruminal pH below 6.2 increases the lag time and decreases the extent of digestion for wheat straw fibre. When grain are introduced into the diet of ruminants fed predominately forage, the pH will decline, the number of amylolytic bacteria tend to increase and the number of fibrolytic bacteria tend to decrease (Henning *et al.*, 1980). Thus, production of fibrolytic enzyme systems by the microbial population and the rate of fibre digestion are likely to decrease (Chakeredza *et al.*, 2001).

Table 3 Apparent digestibility (mean) of the diets fed to the South African Mutton Merino lambs

	Maize cobs inclusion level			SEM
	4%	8%	12%	
Chemical component (%)	Diet 1	Diet 2	Diet 3	
Dry Matter	72.42	74.00	72.48	0.638
Organic Matter	73.67	75.40	73.99	0.658
Acid detergent fibre	39.41	39.28	39.88	2.124
Neutral detergent fibre	43.92	48.53	44.92	1.743
Ether extract	93.50	91.82	92.01	0.394

Energy intake (MJ/day), excretion (MJ/day) and retention (MJ/day) and metabolizable energy content (ME, MJ/kg) of the three different diets are presented in Table 4. No statistical differences ($P > 0.05$) were observed in DM and energy intake and energy excreted as faecal energy, urinary energy and methane gas, for the lambs on the three different diets. Although there were no significant differences between the energy intake and total energy excreted among the three diets, the lambs on Diet 1 tended to have the highest energy intake and total energy excreted (23.28 MJ/day and 9.20 MJ/day, respectively), while the lambs on Diet 2 had the least total energy excreted (8.59 MJ/day). This numerically higher total energy excreted for Diet 1 might be due to their higher daily dry matter and energy intake. A consequence of this higher daily DM intake was that more readily fermentable carbohydrates were available for fermentation in the rumens of the lambs in this treatment, resulting in a higher methane gas production (Nolte & Ferreira, 2004). Urinary energy excretion as a percentage of energy intake did not differ ($P > 0.05$) among the diets, suggesting that the absorbed nutrients were metabolized with similar efficiency for all the lambs (Nolte & Ferreira, 2004).

Table 4 Energy metabolism (mean) of South African Mutton Merino lambs fed the three different diets

	Maize cobs inclusion level			SEM
	4%	8%	12%	
	Diet 1	Diet 2	Diet 3	
Dry matter intake (g/day)	1283.61	1222.92	1197.22	62.044
Gross energy (MJ/kg)	18.133	18.293	18.196	
Energy intake (MJ/day)	23.28	22.37	21.79	1.129
Faecal energy (MJ/day)	6.59	5.92	6.14	0.409
Urinary energy (MJ/day)	0.75	0.88	0.83	0.066
Methane gas production (MJ/day)	1.86	1.79	1.74	0.090
Total energy excreted (MJ/day)	9.20	8.59	8.71	0.528
Faecal energy (% of energy intake)	27.87	26.35	27.88	0.619
Urinary energy (% of energy intake)	3.12	4.01	3.92	0.259
Total energy excreted (% of energy intake)	38.99	38.37	39.80	0.605
Energy retention (MJ/day)	14.08	13.78	13.07	0.630
Energy retention (% of energy intake)	61.01	61.63	60.20	0.605
Metabolizable energy (MJ/kg)	11.06	11.27	10.95	0.111

The efficiency of energy retention was similar (P = 0.8176) for the lambs on Diets 1 to 3, and when expressed as a percentage of energy intake, the diets also did not differ significantly from each other. Since the efficiency of energy retention was similar for the lambs in the three different treatments, the calculated metabolizable energy (ME) content of the diets did not differ between treatments (P = 0.5169) (Nolte & Ferreira, 2004). In their study on Awassi lambs, Al Jassim *et al.* (1996) observed that BW gain was highly correlated with ME retention. In our study, ME retention was similar among all treatments which explains the similar ADG for lambs fed on the three experimental diets.

The ME-values obtained in the digestibility study differs from those calculated from the nutritive values of the feeds (Table 1) (Diet 1: 11.06 *vs.* 10.64; Diet 2: 11.27 *vs.* 10.66 and Diet 3: 10.95 *vs.* 10.62 MJ/kg DM). Therefore, it seems that the calculated ME-values of diets formulated for sheep, cannot under all circumstances be used as an indicator of the real ME-values.

Nitrogen metabolism and retention of the SAMM lambs on the three diets can be seen in Table 5. The numerical higher daily nitrogen intake (g/day) of the lambs on the diet that contained 4% maize cobs (Diet 1) is consistent with their dry matter intake. Faecal nitrogen excretion was similar for the three treatments (P = 0.9561), suggesting a relative similar nitrogen digestion (Nolte & Ferreira, 2004). Urinary and total N excretion (g/day) tended to be lower for lambs on Diet 1 but, when expressed as a percentage of N intake, these lambs excreted significantly less N than those fed the other two diets.

When N retention (g N/kg $BW^{0.75}$/day) was corrected for both endogenous urinary N (EUN) and metabolic faecal N (MFN), there was no significant difference in N retention between Diets 1 to 3. The similar N retention of the lambs in the three different treatments was also reflected in the similar average daily gains obtained during the feeding trial (Stanford *et al.*, 1999). The N retention value obtained for lambs on Diet 1 corresponds to that (1.21 g N/kg $BW^{0.75}$/day) observed by Sheridan *et al.* (2003) for SAMM lambs in a feedlot receiving a similar type of diet (14.56% CP on a DM basis). When N retention was expressed as a percentage of N intake, the lambs fed Diet 1 retained significantly more N than lambs fed the other two diets.

Table 5 Nitrogen metabolism (mean) of South African Mutton Merino lambs fed the three different diets

	Maize cobs inclusion level			SEM
	4%	8%	12%	
	Diet 1	Diet 2	Diet 3	
Dry matter intake (g/day)	1283.61	1222.92	1197.22	62.044
Nitrogen intake (g/day)	31.11	30.73	29.93	1.530
Nitrogen intake (g N/kg BW0.75/day)	1.87	1.84	1.79	0.079
Faecal nitrogen (g/day)	7.54	7.18	7.48	0.498
Urinary nitrogen (g/day)	12.37	17.60	17.37	1.210
Total nitrogen excreted (g/day)	19.90	24.77	24.85	1.479
Faecal nitrogen (% of nitrogen intake)	23.84	22.88	24.80	0.536
Urinary nitrogen (% of nitrogen intake)	38.93^{b}	60.01^{a}	59.16^{a}	3.989
Total nitrogen excreted (% of nitrogen intake)	62.77^{b}	82.90^{a}	83.96^{a}	3.891
Metabolic faecal nitrogen (g/day)	6.42	6.11	5.99	0.310
Endogenous urinary nitrogen (g/day)	2.97	2.98	3.01	0.041
Nitrogen retention (g N/kg BW0.75/day)	1.24	0.90	0.84	0.085
Nitrogen retention (% of nitrogen intake)	67.68^{a}	47.38^{b}	46.38^{b}	3.800

^{a,b,c} Means in the same row with different superscripts differ ($P \leq 0.05$).

Conclusion

The incremental inclusion of maize cobs in the feedlot diets, which were formulated on an isonitrogenous and isoenergetic basis, had no noticeable effect on the growth performance and feed conversion efficiency of the SAMM lambs at a growth rate of *ca.* 0.380 kg/day. There were no apparent differences ($P > 0.05$) in the digestibility of any of the chemical components of the three diets between the lambs, suggesting that the diets had been digested with the same efficiency. No significant differences between diets were observed in terms of energy retention, but lambs on Diet 1 had a significantly higher nitrogen retention, expressed as a percentage of nitrogen intake, than the lambs on the other two diets.

Based on the results obtained in the present trial, it may be inferred that the inclusion of maize cobs up to 12% in a finishing diet of lambs did not adversely affect the production of the animals. Maize cobs, therefore, is an excellent and cheap source of roughage for inclusion in diets of finishing lambs.

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Chapter 4

Effect of maize cobs as roughage source in feedlot finishing diets on proximate chemical composition and physical meat quality characteristics of South African Mutton Merino lambs

Abstract

Maize cobs was evaluated as a roughage source in the finishing diets of 24 South African Mutton Merino (SAMM) lambs. Three diets were formulated on an isonitrogenous and isoenergetic basis (15.50% crude protein (CP) and a metabolizable energy content of 10.64 MJ/kg on a dry matter (DM) basis) that contained maize cobs at a rate of 4, 8 and 12% (Diet 1, 2 and 3, respectively). The lambs were fed *ad libitum* for 24 days, after which the animals were slaughtered. The *longissimus dorsi* and the *biceps femoris* muscles from the right hand side of each carcass were removed. Proximate chemical analyses were carried out on both muscles, while the physical measurements were only conducted on the *M. longissimus dorsi.* Diet did not have a significant effect on the proximate chemical composition of both muscles. However, lambs fed the diet containing 4% maize cobs produced a darker (lower L* value) *longissimus dorsi* muscle with a significant lower cooking loss. It can be concluded that an increase in the inclusion of maize cobs up to a level of 12% in lamb feedlot finishing diets has no adverse affect on the meat quality.

Key words: maize cobs; South African Mutton Merino lambs; physico-chemical attributes

Introduction

In Africa, small ruminants (sheep and goats) are an integral part of small-holder farming systems. In these circumstances, sheep and goats significantly contributes to the total farm income, stability of farming system and human nutrition (Devendra, 1994). According to Sañudo *et al.* (1998) the meat of small ruminants, comprises a large proportion of protein foods in many areas of the developed world. Yet, Babiker *et al.* (1990) stated that about 60% of the world population, particularly in developing countries, are thought to be suffering from some sort of animal protein shortage.

The quality of livestock production are determined to a great extent by quantity and composition of reserve fats. In ruminants, the formation and deposition of fats starts only after a certain age, depending on sex, physiological state and feeding conditions (Banskalieva, 1996). In intensively growing lambs, the protein:energy ratio and composition of the diet are of significant importance for creating the optimum solution for quantity and composition of depot fats (Shindarska *et al.*, 1990, as cited by Banskalieva, 1996).

The South African Mutton Merino (SAMM) is a dual-purpose (mutton and wool) sheep breed and was selected for the study due to its high growth rate and capability to produce a slaughter lamb at an early age with good meat quality attributes (Cloete *et al.*, 2004). It has a high fertility, good conformation, good feed conversion ratio and adaptability and is therefore popular in feedlot production systems (Neser *et al.*, 2000).

The use of maize cobs in finishing feedlot diets was shown to have no negative effect on the production performance of SAMM lambs, however, the effect of this roughage source on the proximate chemical composition and physical meat quality characteristics require elucidation. In the summer rainfall area of South Africa, crop residues (cobs and stovers) from the harvesting of maize are one of the most abundant crop residues produced annually, therefore this study was conducted to determine the effect of maize cobs in feedlot finishing diets on the proximate chemical composition and physical meat quality characteristics feed intake, growth parameters and digestion of SAMM lambs.

Materials and methods

Animals and sampling

Twenty four South African Mutton Merino (SAMM) intact male lambs were used in this investigation. All the lambs were weaned and shorn prior to entering the feedlot at an age of approximately 6 months. All the animals were dewormed with a broad-spectrum drench to eliminate all internal parasites on their arrival at the University of Stellenbosch experimental farm, Welgevallen. The average initial body weight (BW) was 34.9 ± 2.64 kg. The lambs were randomly allocated into individual pens (1 m x 2 m) in an enclosed but adequately ventilated shed with a wooden slatted floor. Lambs were divided in three groups of equal initial weights. Each group was randomly assigned to one of three treatments. The experimental diets contained maize cobs at a rate of 4, 8 and 12% (Diet 1, 2 and 3, respectively; Table 1). Maize cobs used in this study refers to the normal maize cobs used in the *in situ* degradability study in Chapter 2.

Table 1 Physical and chemical composition of the three diets fed to the SAMM lambs

	Maize cobs inclusion level		
	4%	8%	12%
	Diet 1	Diet 2	Diet 3
Physical composition[1]			
Maize meal	21.80	23.00	28.78
Lucerne hay	17.00	17.00	17.00
Citrus pulp	4.00	4.00	2.50
Maize cobs	4.00	8.00	12.00
Hominy Chop	24.74	25.44	25.00
Sunflower hulls	2.80	0.00	0.00
Wheat bran	12.40	9.10	0.00
Full-fat soybeans	2.00	2.00	3.00
Ammonium chloride	0.50	0.50	0.50
Limestone	1.30	1.40	1.30
Mono calcium phosphate	0.00	0.00	0.16
Salt	0.60	0.60	0.60
Urea	0.60	0.70	0.90
Molasses	7.50	7.50	7.50
Premix B[3]	0.25	0.25	0.25
Acid Buf[4]	0.50	0.50	0.50
Avatec[5]	0.02	0.02	0.02
Chemical composition[2]			
Organic matter (%)	92.43	93.05	93.13
Ether extract (%)	4.01	4.29	4.07
Crude Protein (%)	15.15	15.70	15.63
Acid detergent fibre (%)	17.89	16.21	16.10
Neutral detergent fibre (%)	32.89	30.66	30.42
Gross energy (MJ/kg)	18.13	18.29	18.20
Metabolizable energy (MJ/kg)[6]	10.64	10.66	10.62

[1]On an air dry basis; [2]Analysed values on a DM basis; [3]A standard mineral (macro and micro) and vitamin supplement; formulated and supplied by BASF Animal Nutrition (P.O. Box 1783, 1620, Kempton Park, South Africa); [4]A rumen buffer; supplied by Nutec Southern Africa (234 Royston Road, Willowton, Pietermaritzburg, KwaZulu-Natal, South Africa); [5]A growth promoter, lasalocid sodium; supplied by Instavet Import and Export Pty (Ltd) (P.O. Box 346, 2163, Kya Sand, South Africa); [6]Based on the nutritive value of feeds according to laboratory determined values by Senwesko Feeds.

The pelleted diets were formulated according to the NRC (1985) recommendations for lambs, on an isonitrogenous and isoenergetic basis. The lambs had *ad libitum* access to the feed and fresh water. After the 24-day feeding trial and 7-day digestibility trial, the lambs were slaughtered at a commercial abattoir using standard South African procedures. The fasted (16 hrs) average live weight (BW) of the lambs were 42.3 ± 4.22 kg as determined prior to slaughter. After being electrically stunned (4 seconds at 200 volts) the lambs were exsanguinated and the carcasses suspended by the Achilles tendon to bleed. After evisceration, the carcasses were suspended in a cooler at 2°C for 48 h. No electrical stimulation of the carcasses was applied.

Forty-five minutes post-mortem the initial muscle pH (pH_{45}) and temperature ($Temp_{45}$) and after 48 h, the ultimate pH (pH_{48}) and temperature ($Temp_{48}$) were measured with a penetrating glass electrode on a hand-held Crison pH/mV-507 meter. The measurement was taken from within the carcass between the 2nd and 3rd last thoracic vertebrae, 45 mm from the midline. The pH meter was re-calibrated after every fourth reading and the electrode rinsed with distilled water between each measurement. The pH meter contained a temperature probe ensuring automatic adjustment of the pH for temperature. The *M. longissimus dorsi* was removed from the left and right hand side of the carcass (the muscle was removed between the 2nd–3rd last thoracic vertebrae and the 4th–5th lumbar vertebrae) and the *M. biceps femoris* was dissected from the right hind leg to assess measurements of drip loss, cooking loss, meat colour and Warner-Bratzler shear force (WBSF) values. The subcutaneous fat from the removed *M. longissimus dorsi* was used for fat colour measurements. Back-fat thickness was measured with a digital calliper at a site 25 mm off the midline at the 2nd–3rd last thoracic vertebrae and the 4th–5th lumbar vertebrae (where the *M. longissimus dorsi* was removed).

Proximate chemical analysis

Proximate chemical analysis was carried out on the *M. longissimus dorsi* and the *M. biceps femoris* from the right hand side of the carcass. The *M. longissimus dorsi* from the left hand side of the carcass was used for sensory analyses (Chapter 6). The frozen muscle samples were cut into smaller portions, minced three times through a 2 mm sieve to ensure homogeneity, and analysed chemically. Proximate composition was determined according to AOAC methods (AOAC, 2002). The analysis included determination of moisture, ash, protein (N × 6.25) and fat content. The nitrogen analysis were performed on dried, fat free samples, according to the combustion method (Method 990.03, AOAC, 2002) with a Leco FP-428 Nitrogen and Protein Analyzer (Leco Corporation, 3000 Lakeview Avenue, St. Joseph, MI 49085-2396). The moisture content was determined by drying at 100°C for 24 h

and ashing was done at 500°C for 5 h. The lipid content was determined by means of chloroform:methanol (1:2 v/v) extraction according to the method of Lee *et al.* (1996).

Physical analysis

All the physical measurements were conducted on the *M. longissimus dorsi* after the two muscles (*M. longissimus dorsi* and *M. biceps femoris*) had been removed from the carcasses and transported to the laboratory.

For the drip loss determination 1.5 cm thick meat samples, cut perpendicular to the longitudinal axis of the muscle on the caudal side of the removed *M. longissimus dorsi*, were weighed. The samples were placed in netting and then suspended in an inflated plastic bag, ensuring that the sample did not come into contact with the exudate. After a storage period of 24 h at 4°C, the samples were blotted dry with absorbent paper, weighed again and the drip loss was calculated as weight loss expressed as a percentage of the original weight of the sample (Honikel, 1998).

Cooking loss of the *M. longissimus dorsi* was determined by placing weighed, freshly cut samples (1.5 cm thick) in thin-walled plastic bags in a temperature-controlled water bath (preset at 80°C) for 1 h. The samples were then removed from the water bath, cooled in cold water, blotted dry and weighed. Cooking loss was calculated as the difference in sample weight before and after cooking, expressed as a percentage of the initial sample weight (Honikel, 1998).

For the shear force test, three to four 1.27 cm diameter samples were cut randomly from each cooked sample (after determining cooking loss). Care was taken to ensure that no visible connective tissue was included in the cut section. The samples were cut perpendicular to the longitudinal axis of the muscle fibres so that the influence of the myofibrillar proteins on the shear force could be measured (Voisey, 1976). An average maximum shear force was calculated based on the shear force (N/1.27 cm diameter) required to shear the 1.27 cm diameter cylindrical core of cooked meat perpendicular to the grain, at a crosshead speed of 200 mm/min. The shear force measurements (WBSF) were generated with a Warner-Bratzler shear attachment, fitted to an Instron Universal Testing machine. A higher value indicated greater shear force and therefore, tougher meat (Honikel, 1998).

For the colour measurement of the meat, freshly cut steaks were allowed to bloom for 30 mins where after the colour was measured in triplicate, at random positions on the steak surface

(Stevenson *et al.,* 1989) using a Color-guide 45°/0° colorimeter (Cat no: 6805; BYK-Gardner, USA). The colour was expressed by the coordinates L*, a* and b* of the CIELab colour space (Commission Internationale de l'Eclairage, 1976). The L* represents lightness in meat colour, a* value indicates red-green range and b* value the yellow-blue range (Poulson *et al.,* 2004). These values were also used to calculate the hue angle (h_{ab}) and chroma value (C*) using the following equations (Commission Internationale de l'Eclairage, 1976):

Hue angle: $h_{ab} = \tan^{-1} (b^*/a^*)$

Chroma value $C^* = [(a^*)^2 + (b^*)^2]^{1/2}$

Statistical analysis

The effect of incremental inclusion of maize cobs in feedlot finishing diets of SAMM lambs on meat quality characteristics were analysed by using PROC ANOVA and PROC GLM of SAS (2000). Least Square Means were calculated for all effects. Significance was declared at $P \leq 0.05$.

Results and discussion

From a practical point of view, the protein, fat and energy content, based on dry weight basis, is of little use since meat is consumed by weight or servings rather than by amount of dry tissue. For this reason protein, fat and energy are expressed on a wet weight basis, which is important in dietary planning and most informative for consumers (Abu-Tarboush & Dawood, 1993).

The proximate chemical compositions of the *M. longissimus dorsi* and *M. biceps femoris* are presented in Table 2. The three different diets had no significant influence on the proximate composition of the *M. longissimus dorsi* and *M. biceps femoris* of the lambs. The values for protein, fat and consequently energy analysed from the *M. longissimus dorsi* in the current study were higher than that reported by Sheridan (2001) for the *M. semimembranosus* (19.52%, 3.19% and 449.78 kJ, respectively) from SAMM lambs finished on a feedlot diet. Cloete *et al.* (2004), on the other hand, reported proximate chemical values for SAMM sheep that were similar to those found in this study.

A possible explanation for the higher fat content observed in this investigation, compared to that reported by Sheridan (2001), may be due to a difference in final age and final body weight between the two studies, and the fact that different muscles are being compared

Table 2 Mean proximate composition (on an *as is* basis) of *M. longissimus dorsi* and *M. biceps femoris* of the SAMM lambs fed the three different diets

	Maize cobs inclusion level			SEM
	4%	8%	12%	
	Diet 1	Diet 2	Diet 3	
M. longissimus dorsi				
Moisture (%)	74.53	74.01	74.04	0.184
Ash (%)	1.15	1.18	1.16	0.009
Protein (%)	20.14	20.16	20.37	0.116
Fat (%)	4.04	4.20	4.09	0.082
Energy (kJ)[1]	491.86	498.15	497.56	4.248
M. biceps femoris				
Moisture (%)	75.29	75.00	74.77	0.156
Ash (%)	1.20	1.20	1.21	0.009
Protein (%)	19.67	19.58	19.87	0.189
Fat (%)	3.65	3.74	3.74	0.097
Energy (kJ)[1]	469.58	471.22	476.31	5.000

[1] Total energy (kJ) = (g protein in 100g sample × 17) + (g fat in 100g sample × 37) (As gazetted: SA Act No 54 of 1972).

(Martínez-Cerezo *et al.*, 2005). The lambs in the current study were approximately 7 months of age at the end of the trial and weighed on average 44.06 kg, compared to the lambs, weighing on average 42.20 kg at an age of approximately 5 months, used by Sheridan (2001). Marbling fat (intramuscular fat) starts developing only after kidney, intermuscular and subcutaneous fat has been formed, and due to the difference in age, the intramuscular fat of the animals in this study may have developed to a greater extent (Demeyer & Doreau, 1999). Although not significant, the fat content of the *M. longissimus dorsi* in Diet 2 tended (P = 0.7262) to be higher (and consequently a higher calculated energy value) than that of the animals in the other two treatments. This might be due to a little higher gross energy (GE) and calculated metabolizable energy (ME) content of Diet 2 (Table 1), which is an indication that this diet had excess energy for growth and maintenance, which is absorbed and deposited as fat. The fat thickness measured at the 4^{th}–5^{th} lumbar vertebrae tended to be higher (P = 0.8884) for lambs fed Diet 2, which supports the above-mentioned.

Although not significant, there was a tendency that the fat content of both muscles from the lambs fed the three diets had a reverse relationship to the moisture content (r = -0.58, P = 0.1319; r = -0.51, P = 0.1919; r = -0.56, P = 0.1456, respectively for Diet 1, 2 and 3) in the *M.*

biceps femoris. The *M. longissimus dorsi* exhibited significant inverse relationships between fat and moisture contents, except for Diet 2 ($r = -0.75$, $P = 0.0338$; $r = -0.63$, $P = 0.0932$; $r = -0.92$, $P = 0.0011$, respectively for Diet 1, 2 and 3). Muscles with the highest fat content were characterised by lower moisture content (Table 2). This is in accordance with the findings of Ono *et al.* (1984), Dransfield *et al.* (1990) and Martínez-Cerezo *et al.* (2005) who reported the same tendency for cross-bred lambs, Suffolk ram lambs and Spanish sheep breeds, respectively, raised on a feedlot diet.

The diets did not influence ($P > 0.05$) the protein content of the meat, probably because the diets were isonitrogenous (Sheridan *et al.*, 2003) and the fat content of the two muscles investigated, also did not differ significantly between diets.

The mean values for the different dietary effects on the physical attributes investigated are presented in Table 3. No significant differences were observed in the initial, as well as the ultimate pH and temperature readings taken 45 min and 48 h, respectively, post-mortem. The lambs on Diet 1 tended to have a slightly higher mean ultimate pH ($P = 0.0656$) and temperature ($P = 0.7131$) compared to the lambs on the other two diets. It is a well-known fact that the ultimate pH of the muscle is an important contributing factor to the quality of meat (Hoffman *et al.*, 2003). According to Devine *et al.* (1993) an ultimate pH value greater than 5.8 is regarded as undesirable. They also stated that in rested, fed sheep the ultimate pH in the *M. longissimus lumborum* is less than 5.6 and that stressful handling causes a high ultimate pH (Martínez-Cerezo *et al.*, 2005). Lawrie (1985) found that the bacteriological stability of chilled meat is adversely affected by an ultimate pH value above 6.0. The mean pH values of the three diets were all very close or within the normal mutton pH range of 5.4–5.86 (Safari *et al.*, 2001).

Conflicting reports regarding the relationship between pH and tenderness (WBSF) are found in the literature. Consumer surveys have shown that tenderness is considered the most important component of meat quality (Young *et al.*, 1993) and that a WBSF value greater than 49 N is regarded as tough by consumers (Safari *et al.*, 2001). Devine *et al.* (1993) reported an increase in shear force of lamb with higher pH values in the range of 5.4–6.0, while research by Silva *et al.* (1999) oppose these findings. They observed that tenderness of beef assessed by WBSF increased linearly with ultimate pH. Purchas (1990) and Jeremiah *et al.* (1991) found a curvilinear relationship between pH and tenderness, with a minimum tenderness between pH 5.8 and 6.2. Safari *et al.* (2001), on the contrary, found no relationship between pH and shear force value and cooking loss in six diverse lamb genotypes.

Table 3 Physical characteristics of the *M. longissimus dorsi* and subcutaneous fat for the SAMM lambs fed the three different diets

	Maize cobs inclusion level			SEM
	4%	8%	12%	
	Diet 1	Diet 2	Diet 3	
M. longissimus dorsi				
pH_{45}	6.78	6.96	6.72	0.060
$Temperature_{45}$ (°C)	32.14	33.39	32.43	0.700
pH_{48}	5.60	5.41	5.38	0.042
$Temperature_{48}$ (°C)	3.23	2.96	3.14	0.128
Drip loss (%)	1.61	1.58	1.51	0.029
Cooking loss (%)	29.90^{b}	32.89^{a}	30.69^{a}	0.477
Shear force (N/1.27 cm diameter)	35.60	34.65	36.12	0.791
L* value	36.28^{b}	39.40^{a}	38.54^{a}	0.305
a* value	10.95	10.39	10.68	0.119
b* value	7.48^{b}	9.22^{a}	8.59^{a}	0.174
Hue angle (°)	34.11^{b}	41.67^{a}	38.68^{a}	0.729
Chroma	13.35	13.91	13.77	0.124
Fat thickness (2^{nd} – 3^{rd} last thoracic vertebrae)	5.13	4.66	4.75	0.255
Fat thickness (4^{th} – 5^{th} lumbar vertebrae)	2.42	2.54	2.30	0.194
Subcutaneous fat				
L* value	70.54	70.05	71.40	0.759
a* value	0.97	1.80	1.67	0.183
b* value	11.47	11.37	11.38	0.194
Hue angle (°)	85.13	80.78	81.61	0.933
Chroma	11.56	11.55	11.73	0.185

[a,b,c] Means in the same row with different superscripts differ ($P \leq 0.05$).

Velasco *et al.* (2004) concluded that type of supplementary feed does not affect the pH at any time measured post-mortem, nor did it influence the drop in pH values throughout carcass refrigeration. They suggested that animals at pasture, regardless of the energy supplement received, are equally susceptible to stress and displayed the same glycogen muscle content.

There is also a great deal of controversy amongst researchers regarding the relationship between tenderness and the intramuscular fat content of the meat. Silva *et al.* (1999), for example, stated that intramuscular fat content is not correlated with tenderness. This is inconsistent with the negative correlation ($P < 0.001$) obtained by Seideman *et al.* (1987) and Okeudo & Moss (2005) between marbling fat and shear force of the *M. longissimus dorsi* in

cattle and sheep, respectively. Schönfeldt *et al.* (1993) describe a study in which a positive relationship was found between tenderness of meat and fatness of ovine carcasses.

Our results also indicate that the muscle temperature measured after 48 h post-mortem tended to be higher ($P = 0.7131$) in Diet 1, which might be explained by a thicker, although not significant, back-fat thickness measured at the 2^{nd}–3^{rd} last thoracic vertebrae. This finding coincides with the results obtained by Okeudo & Moss (2005), who showed that rate of chilling of sheep carcasses were negatively affected by back fat cover and carcass weight. Large carcasses have a proportionately smaller surface area and the fat cover acts as an insulatorary barrier to heat loss. Moreover, intramuscular fat content also lowers the aggregate thermal conductivity of the muscle (Hill *et al.,* 1967).

Although the three diets had no effect ($P > 0.05$) on the percentage drip loss, it significantly influenced the cooking loss of the *M. longissimus dorsi*. According to the work done by Hopkins & Fogarty (1998), a lower cooking loss is associated with a higher fat content in the meat. Inconsistent with these findings, Schönfeldt *et al.* (1993) and Carpenter & King (1965) reported significantly higher total cooking loss when ovine carcasses had increased fat content. In the current study the values obtained for cooking loss of lambs fed Diet 2 were higher ($P \leq 0.05$) than that of lambs fed Diet 1. This might be due to the intramuscular fat content that tended to be higher ($P = 0.7262$) (Schönfeldt *et al.,* 1993, Carpenter & King, 1965) for animals fed Diet 2 than Diet 1. Purchas *et al.* (1969) showed that although total cooking loss was positively correlated to fat percentage of the carcass side when the whole loin was cooked, cooking loss from the muscular tissue was poorly or negatively correlated to fat percentage of the carcass side.

It is evident from the work done by Safari *et al.* (2001) that a negative correlation exists between cooking loss and juiciness, which will in turn affect the eating quality and acceptability of the meat. Cooking loss and juiciness are significantly ($P < 0.001$) correlated with tenderness assessed by both WBSF and a sensory panel (Silva *et al.,* 1999; Okeudo & Moss, 2005). A reduction in cooking loss will lead to an increase in juiciness, and thus an increase in the tenderness of the meat. Bouton *et al.* (1973) considered that the higher water holding capacity of meat of higher ultimate pH contribute to their high tenderness. Okeudo & Moss (2005) suggest a positive relationship between moisture content of meat and cooking loss and that it is a more important determinant of cooking loss than intramuscular lipid content.

All the lambs displayed L* values above 34. According to Hopkins (1996, as cited by Velasco *et al.,* 2004) this indicates that meat is light-coloured and acceptable to consumers. Meat of animals raised at pasture is generally darker than that of feedlot animals fed concentrate, as lambs at pasture display a greater concentration of blood pigments in muscle than feedlot animals due to their grass intake (Renerre, 1986, as cited by Velasco *et al.,* 2004). The colour of the *M. longissimus dorsi* was significantly darker (lower L*) in the lambs fed Diet 1 than the lambs fed Diet 2 and 3. There was no significant difference in the index of redness among the three treatments, but the yellowness index (b*) was lower ($P \leq 0.05$) and the hue angle narrower ($P \leq 0.05$) in the lambs fed Diet 1.

The difference in meat lightness could have been partially caused by the slight difference in the ultimate pH since high pH meats tend to have a darker colour (Lawrie, 1985; Velasco *et al.*, 2004). This is in accordance with the findings of Young *et al.* (1993) and Priolo *et al.* (2002) who also reported a darker meat colour due to a slightly higher ultimate pH.

The subcutaneous fat colour (Table 3) was not significantly influenced by the diet. According to Mancini & Hunt (2005) fat colour has received less attention in the literature; however, it has been proved that the diet can affect the carotenoid content of the fat, which can influence the fat colour. A lower amount of β-carotene in concentrates than in pasture, results in less carotene accumulation in fat of animals fed concentrates, therefore subcutaneous fat colour from grass fed lambs is more yellow (b*) than that from lambs fed a concentrate (Priolo *et al.,* 2002). Darker subcutaneous fat colour resulted from the redox state of the residual haemoglobin (Mancini *et al.,* 2005), being either deoxyhaemoglobin or methaemoglobin, in the blood capillaries (Irie, 2001).

Conclusion

The objective of this investigation was to determine whether maize cobs, when included at different rates in feedlot finishing diets of SAMM lambs, has a significant effect on lamb quality. Based on the results obtained from this study it may be concluded that the inclusion of maize cobs, at three different rates, had no effect ($P > 0.05$) on the proximate chemical composition of the two muscles, *M. longissimus dorsi* and *M. biceps femoris,* investigated. However, lambs fed the diet containing 4% maize cobs (Diet 1), produce meat with a significantly lower L* value, b* value and consequently a narrower hue angle, than that of lambs fed the other two diets. Results also indicated a significant difference in percentage cooking loss between treatments, with lambs fed Diet 1 having the lowest cooking loss.

However, whether these differences are of such a magnitude that they will influence either purchasing behaviour or consumption satisfaction of consumers requires elucidation.

Based on the results obtained in the present trial, it may be concluded that the inclusion of maize cobs up to 12% in a finishing diet of lambs did not greatly affect the meat quality of the animals adversely.

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Chapter 5

Effects of rumen-protected conjugated linoleic acid (CLA) in feedlot finishing diets on production efficiency, proximate chemical composition and physical meat quality characteristics of South African Mutton Merino lambs

Abstract

Sixteen South African Mutton Merino (SAMM) lambs were used in a feedlot finishing study to evaluate the effect of feeding a diet containing supplemental rumen-protected conjugated linoleic acid (CLA). The two diets were formulated on an isonitrogenous and isoenergetic basis (15.50% crude protein (CP) and a metabolizable energy content of 10.64 MJ/kg on a dry matter (DM) basis). The lambs were fed *ad libitum* for 24 days from average 34.8 ± 2.60 kg to 43.4 ± 2.71 kg body weight, where after they were slaughtered. Individual feed consumption, utilisation, growth and digestion efficiency were investigated. The *longissimus dorsi* and the *biceps femoris* muscles from the right hand side of each carcass were removed for determination of physical and chemical characteristics. No significant differences occurred in the growth performance and feed conversion efficiency of the lambs consuming the two different diets. Both energy and nitrogen retention of the lambs were not affected by diet. Diet also did not have a significant effect on the proximate chemical composition of both muscles. However, diet had a significant effect on the cooking loss and the colour measurements of the *longissimus dorsi* muscle. It can be concluded that 2.5% supplemental rumen-protected CLA in lamb feedlot diets has no adverse affect on their production performance and meat quality.

Key words: conjugated linoleic acid; production efficiency; physico-chemical attributes; South African Mutton Merino lambs; feedlot

Introduction

The concept of certain foods or food components having beneficial effects that go beyond their traditional nutritive value is gaining awareness and acceptance among consumers. Food products have been shown to contain micro-components that have positive effects on human health and disease prevention in addition to those related to its traditional nutritive value. In

recent years, researchers have been exploring the possibility of enhancing the beneficial effects of animal products (milk and meat) through diet manipulation. Food products (milk and meat) from ruminant animals (Dhiman *et al.*, 2000; Mir *et al.*, 2004) are the primary source of conjugated linoleic acid (CLA) for humans. One such option of diet manipulation may be the use of finishing diets supplemented with CLA or with seeds with a high linoleic acid content (Kelly *et al.*, 1998) to improve the concentration of CLA and thus the health benefits of red meat.

The acronym CLA is a collective term used to describe a mixture of positional and geometric isomers of linoleic acid (C18:2 *cis*-9, *cis*-12). CLA has the same chain length as linoleic acid (C18:2n-6), but in CLA the double bonds are conjugated. The bonds in CLA can be either in a *trans* (*t*) or *cis* (*c*) configuration. CLA isomers are intermediates in the biohydrogenation of linoleic acid to *trans*-vaccenic acid and stearic acid (Kelly *et al.*, 1998; Mir *et al.*, 2000). CLA have been associated with a wide range of positive health benefits in humans. The C18:2 *cis*-9, *trans*-11 isomer, sometimes called rumenic acid, is the predominant isomer of CLA and has been shown to have anticarcinogenic properties in animal models (Ha *et al.*, 1987; Ip *et al.*, 1994). The C18:2 *trans*-10, *cis*-12 isomer has been reported to increase growth efficiency (Wiegand *et al.*, 2001) and reduce body fat while increasing lean body mass in growing animals (Park *et al.*, 1999; Azain *et al.*, 2000). Anti-diabetic effects of CLA isomers have also been reported (Houseknecht *et al.*, 1998). For these reasons, increasing the CLA content of foods may increase their nutritive and therapeutic values.

The South African Mutton Merino (SAMM) is a dual-purpose (mutton and wool) sheep breed and was selected for the study due to its high growth rate and capability to produce a slaughter lamb at an early age with good meat quality attributes (Cloete *et al.*, 2004). It has a high fertility, good conformation, good feed conversion ratio and adaptability and is therefore popular in feedlot production systems (Neser *et al.*, 2000).

There is little information on the effect of CLA on production efficiency of lambs and nutritional quality of mutton. This study was therefore conducted to determine the effect of rumen-protected CLA in a feedlot finishing diet on the feed intake, growth parameters, digestion, carcass characteristics, as well as meat quality characteristics of SAMM lambs.

Materials and methods

Animals and feeding trial

Sixteen South African Mutton Merino (SAMM) intact male lambs were used in this investigation. All the lambs were weaned and shorn prior to entering the feedlot at an age of approximately 6 months. All the animals were dewormed with a broad-spectrum drench to eliminate all internal parasites on their arrival at the University of Stellenbosch experimental farm, Welgevallen. The average initial body weight (BW) was 34.8 ± 2.60 kg. The lambs were randomly allocated into individual pens (1 m x 2 m) in an enclosed but adequately ventilated shed with a wooden slatted floor. Lambs were divided in two groups of equal initial weights. Both groups were randomly assigned to one of two treatments: the control and a second diet supplemented with a rumen-protected conjugated linoleic acid (CLA) (Diet 1 and 2, respectively; Table 1). The rumen-protected CLA source (Luta-CLA® 20 P, BASF Animal Nutrition, P.O. Box 1783, 1620, Kempton Park, South Africa) contained 20% CLA, and consisted of two isomers present in the same concentration (10%): C18:2 *cis*-9, *trans*-11 and C18:2 *trans*-10, *cis*-12. The pelleted diets were formulated according to the NRC (1985) recommendations for lambs, on an isonitrogenous and isoenergetic basis. The lambs had *ad libitum* access to the feed and fresh water. They were fed twice daily, for 24 days, at 08:00 and 16:00, and weighed weekly to determine the average daily gain (ADG) for each lamb. Feed refusals were collected daily in the morning before the provision of new feed and water, pooled and weighed weekly to calculate voluntary feed intake, as well as feed conversion efficiency (FCE) (kg feed/kg BW gain) for each lamb.

Digestibility trial

At the end of the feeding trial, twelve of the lambs were used to determine the nutrient digestibility and nitrogen (N) as well as energy retention of the two diets. For the duration of the digestibility trial, the other four lambs were maintained on the diet. Faeces were collected twice daily, for 7 days, at 10:00 and 16:00 whilst the urine was collected with the morning feeding. Twenty mℓ of urine preservative (80 g potassium dichromate and 20 g mercuric chloride dissolved in 1 ℓ distilled water) were added each morning to the urine collection jugs to prevent volatilization of ammonia from the urine. Daily sub-samples of the faeces (10%) and urine (5%) from each lamb were pooled over the whole period, prior to chemical analysis.

Table 1 Physical and chemical composition of the two diets fed to the SAMM lambs

	Diets	
	Diet 1 (Control)	Diet 2 (CLA)
Physical composition[1]		
Maize meal	23.00	14.80
Lucerne hay	17.00	25.24
Citrus pulp	4.00	2.50
Maize cobs	8.00	7.50
Hominy Chop	25.44	25.00
Sunflower hulls	0.00	5.00
Wheat bran	9.10	2.50
Linseed meal	0.00	3.00
Full-fat soybeans	2.00	0.00
Ammonium chloride	0.50	0.50
Limestone	1.40	1.70
Mono calcium phosphate	0.00	0.10
Salt	0.60	0.60
Urea	0.70	0.80
CLA[3]	0.00	2.50
Molasses	7.50	7.50
Premix B[4]	0.25	0.25
Acid Buf[5]	0.50	0.50
Avatec[6]	0.02	0.02
Chemical composition[2]		
Organic matter (%)	93.05	92.56
Ether extract (%)	4.29	5.62
Crude Protein (%)	15.70	14.77
Acid detergent fibre (%)	16.21	20.28
Neutral detergent fibre (%)	30.66	35.21
Gross energy (MJ/kg)	18.29	18.63
Metabolizable energy (MJ/kg)[7]	10.66	10.62

[1]On an air dry basis; [2]Analysed values on a DM basis; [3]Rumen-protected conjugated linoleic acid; supplied by BASF Animal Nutrition (P.O. Box 1783, 1620, Kempton Park, South Africa); [4]A standard mineral (macro and micro) and vitamin supplement; formulated and supplied by BASF Animal Nutrition; [5]A rumen buffer; supplied by Nutec Southern Africa (234 Royston Road, Willowton, Pietermaritzburg, KwaZulu-Natal, South Africa); [6]A growth promoter, lasalocid sodium; supplied by Instavet Import and Export Pty (Ltd) (P.O. Box 346, 2163, Kya Sand, South Africa); [7]Based on the nutritive value of feeds according to laboratory determined values by Senwesko Feeds.

Methane gas production (MJ/day) was calculated as 8% of the gross energy intake (McDonald *et al.*, 1995). Nitrogen retention was corrected both for endogenous urinary N (EUN) and metabolic faecal N (MFN) according to McDonald *et al.* (1988) as follows:

EUN (g) = 0.18 g N/kg $BW^{0.75}$/day

MFN (g) = 5 g N/kg dry matter intake

N retention (g N/kg $BW^{0.75}$/day) = $\{N_{intake} - (N_{faeces} - MFN) - (N_{urine} - EUN)\}/BW^{0.75}$/days

At the end of the 7-day digestibility trial, the sixteen SAMM lambs were slaughtered at a commercial abattoir using standard South African procedures. The fasted (16 hrs) average live weight (BW) of the lambs were 42.1 ± 4.34 kg as determined prior to slaughter. After being electrically stunned (4 seconds at 200 volts) the lambs were exsanguinated and the carcasses suspended by the Achilles tendon to bleed. After evisceration, the weight of the warm carcass of each lamb was recorded. The carcasses were then suspended in a cooler at 2°C and weighed after 48 h to determine the moisture loss in the cooler. Dressing percentage was calculated from the cold carcass weight as a percentage of live BW. No electrical stimulation of the carcasses was applied.

Sampling

Forty-five minutes post-mortem the initial muscle pH (pH_{45}) and temperature ($Temp_{45}$) and after 48 h, the ultimate pH (pH_{48}) and temperature ($Temp_{48}$) were measured with a penetrating glass electrode on a hand-held Crison pH/mV-507 meter. The measurement was taken from within the carcass between the 2nd and 3rd last thoracic vertebrae, 45 mm from the midline. The pH meter was re-calibrated after every fourth reading and the electrode rinsed with distilled water between each measurement. The pH meter contained a temperature probe ensuring automatic adjustment of the pH for temperature. The *M. longissimus dorsi* was removed from the left and right hand side of the carcass (the muscle was removed between the 2nd–3rd last thoracic vertebrae and the 4th–5th lumbar vertebrae) and the *M. biceps femoris* was dissected from the right hind leg to assess measurements of drip loss, cooking loss, meat colour and Warner-Bratzler shear force (WBSF) values. The subcutaneous fat from the removed *M. longissimus dorsi* was used for fat colour measurements. Back-fat thickness was measured with a digital calliper at a site 25 mm off the midline at the 2nd–3rd last thoracic vertebrae and the 4th–5th lumbar vertebrae (where the *M. longissimus dorsi* was removed).

Chemical analysis of the feed and faeces

The faeces were dried at 60°C for 96 h, air-equilibrated and weighed. The feed and faeces were ground through an 1 mm screen and analysed for dry matter (DM), organic matter

(OM), crude protein (CP), ether extract (EE), neutral detergent fibre (NDF), acid detergent fibre (ADF) and gross energy (GE) content (AOAC, 2002). The urine samples were analysed for N and gross energy content (AOAC, 2002). All nitrogen analyses were performed according to the combustion method (Method 990.03, AOAC, 2002) with a Leco FP-428 Nitrogen and Protein Analyzer (Leco Corporation, 3000 Lakeview Avenue, St. Joseph, MI 49085-2396). NDF and ADF were determined with an ANKOM^{220} Fiber Analyzer (ANKOM Technologies, Fairport, NY, USA) and GE by adiabatic oxygen bomb calorimetry (CP500 calorimeter).

Proximate chemical analysis

Proximate chemical analysis was carried out on the *M. longissimus dorsi* and the *M. biceps femoris* from the right hand side of the carcass. The *M. longissimus dorsi* from the left hand side of the carcass was used for sensory analyses (Chapter 6). The frozen muscle samples were cut into smaller portions, minced three times through a 2 mm sieve to ensure homogeneity, and analysed chemically. Proximate composition was determined according to AOAC methods (AOAC, 2002). The analysis included determination of moisture, ash, protein (N × 6.25) and fat content. The nitrogen analysis were performed on dried, fat free samples, according to the combustion method (Method 990.03, AOAC, 2002) with a Leco FP-428 Nitrogen and Protein Analyzer (Leco Corporation, 3000 Lakeview Avenue, St. Joseph, MI 49085-2396). The moisture content was determined by drying at 100°C for 24 h and ashing was done at 500°C for 5 h. The lipid content was determined by means of chloroform:methanol (1:2 v/v) extraction according to the method of Lee *et al.* (1996).

Physical analysis

All the physical measurements were conducted on the *M. longissimus dorsi* after the two muscles (*M. longissimus dorsi* and *M. biceps femoris*) had been removed from the carcasses and transported to the laboratory.

For the drip loss determination 1.5 cm thick meat samples, cut perpendicular to the longitudinal axis of the muscle on the caudal side of the removed *M. longissimus dorsi*, were weighed. The samples were placed in netting and then suspended in an inflated plastic bag, ensuring that the sample did not come into contact with the exudate. After a storage period of 24 h at 4°C, the samples were blotted dry with absorbent paper, weighed again and the drip loss was calculated as weight loss expressed as a percentage of the original weight of the sample (Honikel, 1998).

Cooking loss of the *M. longissimus dorsi* was determined by placing weighed, freshly cut samples (1.5 cm thick) in thin-walled plastic bags in a temperature-controlled water bath (preset at 80°C) for 1 h. The samples were then removed from the water bath, cooled in cold water, blotted dry and weighed. Cooking loss was calculated as the difference in sample weight before and after cooking, expressed as a percentage of the initial sample weight (Honikel, 1998).

For the shear force test, three to four 1.27 cm diameter samples were cut randomly from each cooked sample (after determining cooking loss). Care was taken to ensure that no visible connective tissue was included in the cut section. The samples were cut perpendicular to the longitudinal axis of the muscle fibres so that the influence of the myofibrillar proteins on the shear force could be measured (Voisey, 1976). An average maximum shear force was calculated based on the shear force (N/1.27 cm diameter) required to shear the 1.27 cm diameter cylindrical core of cooked meat perpendicular to the grain, at a crosshead speed of 200 mm/min. The shear force measurements (WBSF) were generated with a Warner-Bratzler shear attachment, fitted to an Instron Universal Testing machine. A higher value indicated greater shear force and therefore, tougher meat (Honikel, 1998).

For the colour measurement of the meat, freshly cut steaks were allowed to bloom for 30 mins where after the colour was measured in triplicate, at random positions on the steak surface (Stevenson *et al.,* 1989) using a Color-guide 45°/0° colorimeter (Cat no: 6805; BYK-Gardner, USA). The colour was expressed by the coordinates L*, a* and b* of the CIELab colour space (Commission Internationale de l'Eclairage, 1976). The L* represents lightness in meat colour, a* value indicates red-green range and b* value the yellow-blue range (Poulson *et al.,* 2004). These values were also used to calculate the hue angle (h_{ab}) and chroma value (C*) using the following equations (Commission Internationale de l'Eclairage, 1976):

Hue angle: $h_{ab} = \tan^{-1} (b^*/a^*)$

Chroma value $C^* = [(a^*)^2 + (b^*)^2]^{1/2}$

Statistical analysis

The effect of supplemental dietary rumen-protected conjugated linoleic acid (CLA) in feedlot finishing diets of SAMM lambs, on their production efficiency and meat quality characteristics were analysed by using PROC ANOVA and PROC GLM of SAS (2000). Least Square Means were also calculated. Significance was declared at $P \leq 0.05$.

Results and discussion

The growth parameters of the SAMM lambs are presented in Table 2. Feed intake, ADG and FCE were not affected ($P > 0.05$) by dietary treatment. In the current study there was no significant reduction in DM intake when supplementing a diet (Diet 2) of finishing SAMM lambs with 2.5% rumen-protected CLA. This contradicts a study by Nigdi *et al.* (1990) who added calcium soaps of long-chain fatty acids to finishing diets of steers and observed a reduction in DM intake with added fat. Similar results were also reported by Haddad & Younis (2004) where DM intake decreased when a rumen protected fat (Ultralac^{TM} 100) was added at a level of 2.5 and 5% to fattening Awassi lamb diets formulated on an isonitrogenous basis. Boggs *et al.* (1987) attributed a portion of the decrease in DM consumption to the increase in caloric density due to chemostatic regulation of voluntary intake.

Similar to our results, Gillis *et al.* (2004) found that the inclusion of 2% rumen-protected CLA salt (calcium salts of palm oil fatty acids and CLA) to a basal diet had no negative effect on DM intake of Angus × Hereford heifers. In contrast, Gassman *et al.* (2000) reported a reduction in DM intake when 2.5% rumen-protected CLA was supplemented to feedlot cattle diets.

Sheridan *et al.* (2003a) observed results similar to that recorded for this investigation as pertaining to ADG and FCE (0.330 kg/day and 5.56 kg feed/kg BW gain, respectively) for SAMM lambs receiving a similar diet at an equivalent initial live weight. Brand *et al.* (2001), on the other hand, found a lower ADG, but a more efficient FCE (0.267 kg/day and 4.80 kg feed/kg BW gain, respectively) for SAMM lambs in a feedlot receiving a diet (15.60% CP) that had been supplemented with 6% canola. Inconsistent with the findings in the current study, Gillis *et al.* (2004) indicated that the ADG was increased ($P \leq 0.05$) and the FCE improved ($P \leq 0.05$) when supplementing the basal diets of Angus × Hereford heifers with 2% rumen-protected CLA. Gassman *et al.* (2000) observed no differences in FCE, but a reduction of 25% in ADG between a control and a rumen-protected CLA supplemented feedlot diet for cattle. Although ADG and FCE did not vary ($P > 0.05$), the FCE of the lambs on Diet 1 was more efficient than for those on Diet 2. The fact that no significant differences occurred in any of the growth parameters may be ascribed to the isonitrogenous and isoenergetic state of the two experimental diets (Table 1).

In terms of cold carcass weight and dressing percentage, no significant differences were observed for the lambs on the two different diets. The values for cold carcass weight and

dressing percentage in the present study are, however, higher than that (19.42 ± 0.777 kg and 47.02 ± 0.647%, respectively) reported by Sheridan *et al.* (2003a) for SAMM lambs weighing 42.20 ± 1.013 kg at slaughter. The dressing percentage (41%) given by Brand *et al.* (2001) for SAMM lambs was also lower than the dressing percentage found in the current study. The differences that occurred in dressing percentage between these two authors and the current study may be due to a difference in the level of gut fill at the time of recording the final body weights (Kirton *et al.*, 1995). Dressing percentage also depends on the forage content of the diet, and therefore on the weight of the digestive tract (Brosh *et al.*, 1995). Skin weight, especially in wool or dual purpose breeds, may also affect dressing percentage when the live weights are recorded (Cloete *et al.*, 2004). If the lambs were shorn prior to entering the feedlot or slaughtering, this will lead to a lower skin weight and thus a higher dressing percentage.

Table 2 Growth parameters, average feed intake and conversion efficiency of South African Mutton Merino lambs receiving the two different feedlot diets (LSMean ± SE)

	Diet 1 (Control)	Diet 2 (CLA)
Initial body weight (kg)	35.00 ± 1.090	34.50 ± 0.779
Final body weight (kg)	44.06 ± 1.120	42.75 ± 0.786
Body weight gain (kg)	9.06 ± 0.427	8.25 ± 0.271
Cumulative feed intake (kg)	47.95 ± 1.481	45.96 ± 1.196
Daily feed intake (kg)	2.00 ± 0.062	1.91 ± 0.050
ADG (kg/day)	0.378 ± 0.018	0.344 ± 0.011
FCE (kg feed/kg BW gain)	5.38 ± 0.299	5.62 ± 0.257
Body weight at slaughter (kg)	42.31 ± 1.839	41.88 ± 1.284
Warm carcass weight (kg)	22.31 ± 0.755	22.36 ± 0.669
Cold carcass weight (kg)	21.89 ± 0.751	21.91 ± 0.654
Moisture loss (%)	1.91 ± 0.198	1.98 ± 0.142
Dressing percentage (%)	51.91 ± 0.774	52.38 ± 0.768

Table 3 shows the apparent digestibility of the different components of the two diets. DM and OM digestibility differed ($P \leq 0.05$) between the two diets. Diet 1 was digested to a greater extent than Diet 2. In contrast, Haddad & Younis (2004) reported an increase in DM digestibility with the addition of a rumen protected fat to fattening Awassi lamb diets. Kronfeld & Donoghue (1980) indicated that a reduction in fibre digestibility with protected tallow diets might have been due to liberation of fat in the rumen, which affected the fermentative digestion of some components of the fibre. Another possible reason for the lower digestibility of DM and OM in Diet 2 may be due to the higher dry matter (g/day) and

energy intake (MJ/day) during the digestibility trial period. Higher feed intakes usually lead to increased passage rates and reduced rumen retention times, resulting in decreased digestibility (Ørskov & McDonald, 1979).

Although there was no difference ($P > 0.05$) in ADF digestibility, the treatments significantly affected the NDF digestibility of the two diets. The two diets also did not differ ($P > 0.05$) in crude fat digestibility. The mean ADF and NDF digestibility values (36.79 and 44.93% respectively) obtained in this study were lower than the corresponding values (52.66 and 62.46% respectively) observed by Sheridan *et al.* (2003a) for SAMM lambs. Nangole *et al.* (1983) reported an ADF digestibility value of 53.0%, which is also higher than that obtained in the current study, for a diet consisting of 87.5% maize cobs fed to Romney Marsh wethers. The low ADF and NDF digestibility observed in the current study might be attributed to low ruminal pH levels (Haddad & Husein, 2004). Haddad *et al.* (1995) stated that a ruminal pH below 6.2 increases the lag time and decreases the extent of digestion for wheat straw fibre. When grain are introduced into the diet of ruminants fed predominately forage, the pH will decline, the number of amylolytic bacteria tend to increase and the number of fibrolytic bacteria tend to decrease (Henning *et al.*, 1980). Thus, production of fibrolytic enzyme systems by the microbial population and the rate of fibre digestion are likely to decrease (Chakeredza *et al.*, 2001).

Table 3 Apparent digestibility (mean) of the diets fed to the South African Mutton Merino lambs

Chemical component (%)	Diet 1 (Control)	Diet 2 (CLA)	SEM
Dry Matter	74.00[a]	68.60[b]	0.880
Organic Matter	75.40[a]	70.04[b]	0.881
Acid detergent fibre	39.28	34.31	1.703
Neutral detergent fibre	48.53[a]	41.32[b]	1.499
Ether extract	91.82	94.10	0.611

[a,b,c] Means in the same row with different superscripts differ ($P \leq 0.05$).

Energy intake (MJ/day), excretion (MJ/day) and retention (MJ/day) and metabolizable energy content (ME, MJ/kg) of the two diets are presented in Table 4. No statistical differences ($P > 0.05$) were observed in DM and energy intake and energy excreted as faecal energy, urinary energy and methane gas, for the lambs on the two different diets. Although there were no significant differences between the energy intake and total energy excreted between the two diets, the lambs on Diet 2 tended to have the highest energy intake ($P = 0.3668$) and total energy excreted ($P = 0.1100$) (25.80 MJ/day and 10.96 MJ/day, respectively). This

numerically higher total energy excreted for Diet 2 might be due to their higher daily dry matter and energy intake. A consequence of this higher daily DM intake was that more easily fermentable carbohydrates were available for fermentation in the rumens of the lambs in this treatment, resulting in a higher methane gas production (Nolte & Ferreira, 2004). Differences ($P \leq 0.05$) occurred in faecal energy and total energy excreted, expressed as a percentage of energy intake. Urinary energy excretion as a percentage of energy intake did not differ ($P > 0.05$) between the diets, suggesting that the absorbed nutrients were metabolized with similar efficiency for all the lambs (Nolte & Ferreira, 2004).

The efficiency of energy retention was similar ($P = 0.6523$) for the lambs on Diets 1 and 2, but when expressed as a percentage of energy intake, the diets differed significantly from each other (61.63% and 57.15%, respectively). In their study on Awassi lambs, Al Jassim *et al.* (1996) observed that BW gain was highly correlated with ME retention. In our study, ME retention was similar between the two treatments which explains the similar ADG for lambs fed on the two experimental diets.

Table 4 Energy metabolism (mean) of South African Mutton Merino lambs fed the two different diets

	Diet 1 (Control)	Diet 2 (CLA)	SEM
Dry matter intake (g/day)	1222.92	1385.04	96.803
Gross energy (MJ/kg)	18.293	18.627	
Energy intake (MJ/day)	22.37	25.80	1.805
Faecal energy (MJ/day)	5.92	7.92	0.578
Urinary energy (MJ/day)	0.88	0.97	0.058
Methane gas production (MJ/day)	1.79	2.06	0.144
Total energy excreted (MJ/day)	8.59	10.96	0.735
Faecal energy (% of energy intake)	26.35^{b}	30.88^{a}	0.797
Urinary energy (% of energy intake)	4.01	3.97	0.292
Total energy excreted (% of energy intake)	38.37^{b}	42.85^{a}	0.796
Energy retention (MJ/day)	13.78	14.84	1.103
Energy retention (% of energy intake)	61.63^{a}	57.15^{b}	0.796
Metabolizable energy (MJ/kg)	11.27^{a}	10.65^{b}	0.123

[a,b,c] Means in the same row with different superscripts differ ($P \leq 0.05$).

The ME-value obtained for Diet 1 in the digestibility study (Table 4) differs from that calculated from the nutritive values of the feeds (Table 1) (Diet 1: 11.27 *vs.* 10.66 MJ/kg DM). Therefore, it seems that the calculated ME-values of these dietary components cannot under all circumstances be used as an indicator of the real ME-values.

Nitrogen metabolism and retention of the SAMM lambs on the two diets can be seen in Table 5. The numerical higher daily nitrogen intake (g/day) of the lambs fed Diet 2 is consistent with their dry matter intake. Faecal nitrogen excretion was similar for the two treatments (P = 0.4757), suggesting a relative similar nitrogen digestion (Nolte & Ferreira, 2004). Total N excretion (g/day), and when expressed as a percentage of N intake, tended to be higher for lambs receiving Diet 2. When N retention (g N/kg $BW^{0.75}$/day) was corrected for both endogenous urinary N (EUN) and metabolic faecal N (MFN), there was no significant difference in N retention between Diets 1 and 2. The similar N retention of the lambs in the two treatments was also reflected in the similar average daily gains obtained during the feeding trial (Stanford *et al.,* 1999). Even when N retention was expressed as a percentage of N intake, the lambs fed the two different diets retained similar amounts of N.

Table 5 Nitrogen metabolism (mean) of South African Mutton Merino lambs fed two different diets

	Diet 1 (Control)	Diet 2 (CLA)	SEM
Dry matter intake (g/day)	1222.92	1385.04	96.803
Nitrogen intake (g/day)	30.73	32.72	2.305
Nitrogen intake (g N/kg BW0.75/day)	1.84	2.00	0.121
Faecal nitrogen (g/day)	7.18	8.19	0.671
Urinary nitrogen (g/day)	17.60	18.87	1.107
Total nitrogen excreted (g/day)	24.77	27.06	1.404
Faecal nitrogen (% of nitrogen intake)	22.88	24.99	0.589
Urinary nitrogen (% of nitrogen intake)	60.01	60.11	4.494
Total nitrogen excreted (% of nitrogen intake)	82.90	85.10	4.204
Metabolic faecal nitrogen (g/day)	6.11	6.93	0.484
Endogenous urinary nitrogen (g/day)	2.98	2.94	0.047
Nitrogen retention (g N/kg BW0.75/day)	0.90	0.94	0.122
Nitrogen retention (% of nitrogen intake)	47.38	45.38	3.757

The proximate chemical compositions of the *M. longissimus dorsi* and *M. biceps femoris* are presented in Table 6. Supplementing Diet 2 with rumen-protected CLA had no significant influence on the proximate composition of the *M. longissimus dorsi* and *M. biceps femoris* of the lambs. The values for protein, fat and consequently energy analysed from the *M. longissimus dorsi* in the current study were higher than that reported by Sheridan (2001) for the *M. semimembranosus* (19.52%, 3.19% and 449.78 kJ, respectively) from SAMM lambs finished on a feedlot diet. Cloete *et al.* (2004), on the other hand, reported proximate chemical values for SAMM sheep that were similar to those found in this study. Jamora & Rhee (1998) suggested that the fat content of muscle tissue of a typical feedlot lamb in the USA sheep industry is 5.25g/100g muscle tissue. Values found in this study were slightly lower than these.

A possible explanation for the higher fat content observed in this investigation, compared to that reported by Sheridan (2001), may be due to a difference in final age and final body weight between the two studies (Demeyer & Doreau, 1999), as the lambs in the current study were approximately two months older (7 months of age) and weighed on average 1.21 kg heavier. The fact that different muscles are being compared may also be a contributing factor (Martínez-Cerezo *et al.,* 2005). Although not significant, the fat content of both the *M. longissimus dorsi* and *M. biceps femoris* in Diet 2 tended (P = 0.3964 and 0.2956, respectively) to be lower (and consequently a lower calculated energy value) than that of the animals fed Diet 1. Research in laboratory animals (Park *et al.,* 1999), swine (Thiel-Cooper *et al.,* 2001; Wiegand *et al.,* 2001) and dairy cattle (Chouinard *et al.,* 1999; Baumgard *et al.,* 2001) has demonstrated that CLA supplementation reduces animal adiposity and alters lipid metabolism. Gassman *et al.* (2000) also reported a decreased intramuscular fat content, and consequently a reduced marbling score of finishing steers fed a diet containing 2.5% rumen-protected CLA. According to Pariza *et al.* (2001) CLA (specifically the C18:2 *trans*-10, *cis*-12 isomer) would block body fat gain, but not necessarily reduce body fat level which had accumulated prior to CLA administration. Results from his study, as well as work done by Gillis *et al.* (2004) indicate that feeding rumen-protected CLA salt does not affect animal performance in sheep and in beef cattle to the same extent as reported for swine (Thiel-Cooper *et al.,* 2001; Wiegand *et al.,* 2001) and laboratory animals (Park *et al.,* 1999).

Although not significant, there was a tendency that the fat content of the two treatments had a reverse relationship to the moisture content (r = -0.63, P = 0.0932; r = -0.45, P = 0.2592, respectively for Diet 1 and 2) in *M. longissimus dorsi.* The *M. biceps femoris* exhibited the same tendency for both treatments (r = -0.51, P = 0.1919; r = -0.48, P = 0.2242, respectively for Diet 1 and 2). Muscles with the highest fat content were characterised by a lower

moisture content (Table 6). This is in accordance with the findings of Ono *et al.* (1984), Dransfield *et al.* (1990) and Martínez-Cerezo *et al.* (2005) who reported the same tendency for cross-bred lambs, Suffolk ram lambs and Spanish sheep breeds, respectively, raised on a feedlot diet.

The treatments did not influence ($P > 0.05$) the protein content of the meat, probably because the diets were isonitrogenous (Sheridan *et al.*, 2003b). There is evidence indicating that dietary CLA may enhance whole body protein accretion in mice (Park *et al.*, 1997). In rats (Azain *et al.*, 2000) and pigs (Ostrowska *et al.*, 1999), CLA appears to enhance lean body mass gain relative to fat mass gain. According to Rowe *et al.* (1999) a negative correlation exists between lipid and protein content of the meat, suggesting that higher intramuscular lipid content will produce a lower muscle protein content. Webb & Casey (1995) reported that carcass fat percentage is negatively correlated with both the percentage muscle and bone in the carcasses of SAMM wethers.

Table 6 Mean proximate composition (on an *as is* basis) of *M. longissimus dorsi* and *M. biceps femoris* of the SAMM lambs fed the two different diets

	Diet 1 (Control)	Diet 2 (CLA)	SEM
M. longissimus dorsi			
Moisture (%)	74.01	74.38	0.214
Ash (%)	1.18	1.17	0.012
Protein (%)	20.16	20.17	0.144
Fat (%)	4.20	4.02	0.103
Energy (kJ)[1]	498.15	491.64	4.754
M. biceps femoris			
Moisture (%)	75.00	75.06	0.189
Ash (%)	1.20	1.20	0.012
Protein (%)	19.58	19.76	0.222
Fat (%)	3.74	3.45	0.133
Energy (kJ)[1]	471.22	463.51	5.916

[1] Total energy (kJ) = (g protein in 100g sample × 17) + (g fat in 100g sample × 37) (As gazetted: SA Act No 54 of 1972).

The mean values for the different effects of the physical attributes investigated are presented in Table 7. No significant differences were observed in the initial, as well as the ultimate pH and temperature readings taken 45 min and 48 h, respectively, post-mortem. The lambs on Diet 1 tended to have a slightly higher mean ultimate pH ($P = 0.5175$) compared to the lambs on Diet 2. It is a well-known fact that the ultimate pH of the muscle is an important contributing factor to the quality of meat (Hoffman *et al.*, 2003). According to Devine *et al.* (1993) an ultimate pH value greater than 5.8 is regarded as undesirable. In rested, fed sheep the ultimate pH in the *M. longissimus lumborum* is less than 5.6 and stressful handling causes a high ultimate pH (Martínez-Cerezo *et al.*, 2005). Lawrie (1985) found that the bacteriological stability of chilled meat is adversely affected by an ultimate pH value above 6.0. The mean pH values of the two diets were all very close or within the normal mutton pH range of 5.4-5.86 (Safari *et al.*, 2001).

Conflicting reports regarding the relationship between pH and tenderness (WBSF) are found in the literature. Consumer surveys have shown that tenderness is considered the most important component of meat quality (Young *et al.*, 1993) and that a WBSF value greater than 49 N is regarded as tough by consumers (Safari *et al.*, 2001). Devine *et al.* (1993) reported an increase in shear force of lambs with higher pH values in the range of 5.4-6.0, while research by Silva *et al.* (1999) oppose these findings. The latter observed that tenderness of beef assessed by WBSF increased linearly with ultimate pH. Purchas (1990) and Jeremiah *et al.* (1991) found a curvilinear relationship between pH and tenderness, with a minimum tenderness between pH 5.8 and 6.2. Whilst Safari *et al.* (2001) found no relationship between pH and shear force value and cooking loss in six diverse lamb genotypes.

There is also a great deal of controversy amongst researchers regarding the relationship between tenderness and the intramuscular fat content of the meat. Silva *et al.* (1999), for example, stated that intramuscular fat content is not correlated with tenderness. This is inconsistent with the negative correlation ($P < 0.001$) obtained by Seideman *et al.* (1987) and Okeudo & Moss (2005) between marbling fat and shear force of the *M. longissimus dorsi* in cattle and sheep, respectively. Schönfeldt *et al.* (1993) describe a study in which a positive relationship was found between tenderness of meat and fatness of ovine carcasses.

According to Koohmaraie (1990) the major factors influencing tenderness are collagen, muscle proteinases and fat deposition rate; particularly, levels of collagen and the relative amounts of highly cross-linked collagen. In the ovine, Young & Braggins (1993) showed that

collagen solubility declined with age but its concentration remained unchanged from 4 months to 5 years of age.

Table 7 Physical characteristics of the *M. longissimus dorsi* and subcutaneous fat for the SAMM lambs fed the two different diets

	Diet 1 (Control)	Diet 2 (CLA)	SEM
M. longissimus dorsi			
pH_{45}	6.96	6.83	0.086
$Temperature_{45}$ (°C)	33.39	32.44	0.488
pH_{48}	5.41	5.34	0.047
$Temperature_{48}$ (°C)	2.96	3.19	0.176
Drip loss (%)	1.58	1.65	0.040
Cooking loss (%)	32.89^{a}	30.97^{b}	0.434
Shear force (N/1.27 cm diameter)	34.65	36.73	0.998
L* value	39.40^{a}	38.06^{b}	0.343
a* value	10.39	10.87	0.156
b* value	9.22^{a}	8.43^{b}	0.164
Hue angle (°)	41.67^{a}	37.72^{b}	0.748
Chroma	13.91	13.83	0.138
Fat thickness (2^{nd} – 3^{rd} last thoracic vertebrae)	4.66	5.20	0.349
Fat thickness (4^{th} – 5^{th} lumbar vertebrae)	2.54	3.73	0.406
Subcutaneous fat			
L* value	70.05^{b}	76.52^{a}	0.878
a* value	1.80^{a}	0.78^{b}	0.168
b* value	11.37	10.98	0.233
Hue angle (°)	80.78^{b}	86.17^{a}	0.885
Chroma	11.55	11.07	0.233

^{a,b,c} Means in the same row with different superscripts differ ($P \leq 0.05$).

Although no literature could be sourced that investigated the effect of CLA on mutton tenderness, a number of studies have been conducted to investigate the effect of dietary CLA on pork quality. For example, Dunshea *et al.* (2005) report a significant reduction in tenderness of pork with the inclusion of CLA in their diets. The decrease in tenderness was consistent with a 2.9 N increase in the shear force value. Results from the current study show that the average shear force value of the *M. longissimus dorsi* from the lambs fed the supplemental dietary CLA (Diet 2) tended to be higher than those fed Diet 1 ($P = 0.2995$).

Our results also indicate that the muscle temperature measured after 48 h post-mortem tended to be higher in Diet 2 ($P = 0.5423$), which might be explained by a thicker, although not significant, back-fat thickness measured at the 2^{nd}–3^{rd} last thoracic vertebrae. This finding coincides with the results obtained by Okeudo & Moss (2005), who showed that rate of chilling of sheep carcasses were negatively affected by back fat cover and carcass weight. Large carcasses have a proportionately smaller surface area and the fat cover acts as an insulatorary barrier to heat loss. Moreover, intramuscular fat content also lowers the aggregate thermal conductivity of the muscle (Hill *et al.,* 1967).

Although the two diets had no effect ($P > 0.05$) on the percentage drip loss, it significantly influenced the cooking loss of the *M. longissimus dorsi.* Dietary CLA supplementation of pig diets resulted in a significant reduction of approximately 5% in drip loss of pork (Dunshea *et al.,* 2005). According to the work done by Hopkins & Fogarty (1998), a lower cooking loss is associated with a higher fat content in the meat. Inconsistent with these findings, Schönfeldt *et al.* (1993) and Carpenter & King (1965) reported significantly higher total cooking loss when ovine carcasses had increased fat content. In the current study the values obtained for cooking loss of lambs fed Diet 1 were higher ($P \leq 0.05$) than that of lambs fed Diet 2. This might be due to the intramuscular fat content that tended to be higher ($P = 0.3964$) (Schönfeldt *et al.,* 1993, Carpenter & King, 1965) in Diet 1 than in Diet 2. Purchas *et al.* (1969) showed that although total cooking loss was positively correlated to fat percentage of the carcass side when the whole loin was cooked, cooking loss from the muscular tissue was poorly or negatively correlated to fat percentage of the carcass side. It is evident from the work done by Safari *et al.* (2001) that a negative correlation exists between cooking loss and juiciness, which will in turn affect the eating quality and acceptability of the meat. Cooking loss and juiciness are significantly ($P < 0.001$) correlated with tenderness assessed by both WBSF and a sensory panel (Silva *et al.,* 1999; Okeudo & Moss, 2005). A reduction in cooking loss will lead to an increase in juiciness, and thus an increase in the tenderness of the meat. Bouton *et al.* (1973) considered that the higher water holding capacity of meat of higher ultimate pH contribute to their high tenderness. Okeudo & Moss (2005) suggest a positive relationship between moisture content of meat and cooking loss and that it is a more important determinant of cooking loss than intramuscular lipid content.

All the lambs displayed L* values above 34. According to Hopkins (1996, as cited by Velasco *et al.,* 2004) this indicates that meat is light-coloured and acceptable to consumers. The colour of the *M. longissimus dorsi* was significantly darker (lower L*) in the lambs fed Diet 2 than the lambs fed Diet 1. In accordance with our results, Joo *et al.* (2002) reported that 5% dietary CLA caused significantly lower L* values than the control in pork. They

assumed that dietary CLA improved the oxidative stability of intramuscular fat in pork. Hur *et al.* (2004) again found a higher L* value by substitution of CLA sources for fat in beef patties after 7 days of cold storage. There was no significant difference in the index of redness between the two treatments, but the yellowness index (b*) was higher ($P \leq 0.05$) and the hue angle wider ($P \leq 0.05$) in the lambs fed Diet 1. A study by Poulson *et al.* (2004) showed that meat from steer calves fed supplemental rumen-protected CLA had a lower red colour stability over a period of 15 days than calves not receiving the supplementation. Their results suggested that feeding CLA, which is a rich source of unsaturated fatty acids, will decrease the red colour in meat, mainly because of oxidation. However, Du *et al.* (2000) suggested that dietary CLA treatment not only reduced lipid oxidation, but also improved colour stability during storage. Thiel-Cooper *et al.* (1998) also reported increases in a* values with increasing levels of CLA, from 0 to 1% in the diet, suggesting that dietary CLA may protect meat colour.

The significant lower yellowness index of meat from lambs fed Diet 2 is also in accordance with the findings of Joo *et al.* (2002) and Poulson *et al.* (2004). Poulson *et al.* (2004) noted by means of visual evaluation, that the meat from calves fed supplemental rumen-protected CLA appeared to brown faster than that of animals not receiving supplemental rumen-protected CLA. The colour of meat is due to a balance between oxymyoglobin oxidation and metmyoglobin reduction (Faustman & Cassens, 1990) and the rate of meat discolouration is closely related to the rate of myoglobin oxidation induced by lipid oxidation (Yin & Faustman, 1993). Young *et al.* (1993) and Priolo *et al.* (2002) reported that a darker meat colour might be due to a slightly higher ultimate pH, since high pH meats tend to have a darker colour (Lawrie, 1985; Velasco *et al.*, 2004). This was not the case in the current study.

According to Mancini & Hunt (2005) fat colour has received less attention in the literature; however, it has been proved that the diet can affect carotenoid content of the fat, which can influence the fat colour. French *et al.* (2000) reported an inverse and linear correlation ($r = -0.52$) between the yellowness of subcutaneous fat and the amount of dietary concentrate. This was due to a lower amount of β-carotene in concentrates than in pasture, resulting in less carotene accumulation in fat of animals fed concentrates. Similarly, Priolo *et al.* (2002) found that subcutaneous fat colour from grass fed lambs was more yellow (b*) than that from lambs fed a concentrate. The fact that the lambs in both treatments received a concentrate-based diet, accounts for the lack of significant difference in yellowness observed. Mancini *et al.* (2005) obtained results indicating that darker subcutaneous fat colour resulted from the redox state of the residual haemoglobin, being either deoxyhaemoglobin or methaemoglobin, in the

blood capillaries (Irie, 2001). This might be a possible explanation for the significantly darker subcutaneous fat colour observed for Diet 1.

Conclusion

The results from this study indicate that the inclusion of rumen-protected linoleic acid (CLA) in feedlot finishing diets of SAMM lambs had no significant effect on the growth performance and feed conversion efficiency. However, the DM, OM and NDF digestibility were adversely affected by the inclusion of rumen-protected CLA. The reduced DM and OM digestibility might be due the higher dry matter intake during the digestibility trial period, which lead to increased passage rate and reduced rumen retention times, resulting in decreased digestibility. Lambs fed the CLA diet had significantly lower energy retention, when expressed as a percentage of energy intake, however, the nitrogen content of the two feedlot diets were metabolised with similar efficiency. The fact that no difference occurred in energy retention (MJ/kg) explains the similar ADG for the lambs fed the two experimental diets.

Dietary treatments also had no effect ($P > 0.05$) on the proximate chemical composition of the two muscles, *M. longissimus dorsi* and *M. biceps femoris,* investigated. Percentage cooking loss differed ($P \leq 0.05$) between the two diets, and lambs fed the control diet (Diet 1) produced meat with a significantly higher L* and b* value and consequently a wider hue angle.

Based on the results obtained in the present study it may be concluded that supplementing feedlot finishing diets of SAMM lambs with 2.5% rumen-protected CLA did not have a negative effect on production efficiency, as well as on meat quality.

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Chapter 6

Effects of rumen-protected conjugated linoleic acid (CLA) in feedlot finishing diets on fatty acid composition, cholesterol content and sensory characteristics of the meat of South African Mutton Merino lambs

Abstract

Sixteen South African Mutton Merino (SAMM) lambs were used in a feedlot finishing investigation to evaluate the effect of feeding a diet containing supplemental rumen-protected conjugated linoleic acid (CLA) on the muscle, as well as adipose tissue fatty acid composition, cholesterol content and sensory characteristics of meat. Two diets were formulated on an isonitrogenous and isoenergetic basis (15.50% crude protein (CP) and a metabolizable energy content of 10.64 MJ/kg on a dry matter (DM) basis). The lambs were fed *ad libitum* for 24 days, where after they were slaughtered. The *longissimus dorsi* and the *biceps femoris* muscles of each carcass were removed for determination of fatty acid composition, cholesterol content and sensory characteristics. Three adipose tissue types (subcutaneous, intermuscular, as well as kidney and caul fat) were also analysed for fatty acid composition. Diet had no significant effect on any of the five sensory attributes measured. Fatty acid composition was significantly affected by dietary treatment. Palmitic (C16:0), stearic (C18:0) and oleic (18:1n-9) acid were the major fatty acids in both muscles, as well as in the three adipose tissue types. The rumen-protected CLA increased the CLA (C18:2n-6) contents in both the *longissimus dorsi* and *biceps femoris* muscles, although the increase was only significant for the *longissimus dorsi* muscle. Cholesterol content of both muscle was not significantly influenced by the dietary treatment. It can be concluded that although 2.5% supplemental rumen-protected CLA had no major affect on the sensory characteristics, its variable effect on fatty acid composition of meat and fat depots of SAMM lambs warrants further investigation.

Key words: conjugated linoleic acid; fatty acid composition; cholesterol content; sensory attributes; South African Mutton Merino lambs; feedlot

Introduction

Fatty acids are the most important lipid fraction of meat (Russo *et al.,* 1999). The fat type or fatty acid composition of meat is an important diet or health concern to consumers. It is also a primary factor that determines product shelf-life or storage stability and flavour (Webb & Casey, 1995). The fatty acid composition of tissues derived from ruminants is less affected by dietary lipid composition than that of monogastric animals (Rhee, 1992). The great disparity between dietary and tissue fat in ruminants arises primarily from hydrogenation of dietary lipids by rumen microbes (Byers & Schelling, 1993). The ruminal conversion (saturation) of dietary unsaturated fatty acids, however, is normally not complete and bovine studies have shown that dietary fatty acid composition differences can result in differences in tissue fatty acid composition (Marmer *et al.,* 1984; Chilliard, 1993).

The amount of fat in the human diet, and especially the proportion of saturated fatty acids (Kritchevsky, 1998) and dietary cholesterol levels (Abu-Tarboush & Dawood, 1993) have been considered as major risk factors for coronary heart diseases (CHD). The ratio between polyunsaturated and saturated fatty acids (P:S) and the ratio between n-6 and n-3 fatty acids are considered two important indexes for nutritional evaluation of fat (Wood *et al.,* 2003). According to nutritional guidelines, a P:S ratio of 0.45 or higher (Warris, 2000) and a n-6:n-3 ratio of 4 or lower is recommended (Enser *et al.,* 1998). Ruminant meats have a low P:S ratio because of the hydrogenating action of the rumen microorganisms on dietary fatty acids but the n-6:n-3 ratio is beneficially low, especially on grass diets (Enser *et al.,* 1998). Only in the last few years have researchers begun to oppose the so-called "danger" of saturated fatty acids and the "goodness" of unsaturated fatty acids. For example, it has been proved that stearic acid (C18:0) is easily converted into oleic acid (C18:1n-9) which, from a dietetic point of view, is one of the desirable fatty acids (Rhee, 1992). The danger has been noticed of excessive consumption of polyunsaturated fats that are readily oxidised with the formation of radicals of oxygen and aldehydes, which are both compositions that became part of carcinogenesis and ageing of tissues (Russo *et al.,* 1999).

Consumers are very conscious of their dietary intake of high-fat animal food that contains saturated fatty acids and cholesterol, which in turn, elevate serum cholesterol (Flynn *et al.*, 1985). Jiminez-Colmenero *et al.* (2001) suggested that cholesterol intake should be limited to 300 mg/day. Rhee *et al.* (1982) reported that cholesterol may be avoided by selecting meat with little or no marbling fat and by trimming away separable fat, especially subcutaneous or external fat. The benefit of removing the separable fat is to reduce the intake of

triacylglycerols rather than cholesterol intake since triacylglycerols is the starting material for the synthesis of cholesterol (Rhee *et al.,* 1982). It is generally accepted that, in humans, plasma cholesterol concentration is influenced by the fatty acid composition of dietary fat (Flynn *et al.,* 1985). It is a well-known fact that palmitic acid (C16:0) increases total serum cholesterol levels, whereas stearic acid (C18:0) has no effect. Oleic acid (C18:1n-9) seems to reduce both the low-density lipoprotein (LDL) cholesterol and triglycerides content of the blood, without reducing the high-density lipoprotein (HDL) cholesterol (Grundy, 1997; Russo *et al.,* 1999).

For ruminants, the most important n-3 fatty acid source is grass and linseed (Choi *et al.,* 2000). However, to be effective at increasing the tissue n-3 polyunsaturated fatty acid (PUFA) content, the dietary PUFA should escape biohydrogenation in the rumen.

As far as most consumers are concerned, meat should contain a very small amount of fat. Too much fat discourages the purchase of meat and is normally removed either before cooking or during the meal by the consumer. However, some fat is always present in meat and indeed is required to impart flavour and juiciness (Melton, 1990). Consumers tend to evaluate meat quality on the basis of tenderness, juiciness and flavour of the cooked meat (Carpenter, 1966). The acceptability of red meat after purchase is determined almost exclusively by the satisfaction derived from its consumption (Jeremiah *et al.,* 1991). Although texture or tenderness is the predominant quality determinant and is probably the most important sensory characteristic of red meat (Koohmaraie, 1990), a species specific flavour is also an important sensory attribute (Sink, 1979).

The South African Mutton Merino (SAMM) is a dual-purpose (mutton and wool) sheep breed and was selected for the study due to its high growth rate and capability to produce a slaughter lamb at an early age with good meat quality attributes (Cloete *et al.,* 2004). It has a high fertility, good conformation, good feed conversion ratio and adaptability and is therefore popular in feedlot production systems (Neser *et al.,* 2000).

This study was conducted to determine the effect of rumen-protected CLA in a feedlot finishing diet on the fatty acid composition, cholesterol content and sensory characteristics of SAMM lambs.

Materials and methods

Animals and sampling

Sixteen South African Mutton Merino (SAMM) intact male lambs were used in this investigation. All the lambs were weaned and shorn prior to entering the feedlot. All the animals were dewormed with a broad-spectrum drench to eliminate all internal parasites on their arrival at the University of Stellenbosch experimental farm, Welgevallen. An extra group of eight lambs, selected at random, was slaughtered before the feeding trial commenced to serve as an index of the initial fatty acid composition of the lambs (Initial baseline group). The average initial body weight (BW) of the lambs used in this investigation was 34.8 ± 2.60 kg. The lambs were randomly allocated into individual pens (1 m x 2 m) in an enclosed but adequately ventilated shed with a wooden slatted floor. Lambs were divided in two groups of equal initial weights. Both groups were randomly assigned to one of two treatments. The pelleted diets were formulated according to the NRC (1985) recommendations for lambs, on an isonitrogenous and isoenergetic basis. Diet 1 was formulated to serve as a control diet, whilst Diet 2 was supplemented with a rumen-protected conjugated linoleic acid (CLA) (Table 1). The lambs had *ad libitum* access to the feed and fresh water. The rumen-protected CLA source (Luta-CLA® 20 P, BASF Animal Nutrition, P.O. Box 1783, 1620, Kempton Park, South Africa) contained 20% CLA, and consisted of two isomers present in the same concentration (10%): C18:2 *cis*-9, *trans*-11 and C18:2 *trans*-10, *cis*-12.

After the 24-day feeding trial and digestibility trial of a week, the lambs were slaughtered at a commercial abattoir using standard South African procedures. After being electrically stunned (4 seconds at 200 volts) the lambs were exsanguinated and the carcasses suspended by the Achilles tendon to bleed. After evisceration, the carcasses were suspended in a cooler at 2°C for 48 hours. No electrical stimulation of the carcasses was applied.

The carcass characteristics and physical meat quality attributes of the lambs have been discussed in Chapter 5. The *M. longissimus dorsi* was removed from the left and right hand side of the carcass (the muscle was removed between the 2^{nd}–3^{rd} last thoracic vertebrae and the 4^{th}–5^{th} lumbar vertebrae) and the *M. biceps femoris* was dissected from the right hind leg. A subcutaneous fat, intermuscular fat (behind the *M. biceps femoris)*, as well as a kidney and caul fat sample from each carcass were taken. The muscle and fat samples were labelled, vacuum packed, frozen and stored at –18°C until further analysis.

Table 1 Physical and chemical composition of the two diets fed to the SAMM lambs

	Diets	
	Diet 1 (Control)	Diet 2 (CLA)
Physical composition[1]		
Maize meal	23.00	14.80
Lucerne hay	17.00	25.24
Citrus pulp	4.00	2.50
Maize cobs	8.00	7.50
Hominy Chop	25.44	25.00
Sunflower hulls	0.00	5.00
Wheat bran	9.10	2.50
Linseed meal	0.00	3.00
Full-fat soybeans	2.00	0.00
Ammonium chloride	0.50	0.50
Limestone	1.40	1.70
Mono calcium phosphate	0.00	0.10
Salt	0.60	0.60
Urea	0.70	0.80
CLA[3]	0.00	2.50
Molasses	7.50	7.50
Premix B[4]	0.25	0.25
Acid Buf[5]	0.50	0.50
Avatec[6]	0.02	0.02
Chemical composition[2]		
Organic matter (%)	93.05	92.56
Ether extract (%)	4.29	5.62
Crude Protein (%)	15.70	14.77
Acid detergent fibre (%)	16.21	20.28
Neutral detergent fibre (%)	30.66	35.21
Gross energy (MJ/kg)	18.29	18.63
Metabolizable energy (MJ/kg)[7]	10.66	10.62

[1]On an air dry basis; [2]Analysed values on a DM basis; [3]Rumen-protected conjugated linoleic acid; supplied by BASF Animal Nutrition (P.O. Box 1783, 1620, Kempton Park, South Africa); [4]A standard mineral (macro and micro) and vitamin supplement; formulated and supplied by BASF Animal Nutrition; [5]A rumen buffer; supplied by Nutec Southern Africa (234 Royston Road, Willowton, Pietermaritzburg, KwaZulu-Natal, South Africa); [6]A growth promoter, lasalocid sodium; supplied by Instavet Import and Export Pty (Ltd) (P.O. Box 346, 2163, Kya Sand, South Africa); [7]Based on the nutritive value of feeds according to laboratory determined values by Senwesko Feeds.

Fatty acid analysis

Fatty acid analysis were carried out on the *M. longissimus dorsi* and the *M. biceps femoris* from the right hand side of the carcass as well as on the mentioned fat depots. The frozen muscle samples were cut into smaller portions, minced three times through a 2 mm sieve to ensure homogeneity, and analysed.

The fatty acid content was determined by the method described by Tichelaar *et al.* (1998). After thawing the meat a 2 g sample was extracted with a chloroform:methanol (2:1 v/v) solution according to a modified method of Folch *et al.* (1957). All the extraction solvents contained 0.01% butylated hydroxytoluene (BHT) as an antioxidant. A polytron mixer (Kinematica, type PT 10-35, Switzerland) was used to homogenize the sample within the extraction solvent. Heptadecanoic acid (C17:0) was used as an internal standard to quantify the individual fatty acids. A sub-sample of the extracted lipids was transmethylated for 2 h at 70°C using methanol:sulphuric acid (19:1 v/v) as transmethylating agent. After cooling, the resulting fatty acid methyl esters (FAMEs) were extracted with water and hexane. The top hexane phase was transferred to a spotting tube and dried under nitrogen.

The FAMEs were purified by TLC (silica gel 60 plates) and analysed by gas-liquid chromatography (GLC). A Varian Model 3300 was used that is equipped with flame ionisation detection using 60 m BPX70 Capillary columns of 0.25 mm internal diameter (SGE, Australia). Gas flow rates were: hydrogen, 25 ml/min; and hydrogen carrier gas 2-4 ml/min. Temperature programming was linear at 3°C/min, with an initial temperature of 150°C, a final temperature of 220°C, an injector temperature of 240°C and a detector temperature of 250°C. The FAMEs in the total lipids were identified by comparison of the retention times with those of a standard FAME mixture (SupelcoTM 37 Component FAME Mix, Catalogue Number 18919-1AMP, Lot number, LB-16064. Sigma Aldrich Inc. North Harrison Road, Bellefonte, PA 16823-0048, USA).

Cholesterol analysis

From the same lipid extraction used for fatty acid determination, a sub-sample was used for cholesterol determination. After drying the sub-sample under nitrogen, Stigmasterol (3-B-hydroxy-24-ethyl-5.22-cholestadiene; Sigma Chemical Co., St Louis, MO, USA) was added as internal standard and 6% ethanolic potassium hydroxide (KOH) was used to saphonifize the extraction for 2 h at 70°C in a heating block. After cooling, distilled water and hexane were added and the resulting extraction was analysed by GLC (Varian Model 3700, equipped with flame ionisation detection). A 1.2 m glass column of 2 mm internal diameter packed with 3% SP2401 on 100/120 mesh Supelcoport (Supelco Inc., Bllefonte, PA, USA) was used.

Gas flow rates were: hydrogen, 20 ml/min; air, 200 ml/min and nitrogen (carrier gas), 25 ml/min. Temperatures were as follow: injector temperature (280°C); column temperature (225°C) and detector temperature (290°C).

Descriptive sensory analysis

The frozen *M. longissimus dorsi* from the left hand side of the carcass was used for sensory analysis. The *longissimus dorsi* muscle samples were defrosted at a temperature of 3-4°C for a period of 24 h, wrapped individually in cooking bags and placed fat-side up on an aluminium foil covered rack of an open roasting pan. The samples were cooked at 160°C in two electric Defy 835 ovens connected to a computerized electronic temperature control system (Viljoen *et al.,* 2001) to an internal temperature of 68°C (AMSA, 1978). Following a standing period of 5 mins at room temperature, all visible subcutaneous fat was removed from each sample. Fourteen 1.5 cm x 1.5 cm cubed samples were taken from the middle of each sample and wrapped immediately in aluminium foil. The samples were placed in preheated glass ramekins, marked with a three digit random code and were kept in a preheated oven at 100°C and evaluated within 10 mins.

A seven-member descriptive panel was selected and trained according to the generic descriptive analysis techniques and the procedures described in the American Meat Science Association guidelines (AMSA, 1978). The panel evaluated the meat for the following sensory attributes: aroma intensity, initial impression of juiciness, sustained juiciness, tenderness and overall lamb flavour by means of an unstructured 100 mm line scale. Table 2 depicts the definitions of the attributes used in the sensory analysis. The panelists were seated in individual booths in a temperature and light-controlled room, receiving a set of three samples served in a complete randomised order per session. Crackers, apple slices and distilled water were used to cleanse the palate between samples (AMSA, 1978).

Statistical analysis

The effect of supplemental dietary rumen-protected conjugated linoleic acid (CLA) in feedlot finishing diets of SAMM lambs, on the fatty acid composition and cholesterol content of the *M. longissimus dorsi* and *M. Biceps femoris* were analysed by using the nonparametric PROC ANOVA of SAS (2000). Least Square Means were also calculated. Significance was declared at $P \leq 0.05$.

The experiment consisted of a completely randomized design with two dietary treatments (Diet 1 and 2). An experimental unit was regarded as a carcass from which samples were

taken for measurements. The sensory scores were subjected to analysis of variance using SAS (SAS, 2000). The Shapiro-Wilk test was performed to test for non-normality (Shapiro & Wilk, 1965). In some cases deviations from normality were the cause of one or two outliners, which were removed before the final analysis. Student's t-Least Significant Differences (LSD) were calculated at the 5% significance level to compare treatment means.

Table 2 Definition of attributes for descriptive sensory analysis

Attribute and Scale	Definition
Lamb aroma 0=Low; 100=High	Aroma associated with the animal species
Initial juiciness 0=Dry; 100= Juicy	The amount of fluid exuded on the cut surface when pressed between forefinger and thumb
Sustained juiciness 0=Dry; 100=Juicy	Degree/amount of water perceived during mastication
Tenderness 0=Tough; 100=Tender	Force needed to compress the sample of meat between molar teeth on the first bite
Lamb flavour 0=Low; 100=High	Flavour associated with the animal species

Results and discussion

The fatty acid profile of the two diets (Diet 1 and 2) fed to the lambs are presented in Table 3. Diet 2 contained a higher concentration of total saturated fatty acids compared to Diet 1 (31.88 *vs* 18.33 g/100 g). Conversely, the concentration of total unsaturated fatty acids was higher for Diet 1 than for Diet 2 (81.67 *vs* 68.12 g/100 g), with oleic acid (C18:1n-9) (29.25 g/100 g) being the most prominent mono-unsaturated fatty acid (MUFA) and linoleic acid (CLA, C18:2n-6) (47.83 g/100 g) and α-linolenic acid (C18:3n-3) (3.67 g/100 g) the most prominent polyunsaturated fatty acids (PUFA). This resulted in Diet 1 having a higher PUFA:SFA ratio than Diet 2 (2.83 *vs* 1.29). The inclusion of full-fat soybeans in Diet 1 might explain the higher concentration of CLA (C18:2n-6) in Diet 1 compared to Diet 2, as C18:2n-6 is the predominant fatty acid in soybeans (Beaulieu *et al.*, 2002). Palmitic acid (C16:0) was the most predominant saturated fatty acid (SFA) in Diet 2 (19.13 g/100 g), followed by stearic acid (C18:0) (11.11 g/100 g). According to Rhee *et al.* (2000), palmitic and stearic acid are two primary fatty acids. As in Diet 1, oleic acid (C18:1n-9) was the most predominant

Table 3 Fatty acid profile (g/100g of identified fatty acids) of the two diets fed to the SAMM lambs

Fatty acid	Diets	
	Diet 1 (Control)	Diet 2 (CLA)
14:0	0.11	0.34
16:0	13.77	19.13
18:0	3.24	11.11
20:0	0.64	0.65
22:0	0.34	0.28
24:0	0.24	0.37
14:1	0.00	0.00
16:1n-7	0.13	0.14
18:1n-9	29.25	26.47
20:1n-9	0.35	0.26
22:1n-9	0.06	0.06
24:1n-9	0.04	0.09
18:2n-6	47.83	32.29
18:3n-6	0.03	0.03
18:3n-3	3.67	8.40
20:2n-6	0.05	0.04
20:3n-6	0.03	0.03
20:3n-3	0.03	0.07
20:4n-6	0.03	0.00
20:5n-3	0.05	0.05
22:2n-6	0.04	0.04
22:5n-3	0.04	0.06
22:6n-3	0.05	0.09
SFA[1]	18.33	31.88
MUFA[2]	29.83	27.02
PUFA[3]	51.84	41.10
DFA[4]	84.91	79.23
PUFA:SFA[5]	2.83	1.29
Σn-6[6]	48.01	32.43
Σn-3[7]	3.83	8.67
n-6:n-3[8]	12.52	3.74

[1]Saturated fatty acids; [2]Mono-unsaturated fatty acids; [3]Polyunsaturated fatty acids; [4]Desirable fatty acids = total unsaturated fatty acids + stearic acid; [5]Ratio of polyunsaturated to saturated fatty acids; [6]Total omega-6 fatty acids; [7]Total omega-3 fatty acids; [8]Ratio of omega-6 to omega-3 fatty acids.

MUFA in Diet 2 (26.47 g/100 g). Diet 2 had less PUFA than Diet 1 with CLA (C18:2n-6) being the dominant PUFA (32.29 g/100 g). α-Linolenic acid (C18:3n-3) was the only other PUFA in Diet 1 and 2 with a concentration above 1 g/100 g (3.67 and 8.40 g/100 g, respectively). The higher concentration of α-linolenic acid reported for Diet 2 is due the inclusion of linseed meal in the diet, since linseed is rich in this particular fatty acid (Demirel *et al.*, 2004; Raes *et al.*, 2004b).

The fatty acid composition of the *M. longissimus dorsi* and *M. biceps femoris* of the lambs are presented in Table 4. Results obtained in this investigation indicated that supplementing a diet with rumen-protected conjugated linoleic acid (CLA) (Diet 2) had a significant effect on the fatty acid composition of lamb *M. longissimus dorsi* and *M. biceps femoris*. Palmitic (C16:0), stearic (C18:0) and oleic (C18:1n-9) acid comprised the greatest proportions of the fatty acids in both muscles, as also reported by Webb *et al.* (1997) and Hoffman *et al.* (2003) for SAMM wethers and South African Merino sheep, respectively. This is also consistent with the results obtained by Rowe *et al.* (1999) who examined the effect of different types of diets on the fatty acid composition of adipose depots of lambs. Other fatty acids with proportions above 1g/100 g are the SFA C14:0 (myristic), the MUFA C16:1n-7 (palmitoleic) and the PUFA's C18:2n-6 (CLA) and C22:5n-3 (docosapentaenoic, DPA). Eicosatrienoic acid (C20:3n-3) and docosahexaenoic acid (C22:6n-3, DHA) was also prominent in the *M. biceps femoris* and the *M. longissimus dorsi*, respectively.

The palmitic acid (C16:0) concentration in the intramuscular fat of the *M. longissimus dorsi* and *M. biceps femoris* was not affected ($P > 0.05$) by feeding regimen. Although the dietary C16:0 concentration was higher in Diet 2 than in Diet 1 (19.13 *vs* 13.77 g/100 g), the C16:0 concentration in both muscles was not significantly different between lambs fed Diet 1 and 2. This finding coincides with that of Rhee *et al.* (2000) and Sheridan (2001). Sheridan (2001) stated that animals on a diet containing less palmitic acid, may synthesize it by hydrogenation and breakdown of chained fatty acids. However, the excess C16:0 in Diet 2 was probably desaturated to palmitoleic acid (C16:1n-7) or desaturated and elongated to C18:1n-9. A review by Chilliard (1993) indicated that fatty acid synthesis in ruminants is more sensitive to inhibition by saturated fatty acids. In other words, the *de novo* synthesis of C16:0 could have been less in lambs fed Diet 2 than lambs fed Diet 1, because Diet 2 provided much more exogenous C16:0, which is inhibitory to the *de novo* synthesis of C16:0, than Diet 1. The stearic acid (C18:0) concentration was also not influenced by diet. The same trend that was evident with C16:0, could be seen in the C18:0 concentration, since Diet 1 contained less stearic acid than Diet 2 (3.24 *vs* 11.11 g/100 g). Unlike C16:0, C18:0 may be produced by

Table 4 Fatty acid composition (g/100 g of identified fatty acids) of the *M. longissimus dorsi* and *M. biceps femoris* of the SAMM lambs fed the two different diets

	M. longissimus dorsi				*M. biceps femoris*			
Fatty acid	Baseline[1]	Diet1 (Control)	Diet 2 (CLA)	SEM	Baseline	Diet 1(Control)	Diet 2 (CLA)	SEM
14:0	2.017	2.684	2.689	0.218	1.625	1.517	1.776	0.147
16:0	22.939	26.721	26.807	0.650	23.552	25.650	26.005	0.515
18:0	18.931^{a}	15.689^{b}	17.674^{ab}	0.497	19.465^{a}	16.065^{b}	17.277^{b}	0.400
20:0	0.218^{a}	0.103^{b}	0.129^{b}	0.017	0.219^{a}	0.136^{b}	0.120^{b}	0.014
22:0	0.115^{a}	0.064^{b}	0.056^{b}	0.008	0.199^{a}	0.101^{b}	0.065^{b}	0.021
24:0	1.078^{a}	0.831^{ab}	0.489^{b}	0.086	1.013	0.508	0.549	0.114
14:1	0.105	0.101	0.100	0.006	0.130	0.103	0.085	0.009
16:1n-7	1.341^{b}	1.763^{a}	1.586^{ab}	0.065	1.377^{b}	1.820^{a}	1.643^{ab}	0.068
18:1n-9	34.454^{b}	39.618^{a}	37.679^{a}	0.692	34.847^{c}	42.255^{a}	38.976^{b}	0.840
20:1n-9	0.129	0.098	0.089	0.008	0.140	0.113	0.107	0.008
22:1n-9	0.092	0.133	0.098	0.037	0.113^{a}	0.074^{b}	0.065^{b}	0.008
24:1n-9	0.317^{a}	0.414^{a}	0.138^{b}	0.065	0.294	0.205	0.189	0.026
18:2n-6	6.813^{a}	5.002^{b}	6.345^{a}	0.269	8.655	6.610	7.914	0.350
18:3n-6	0.100^{a}	0.041^{b}	0.051^{b}	0.009	0.154	0.081	0.097	0.013
18:3n-3	1.379^{a}	0.498^{c}	0.941^{b}	0.086	1.258^{a}	0.613^{b}	0.977^{ab}	0.092
20:2n-6	0.258	0.085	0.082	0.046	0.326	0.095	0.131	0.046
20:3n-6	0.440^{a}	0.170^{b}	0.207^{b}	0.040	0.416^{a}	0.273^{b}	0.262^{b}	0.024
20:3n-3	2.584^{a}	0.833^{b}	1.187^{b}	0.293	2.498^{a}	1.464^{b}	1.509^{b}	0.137

Table 4 Fatty acid composition (g/100 g of identified fatty acids) of the *M. longissimus dorsi* and *M. biceps femoris* of the SAMM lambs fed the two different diets (continue)

	M. longissimus dorsi				*M. biceps femoris*			
Fatty acid	Baseline[1]	Diet1 (Control)	Diet 2 (CLA)	SEM	Baseline	Diet 1(Control)	Diet 2 (CLA)	SEM
20:4n-6	0.099[a]	0.059[b]	0.058[b]	0.007	0.135[a]	0.062b	0.087[b]	0.011
20:5n-3	1.187[a]	0.186[b]	0.673[ab]	0.153	0.736[a]	0.310[b]	0.388[b]	0.059
22:2n-6	0.203	0.178	0.316	0.056	0.567	0.163	0.177	0.107
22:5n-3	3.270	3.124	2.013	0.532	1.505	1.585	1.224	0.289
22:6n-3	1.932	1.603	0.594	0.441	0.776	0.198	0.377	0.138
SFA[2]	45.299	46.092	47.842	0.669	46.073	43.977	45.791	0.503
MUFA[3]	36.437[b]	42.128[a]	39.691[a]	0.698	36.902[c]	44.570[a]	41.065[b]	0.845
PUFA[4]	18.264[a]	11.780[b]	12.466[b]	1.119	17.025[a]	11.452[b]	13.143[b]	0.812
DFA[5]	73.632	69.597	69.831	0.768	73.392	72.088	71.486	0.562
PUFA:SFA[6]	0.414[a]	0.265[b]	0.263[b]	0.030	0.371	0.265	0.289	0.019
Σn-6[7]	7.913[a]	5.535[b]	7.058[a]	0.323	10.252[a]	7.284[b]	8.668[ab]	0.430
Σn-3[8]	10.351[a]	6.245[ab]	5.408[b]	0.948	6.773	4.168	4.475	0.486
n-6:n-3[9]	0.927	1.201	1.386	0.093	1.698	1.924	2.297	0.171

[a,b,c] Means in the same row, within a muscle, with different superscripts differ ($P \leq 0.05$).

[1]Initial Baseline group; [2]Saturated fatty acids; [3]Mono-unsaturated fatty acids; [4]Polyunsaturated fatty acids; [5]Desirable fatty acids = total unsaturated fatty acids + stearic acid; [6]Ratio of polyunsaturated to saturated fatty acids; [7]Total omega-6 fatty acids; [8]Total omega-3 fatty acids; [9]Ratio of omega-6 to omega-3 fatty acids.

elongation of C16:0 and/or through ruminal hydrogenation of 18-carbon unsaturated fatty acids (Ekeren *et al.*, 1992).

The two diets had no effect on the total and individual SFA, and total PUFA and DFA concentrations ($P > 0.05$) in both muscles. Diet had an effect on the total MUFA in both muscles, although only significantly for the *M. biceps femoris.* Dietary CLA was reported to increase the levels of SFA and decrease MUFA concentration in milk (Kelly *et al.*, 1998) and breast muscles from broilers (Sirri *et al.*, 2003) through inhibiting the activity of stearoyl CoA desaturase. The decreased concentration of total MUFA in both muscles, although only significantly for the *M. biceps femoris,* might be due to inclusion of the rumen-protected CLA in Diet 2. The intramuscular fat of both muscles contained a higher concentration of total SFA's and lower concentration of total PUFA's than were present in both diets. This may be due to the biohydrogenation of dietary unsaturated fatty acids by ruminal microorganisms (Byers & Schelling, 1993; Banskalieva *et al.*, 2000). Compared to Diet 1, the inclusion of linseed meal in Diet 2 may be responsible for the significant increase in the α-linolenic acid (C18:3n-3) concentration in the *M. longissimus dorsi.* Demirel *et al.* (2004) found that linseed doubled the proportion of C18:3n-3 in the *M. semimembranosus* of ram lambs, compared to lambs fed a diet containing Megalac®. Our observation is consistent with the findings of Raes *et al.* (2004b) for Belgian Blue young bulls fed a diet containing either crushed or extruded linseed. They also reported an increase in the total n-3 concentration of the *M. longissimus thoracis* which was not the case in the current study for either the *M. longissimus dorsi* or the *M. biceps femoris.* According to Scollan *et al.* (2003) the efficiency of incorporating dietary PUFA in the meat of ruminant animals is generally less than 5%, which relates to the high degree of biohydrogenation of the PUFA in the rumen.

There was an increase in CLA (C18:2n-6) deposition in both the *M. longissimus dorsi* and *M. biceps femoris* of the lambs fed Diet 2, although this increase was only significant for the *M. longissimus dorsi.* A possible explanation might be due to a larger variation among the fatty acid concentration measurements. This increased deposition occurred although Diet 1 contained a higher concentration of this particular fatty acid. This might be explained by the fact that the supplemental CLA in Diet 2 was rumen-protected, and demonstrate some resistance to ruminal biohydrogenation. Scollan *et al.* (2003) also reported a significant increased concentration of C18:2n-6 in intramuscular fat of beef muscle fed a diet containing a rumen-protected lipid source. This source comprised soybeans, linseed as well as sunflower seed oils. Beaulieu *et al.* (2002) found that dietary soybean oil did not affect the CLA (C18:2n-6) content, the predominant fatty acid in soybean oil, of muscle tissue.

Although not significant, the total PUFA concentrations in both muscles were lower for Diet 1 than for Diet 2. Choi *et al.* (2000) found that in animals with a higher intramuscular fat content, the proportion of PUFA is lower. This agrees with results from the previous chapter (Chapter 5), where it was found that the intramuscular fat content of both muscles investigated, tended to be higher for Diet 1 than for Diet 2 ($P = 0.3964$). This is mainly due to the fact that a higher intramuscular fat content is accompanied with a higher proportion of triacylglycerols relative to phospholipids. Phospholipids are characterised by a high PUFA content while triacylglycerols are mainly composed of SFA and MUFA (Raes *et al.*, 2004a). As a result of the lower intramuscular fat content compared to that reported by Hoffman *et al.* (2003) for crossbred lambs, the PUFA:SFA ratio (P:S ratio) in the current study is relatively high. This ratio (≈ 0.26) is closer to the recommended minimum value of 0.45 (Warris, 2000) and much higher than the average value of 0.119 reported by Hoffman *et al.* (2003).

The P:S ratio is an important guideline illustrating the total impact of SFA on blood cholesterol. This ratio is lower in ruminant than non-ruminant meat because of the biohydrogenation of dietary unsaturated fatty acids by ruminal microorganisms (Byers & Schelling, 1993; Banskalieva *et al.*, 2000). This is the main factor causing ruminant meat to have a P:S ratio below the value of 0.45 required in the human diet (Warris, 2000). However, De Smet *et al.* (2004) made the statement that for ruminants the P:S ratio is mainly influenced by genetics, in particular the overall fat level of the animal, and to a much lesser extent by the diet.

Desirable fatty acids (DFA), according to the health classification of Rhee (1992), are the sum of all unsaturated fatty acids and stearic acid (C18:0). Palmitic (C16:0), stearic (C18:0) and oleic (C18:1n-9) acids, as mentioned before, represented the majority of the fatty acids measured in both muscles. It is a well-known fact that C16:0 increases the lipid levels in blood, and thus increases total serum cholesterol levels, whereas C18:0 has no effect. C18:1n-9 lowers lipaemia by reducing both the low-density lipoprotein (LDL) cholesterol and triglycerides content of the blood, without reducing the high-density lipoprotein (HDL) cholesterol (Grundy, 1997; Russo *et al.*, 1999).

It was observed in the current study that lambs fed the diet that contained the highest n-6:n-3 ratio (Diet 1), tended to have the lowest n-6:n-3 ratio in the intramuscular fat of both muscles. Rizzi *et al.* (2002) reported the same tendency in intramuscular fat of hind leg muscles from male Sarda lambs. The extent to which dietary unsaturated fatty acids escape hydrogenation appears to depend on microbial growth condition that influences rate of lipolysis and biohydrogenation (Rizzi *et al.*, 2002). In the current study, addition of partially crushed,

formaldehyde treated or whole linseed, instead of linseed meal, could be a better strategy for delivery of unsaturated fatty acids. According to Bas & Morand-Fehr (2000), there is a considerable protection against hydrogenation of intracellular triacylglycerols in the rumen when the feed is not crushed or in a ground form, and a part of α-linolenic acid (C18:3n-3) would thus resist hydrogenation.

The n-6:n-3 ratio of intramuscular fat is highly influenced by the fatty acid composition of the diet fed to the animals. Including n-3 sources in the diet of animals increases the total n-3 content, which is mostly associated with decreased deposition of intramuscular n-6 fatty acids, and lowers the n-6:n-3 ratio (Raes *et al.,* 2004a). Scollan *et al.* (2001) also found that with the inclusion of a source rich in linolenic acid (bruised whole linseed), the concentration of linolenic acid in muscle of beef cattle was increased by 100%. This was not the case in the current study, as the total n-3 fatty acid content was not significantly increased in intramuscular fat of both muscles from lambs consuming Diet 2. Doreau & Ferlay (1994) stated that the increase of intramuscular C18:3n-3 content is limited. In ruminants the degree of biohydrogenation of linoleic acid (C18:2n-6) and C18:3n-3 was estimated at 80 and 92%, respectively, thus lowering the duodenal availability of C18:3n-3 relatively more than C18:2n-6 compared to their dietary levels (Doreau & Ferlay, 1994). According to nutritional guidelines, a n-6:n-3 ratio of less than 4 is recommended (Enser *et al.,* 1998). The ratios of intramuscular fat obtained in this study for both muscles, is well below the upper limit of the nutritional recommended ratio. Finishing ruminants on pasture can decrease the n-6:n-3 ratio, while concentrate (grains are normally high in C18:2n-6) fed ruminants give higher ratios around 6-10 (Enser *et al.,* 1998). This can be explained by the fact that grass has higher n-3 PUFA's, primarily as C18:3n-3, than grain- based diets, which are normally high in C18:2n-6 (Moloney *et al.,* 2001).

According to Enser (1995) grazing in lambs may increase the C18:0 concentrations in adipose tissue, while grain concentrates may increase C18:1n-9 and C18:2n-6. Westerling & Hedrick (1979) reported that the C16:0 percentage in intramuscular fat was similar between animals fed an 82.4% maize diet and those grazed on predominantly fescue grass, whereas the C18:1n-9 percentage was greater in grain-fed animals and C18:0, C18:2n-6 and C18:3n-3 percentage were higher in forage-fed animals. In the study by Marmer *et al.* (1984), however, the C18:2n-6 percentage of intramuscular fat was higher in animals fed a 79% corn diet than in animals fed a forage diet. These differences may be related to differences in fatty acid composition that could have been present between the two grain or two forage diets.

The effect of dietary treatment on fatty acid composition of subcutaneous, kidney and caul, and intermuscular adipose tissue are presented in Table 5. Fatty acids that were most represented in these three different adipose tissue types, were the same as the ones found for the intramuscular fat of both muscles. Inconsistent with our study, Webb & Casey (1995) found that C18:0 was the most abundant saturated fatty acid in the subcutaneous adipose tissue of SAMM lambs, followed by C16:0. On average between the two treatments, 48.02 g/100 g of the fatty acids in the subcutaneous adipose tissue were saturated, while 45.08 g/100 g were mono-unsaturated and 6.90 g/100 g polyunsaturated. These values coincides with those reported by Russo *et al.* (1999) for subcutaneous fat of Apennine male lambs, but not with those values found by Sheridan *et al.* (2003) and Cloete *et al.* (2004) for SAMM lambs. C18:1n-9 was the most abundant mono-unsaturated fatty acid and C18:2n-6 (CLA) the most abundant polyunsaturated fatty acid in the three adipose tissues types. The total SFA concentration in the subcutaneous fat was significantly higher and total MUFA concentration lower ($P \leq 0.05$) for Diet 2, compared to Diet 1. Kelly *et al.* (1998) and Sirri *et al.* (2003) reported the same effect for milk and breast muscles from broilers, respectively.

Webb *et al.* (1994) and Webb & Casey (1995) reported that the proportion of fatty acids in the subcutaneous adipose tissue of the later maturing SAMM was not affected by slaughter weight. The latter authors found that the proportion of saturated fatty acids in the subcutaneous adipose tissue tended to decrease with increasing live weight. Effect of slaughter weight on the proportion of mono-unsaturated or polyunsaturated subcutaneous fatty acids was negligible. Banskalieva (1996), on the contrary, found that with increasing age and live weight of lambs, the C14:0 and C16:0 content of the subcutaneous fat decreased ($P \leq 0.05$), while those of C18:0 and C18:1n-9 increased considerably ($P \leq 0.05$). Inconsistent with Banskalieva (1996), results obtained in the current study demonstrate that with increased age and slaughter weight (Initial baseline group *vs* Diet 1 and 2), the C14:0 concentration did not differ ($P > 0.05$), while the C16:0 concentration of both the subcutaneous and kidney and caul fat increased significantly. In the current study, the concentration of C18:0 in the subcutaneous fat increased significantly with age and slaughter weight, which is also inconsistent with that reported by Banskalieva (1996).

In studies by Webb *et al.* (1994) and Banskalieva (1996), it was found that a high energy diet significantly increased the deposition of C18:1n-9 in the subcutaneous fat of SAMM wethers and male lambs, respectively. A similar increase in the concentration of this particular fatty acid was observed by Casey *et al.* (1988). In the current study, the significantly higher concentration of C18:1n-9 found in the subcutaneous fat of the lambs, might also be due to

the high energy diet fed to the lambs (Webb *et al.*, 1994). In Chapter 5, the metabolizable energy (ME) value obtained for Diet 1 in the digestibility study differed from that calculated from the nutritive values of the feeds (Table 1) (Diet 1: 11.27 vs. 10.66 MJ/kg DM), causing Diet 1 to have a higher ($P \leq 0.05$) ME-content than Diet 2. As mentioned earlier, feeding a diet containing linseed meal to finishing lambs resulted in a significant increased deposition of α-linolenic acid (C18:3n-3) in the subcutaneous, intermuscular and kidney and caul fat. Webb *et al.* (1994), on the other hand, found that a greater concentration of C18:3n-3 were deposited in the subcutaneous fat of wethers fed a moderately high energy (10.2 MJ ME/kg DM) diet.

The cholesterol content of both the *longissimus dorsi* (62.32 and 68.83 mg/100g, respectively for Diet 1 and 2) and *biceps femoris* (80.99 and 85.17 mg/100g, respectively for Diet 1 and 2) muscles was not significantly influenced by the inclusion of rumen-protected CLA in the diet (Diet 2). Rowe *et al.* (1999) reported a cholesterol value of 57.76 mg/100g in *longissimus dorsi* muscles of drylot lambs slaughtered when their body weights ranged from 29-31 kg. Lee *et al.* (1994) reported that CLA significantly reduced serum triglycerides and low-density lipoprotein (LDL) cholesterol levels in rabbits, thus reducing atherosclerotic plaque formation in their aortas. In human adipose tissue it was found that 80% C18:2 *cis*-9, *trans*-11 and 20% C18:2 *trans*-10, *cis*-12 CLA supplement significantly reduced very low-density lipoprotein cholesterol concentrations (Rainer & Heiss, 2004).

Table 5 Fatty acid composition (g/100 g of identified fatty acids) of the three adipose tissue types of the SAMM lambs fed the two different diets, as well as that of the Initial baseline group

	Subcutaneous fat				Kidney and caul fat				Intermuscular fat		
		Diet 1	Diet 2			Diet 1	Diet 2		Diet 1	Diet 2	
Fatty acids	Baseline[1]	Control	CLA	SEM	Baseline	Control	CLA	SEM	Control	CLA	SEM
14:0	2.921	4.033	4.450	0.400	3.913	3.903	3.851	0.275	5.271	4.940	0.473
16:0	25.835^{b}	28.949^{a}	30.152^{a}	0.634	22.465^{b}	28.634^{a}	26.807^{a}	0.684	28.270	27.773	0.638
18:0	20.179^{a}	10.526^{b}	13.830^{b}	1.089	27.794	28.241	31.490	0.834	21.310	23.329	0.958
20:0	0.265^{a}	0.134^{b}	0.180^{ab}	0.021	0.340^{a}	0.239^{b}	0.232^{b}	0.019	0.142	0.180	0.014
22:0	0.226	0.130	0.257	0.054	0.466^{a}	0.585^{a}	0.094^{b}	0.196	0.047	0.067	0.006
24:0	1.020	1.289	2.107	0.407	1.281	0.786	0.120	0.271	0.209	0.227	0.077
14:1	0.144^{c}	0.791^{a}	0.426^{b}	0.066	0.078	0.083	0.057	0.009	0.151	0.139	0.014
16:1	1.292^{b}	2.376^{a}	1.919^{ab}	0.172	0.889	0.941	0.880	0.058	1.715	1.326	0.115
18:1n-9	40.717^{ab}	44.580^{a}	38.856^{b}	0.913	35.024^{a}	29.582^{b}	27.382^{b}	0.846	35.975	34.782	0.961
20:1n-9	0.146	0.159	0.116	0.007	0.150	0.119	0.132	0.012	0.099	0.102	0.005
22:1n-9	0.028	0.103	0.048	0.023	0.110	0.023	0.026	0.030	0.017	0.019	0.002
24:1n-9	0.596	0.303	0.480	0.140	0.113	0.354	0.363	0.062	0.238	0.156	0.046
18:2n-6	3.338	4.463	4.452	0.501	4.013	4.867	6.040	0.531	4.903	4.351	0.306
18:3n-6	0.038	0.048	0.041	0.006	0.031	0.036	0.030	0.004	0.023	0.023	0.001
18:3n-3	1.104^{a}	0.593^{b}	1.150^{a}	0.077	1.113^{a}	0.656^{b}	1.310^{a}	0.083	0.879^{b}	1.407^{a}	0.092
20:2n-6	0.054	0.043	0.062	0.008	0.059	0.051	0.064	0.008	0.042	0.054	0.007
20:3n-6	0.115	0.137	0.140	0.019	0.085	0.069	0.097	0.009	0.073	0.051	0.007

Table 5 Fatty acid composition (g/100 g of identified fatty acids) of the three adipose tissue types of the SAMM lambs fed the two different diets, as well as that of the Initial baseline group (continue)

	Subcutaneous fat				Kidney and caul fat				Intermuscular fat		
		Diet 1	Diet 2			Diet 1	Diet 2		Diet 1	Diet 2	
Fatty acids	Baseline[1]	Control	CLA	SEM	Baseline	Control	CLA	SEM	Control	CLA	SEM
20:3n-3	0.428	0.371	0.323	0.037	0.229	0.175	0.151	0.021	0.155	0.114	0.012
20:4n-6	0.059	0.089	0.082	0.015	0.092	0.040	0.052	0.011	0.029	0.075	0.016
20:5n-3	0.172[a]	0.078[b]	0.114[ab]	0.015	0.290[a]	0.034[b]	0.043[b]	0.057	0.057	0.052	0.008
22:2n-6	0.130	0.214	0.056	0.037	0.488[a]	0.035[b]	0.055[ab]	0.095	0.033	0.121	0.028
22:5n-3	0.527	0.254	0.414	0.091	0.588	0.336	0.477	0.118	0.186	0.455	0.106
22:6n-3	0.664	0.337	0.345	0.061	0.388	0.210	0.247	0.042	0.177	0.258	0.040
SFA[2]	50.445[a]	45.060[b]	50.976[a]	1.001	56.259[b]	62.389[a]	62.594[a]	0.900	55.249	56.516	0.874
MUFA[3]	42.924[b]	48.313[a]	41.845[b]	0.974	36.364[a]	31.102[b]	28.840[b]	0.837	38.194	36.524	0.913
PUFA[4]	6.630	6.626	7.179	0.526	7.377	6.509	8.566	0.560	6.557	6.960	0.363
DFA[5]	69.734[a]	65.466[b]	62.854[b]	0.925	71.535[a]	65.852[b]	68.896[ab]	0.787	66.061	66.813	0.980
P:S[6]	0.132	0.153	0.143	0.012	0.136	0.104	0.138	0.011	0.120	0.123	0.007
Σn-6	3.734	4.994	4.833	0.513	4.768	5.098	6.338	0.563	5.103	4.676	0.301
Σn-3	2.896[a]	1.633[b]	2.346[ab]	0.182	2.608[a]	1.411[b]	2.228[ab]	0.198	1.454[b]	2.285[a]	0.174
n-6:n-3	1.488	3.097	2.130	0.288	2.136	4.282	3.357	0.425	3.807[a]	2.113[b]	0.321

[a,b,c] Means in the same row, within a adipose tissue type, with different superscripts differ ($P \leq 0.05$).

[1]Initial Baseline group; [2]Saturated fatty acids; [3]Mono-unsaturated fatty acids; [4]Polyunsaturated fatty acids; [5]Desirable fatty acids = total unsaturated fatty acids + stearic acid; [6]Ratio of polyunsaturated to saturated fatty acids; [7]Total omega-6 fatty acids; [8]Total omega-3 fatty acids; [9]Ratio of omega-6 to omega-3 fatty acids.

Mean values for the sensory quality characteristics of the *M. longissimus dorsi* samples are presented in Table 6. Although all the sensory quality characteristics were similar ($P > 0.05$) for the two dietary treatments, the lambs fed Diet 1 tended to have the highest scores for aroma, tenderness and flavour, and the lowest for initial and sustained juiciness. When comparing the mean values for the attributes measured, it is clear that the samples from both treatments were perceived as being high in lamb aroma, high in both initial and sustained juiciness and very tender and flavourful, as all the mean values were above 80.

Koohmaraie *et al.* (1990) and Safari *et al.* (2001) reported that the most important contributing sensory attribute to eating quality is tenderness, with flavour and juiciness also contributing significantly, although to a lesser extent. Consumer surveys have also shown that tenderness is considered the most important component of meat quality (Young *et al.*, 1993). Results obtained by Devine *et al.* (1993) and Safari *et al.* (2001) indicated that a significant ($P < 0.001$), although negative, correlation exists between Warner-Bratzler shear force (WBSF) value and taste panel tenderness rating. In the current study the meat of lambs fed Diet 1 tended to be more tender than that of lambs fed Diet 2 ($P = 0.2224$) (Table 6). The corresponding WBSF values for lambs fed Diet 1 and 2, obtained from work done in Chapter 5, was 34.65 and 36.73 N/1.27 cm diameter ($P = 0.2995$), respectively. This is in accordance with the findings of Devine *et al.* (1993) that meat with a higher WBSF value will be less tender than meat with a lower WBSF value. It is well documented that WBSF value is more closely related to the myofibrillar than the connective tissue component of meat, especially when changes in strength of connective tissue are either small or similar (Bouton *et al.*, 1975). As the lambs in this investigation were of the same age, originated from the same flock (therefore raised under the same environment) it can be expected that they would have similar levels (and chemical form) of connective tissue. Any variation would therefore be due to inherent animal variation. According to Devine *et al.* (1993) the connective tissue contribution to toughness in lamb *M. longissimus dorsi* is relatively unimportant.

There is a great deal of controversy amongst researchers regarding the relationship between tenderness and the intramuscular fat content of the meat. Silva *et al.* (1999), for example, stated that intramuscular fat content is not correlated with tenderness. This is inconsistent with the negative correlation ($P < 0.001$) obtained by Seideman *et al.* (1987) and Okeudo & Moss (2005) between marbling fat and shear force of the *M. longissimus dorsi* in cattle and sheep, respectively. Schönfeldt *et al.* (1993) describe a study where a positive relationship was found between tenderness of meat and fatness of ovine carcasses. With reference to the findings of the latter authors, the fact that no significant difference was observed in tenderness between the two treatments in the current study, might be explained by the intramuscular fat

content of the two treatments (4.20 and 4.02% for Diet 1 and 2, respectively; Chapter 5), which did not differ significantly (P = 0.3964) from each other.

Dryden & Marchello (1970) reported that juiciness is related to both the capacity of the muscle to release its constitutive water (initial juiciness) and the infiltrated fat content (sustained juiciness). The melted lipid and water in the muscle constitutes a broth that, when retained in the meat, is released upon chewing. This broth may also stimulate the flow of saliva, and thus improve the meat's apparent juiciness (Forrest *et al.,* 1975). However, the major contributor to the sensation of juiciness is the water remaining in the cooked meat (Forrest *et al.,* 1975). Results from the previous chapter (Chapter 5) have shown that, although not significant, there was a tendency that the intramuscular fat content of the two treatments had a reverse relationship to the moisture content (r = -0.63, P = 0.0932; r = -0.45, P = 0.2592, respectively for Diet 1 and 2) in the *M. longissimus dorsi.* Muscles with the highest fat content were characterised by a lower moisture content. It was evident that the *M. longissimus dorsi* of lambs on Diet 1 tended (P = 0.3964) to have a higher fat content, and therefore a lower moisture content, compared to lambs on Diet 2. This might explain the tendency for Diet 1 to have a lower score for both initial and sustained juiciness than Diet 2. Schönfeldt *et al.* (1993) stated that the *M. longissimus thoracis et lumborum* cuts of sheep with a carcass fat classification of 1 contained significantly more expressible moisture than the corresponding cuts with a fat classification of 3. The sustained juiciness of the cuts with a carcass fat classification of 1 were therefore significantly ($P \leq 0.01$) higher in comparison to the corresponding cuts with a fat classification of 2 and 3. McKeith *et al.* (1985), on the contrary, found no relationship between water holding capacity values and intramuscular fat content.

It was evident from Chapter 5, that lambs fed the diet supplemented with rumen-protected conjugated linoleic acid (CLA) (Diet 2), exhibited a significantly lower cooking loss than of lambs fed Diet 1. Safari *et al.* (2001) stated that a negative correlation exists between cooking loss and juiciness, which will in turn affect the eating quality and acceptability of the meat. This might also explain the tendency for Diet 1 to have lower scores for juiciness, both initial and sustained, than Diet 2. Cooking loss and juiciness are significantly ($P < 0.001$) correlated with tenderness assessed by both WBSF and a sensory panel (Silva *et al.,* 1999; Okeudo & Moss, 2005). A reduction in cooking loss will lead to an increase in juiciness, and thus an increase in the perceived tenderness of the meat.

Aroma and flavour are important components of eating quality of all foods, and are sometimes used as determining criteria in the acceptance or rejection of the product,

especially in lamb and mutton meats (Mottram, 1992). The meaty flavours of cooked meat can be divided into two groups, those formed from lipid oxidation, particularly polyunsaturated fatty acids (PUFA), and those formed from the Maillard reaction (Mottram, 1998). In a literature summary of Owens & Gardner (1999), age, perhaps through increasing carcass fatness, was positively associated with flavour desirability. Although flavour intensity increased with increasing *longissimus* muscle lipid content, greater maturity of lean tissue reduced flavour intensity (Owens & Gardner, 1999). Aroma components are generally more soluble in fat and tend to be retained for longer in the fat matrix (De Roos, 1997). Aroma and flavour did not differ ($P > 0.05$) between treatments, and might be explained by the fact that the intramuscular fat content of the *M. longissimus dorsi*, as observed in Chapter 5, did not differ ($P > 0.05$) between Diet 1and 2. In a study by Young *et al.* (1993) a positive relationship was found between mutton aroma and flavour of the lean meat, as it is inextricably linked. In the current study, both these two attributes received similar scores from the descriptive panel.

Sañudo *et al.* (2000) reported consistent correlations between major fatty acids and sensory scores for aroma and flavour intensity. The correlation were positive for stearic acid (C18:0), oleic acid (C18:1n-9) and linolenic acid (C18:3n-3), and negative for linoleic acid (C18:2n-6). The n-6 and n-3 polyunsaturated fatty acids are important contributors to the aroma and flavour of ruminant meats fed grain or forage, respectively. The overall conclusion seems to be that the odours and flavours associated with feedlot-diet (grain-fed) meat are less intense than those from forage (grass), and are more preferred (Melton, 1990).

Table 6 Means for the sensory quality characteristics of the *M. longissimus dorsi* of the SAMM lambs as influenced by the two dietary treatments

Treatment	Aroma	Initial juiciness	Sustained juiciness	Tenderness	Flavour
Diet 1	85.08	86.75	85.13	84.40	85.56
Diet 2	84.87	87.38	85.38	82.10	84.68
LSD ($P \leq 0.05$)[1]	2.28	3.59	2.84	11.23	2.39

[1]LSD=Least significant difference ($P \leq 0.05$).

Conclusion

It was evident from the results obtained from this investigation that palmitic (C16:0), stearic (C18:0) and oleic (C18:1n-9) acid comprised the greatest proportions of the fatty acids,

measured on both the *longissimus dorsi* and *biceps femoris* muscles. The total and individual SFA, and total PUFA and DFA concentrations in both muscles was not affected ($P > 0.05$) by the inclusion of rumen-protected CLA, however the total MUFA concentration decreased in both muscles, although only significantly for the *M. biceps femoris.*

Although only significant for the *M. longissimus dorsi,* an increased CLA (C18:2n-6) deposition was observed in both the *M. longissimus dorsi* and *M. biceps femoris* of the lambs fed the diet containing rumen-protected CLA, resulting in healthier lamb. This increased deposition occurred although the Control diet contained a higher concentration of this particular fatty acid. A possible explanation for this increase might be due the fact that the supplemental CLA was rumen-protected, and demonstrate some degree of protection from the hydrogenating action of the rumen microorganisms.

The cholesterol content of both the *M. longissimus dorsi* and *M. biceps femoris* was not significantly influenced by the inclusion of rumen-protected CLA in the diet.

Inclusion of the rumen-protected CLA, measured on the *M. longissimus dorsi,* had no significant effect on any of the five sensory quality characteristics measured. From the results obtained it is clear that the samples from both treatments were perceived as being high in lamb aroma, high in both initial and sustained juiciness and very tender and flavourful, as all the mean values were above 80 on an unstructured 100 mm line scale.

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Chapter 7

GENERAL CONCLUSION

Sheep farmers in South Africa are faced with major constraining factors such as ever increasing production and feed costs, low product prices and a declining consumer demand for lamb and mutton. This results in profit margins becoming smaller and smaller. Thus, to survive economically, sheep farmers need to run their enterprises in the most effective way possible (Hoffman *et al.,* 2003). This unprecedented increase in the cost of conventional ingredients used in compounding livestock feed, and the decrease in availability has necessitated intensive investigations into the use of crop residues, such as maize cobs, which are largely wasted in the absence of a ruminant component in the system. In any livestock production system the key to success is to raise as many offspring as possible. Quite often lambs are raised on poor quality forage, resulting in higher mortality rates, slower growth rates and reduced reconception of the ewes in the following mating season. In such conditions lambs may have to be weaned earlier and finished in feedlots (Nolte & Ferreira, 2004).

Quality and cost of livestock production are determined to a great extent by quantity and composition of reserve fats (Banskalieva, 1996). Therefore, fatty acid composition plays an important role in the definition of meat quality, due to its relation to differences in sensory attributes, especially flavour (Melton, 1990), and in nutritional value of fat for human consumption. As lamb and mutton is partly composed of saturated fatty acids (Webb & Casey, 1995), the amount of fat in the human diet, and especially the proportion of saturated fatty acids have been considered as major risk factors for coronary heart diseases (CHD) (Kritchevsky, 1998). Recently, research has focused on a group of fatty acids characteristic of fat from ruminants, the conjugated linoleic acid (CLA), due to a wide range of positive health benefits in humans associated with it (Pariza *et al.,* 2001).

This project was conducted to obtain more information on the *in situ* rumen degradability of maize cobs, as well as the effect of incremental inclusion of maize cobs in feedlot finishing diets on the production efficiency and meat quality characteristics of South African Mutton Merino (SAMM) lambs. A second independent study was undertaken to determine whether rumen-protected conjugated linoleic acid (CLA) have a effect on production efficiency, proximate chemical composition and physical meat quality characteristics, as well as on fatty acid composition, cholesterol content and sensory attributes of SAMM lambs.

Results obtained from the *in situ* degradability study clearly established that, on average, the degradability of maize cobs is comparable with that of oat hay. In the first investigation, feed intake and feed conversion efficiency (FCE) were not affected ($P > 0.05$) by including maize cobs up to a level of 12% in the feedlot diets of the SAMM lambs at a growth rate of *ca.* 0.380 kg/day. The diets were formulated on an isonitrogenous and isoenergetic basis. There were no differences ($P > 0.05$) in the apparent digestibility of dry matter (DM), organic matter (OM), acid and neutral detergent fibre (ADF and NDF, respectively) or ether extract of the three diets between the lambs, suggesting that the diets had been digested with the same efficiency.

No significant differences between diets were observed in terms of energy retention, but lambs on the diet containing 4% maize cobs had a significantly higher nitrogen (N) retention, expressed as a percentage of nitrogen intake, than the lambs on the other two diets containing 8 and 12% maize cobs. The similar average daily gains obtained during the feeding trial might be due to the similar N retention of the lambs fed the three different diets (Stanford *et al.,* 1999).

Regarding the proximate chemical compositions of both the *M. longissimus dorsi* and *M. biceps femoris*, incremental inclusion of maize cobs did not significantly influence the moisture, ash, protein or fat content. Results obtained also indicated that feeding lambs the diet containing 4% maize cobs, produce a significantly darker (lower L* value) meat, with a lower b* value (yellowness) ($P \leq 0.05$) and consequently a narrower hue angle, than that of lambs fed the other two diets. Percentage cooking loss has been significantly affected, with lambs fed the diet containing 4% maize cobs, having the lowest cooking loss. However, whether these differences are of such a magnitude that they will influence either purchasing behaviour or consumption satisfaction of consumers requires elucidation.

No differences ($P > 0.05$) were observed as pertaining to ultimate pH and tenderness (Warner-Bratzler shear force, WBSF). It is a well-known fact that the ultimate pH of muscle is an important contributing factor to the quality of meat (Hoffman *et al.,* 2003) and according to literature conflicting reports regarding the relationship between pH and tenderness (WBSF) exist.

The results from the second study indicate that the inclusion of 2.5% rumen-protected linoleic acid (CLA) in feedlot finishing diets of SAMM lambs did not have any significant effect on their growth performance and feed conversion efficiency. Neither carcass weight nor dressing percentage was affected by the supplemental rumen-protected CLA. The lambs fed the supplemented diet, digested DM, OM and NDF digestibility less efficient than lambs fed the Control diet. The higher dry matter intake during the digestibility trial period, which lead

to increased passage rate, reduced rumen retention times, might explain the reduced DM and OM digestibility. When expressed as a percentage of energy intake, energy retention was significantly lower for lambs fed the CLA Diet, however, the nitrogen content of the two feedlot diets were metabolised with similar efficiency. The fact that no difference occurred in energy (MJ/kg) and nitrogen retention (g N/kg $BW^{0.75}$/day) explains the similar ADG for the lambs fed the two experimental diets.

Dietary treatments also had no effect ($P > 0.05$) on the proximate chemical composition of the two muscles, *M. longissimus dorsi* and *M. biceps femoris,* investigated. Percentage cooking loss differed ($P \leq 0.05$) between the two diets, and lambs fed the Control diet produced significantly lighter (higher L* value) meat with a higher b* value (yellowness) ($P \leq 0.05$) and consequently a wider hue angle.

Regarding the fatty acid composition, measured on both the *longissimus dorsi* and *biceps femoris* muscles, palmitic (C16:0), stearic (C18:0) and oleic (C18:1n-9) acid comprised the greatest proportions of the fatty acids, as also reported by Webb *et al.* (1997) and Hoffman *et al.* (2003) for SAMM wethers and South African Merino sheep, respectively. The inclusion of rumen-protected CLA had no effect on the total and individual SFA, and total PUFA and DFA concentrations ($P > 0.05$) in both muscles, however the total MUFA concentration decreased in both muscles, although only significantly for the *M. biceps femoris* (Kelly *et al.,* 1998; Sirri *et al.,* 2003).

An increased CLA (C18:2n-6) deposition was observed in both the *M. longissimus dorsi* and *M. biceps femoris* of the lambs fed the diet containing rumen-protected CLA, although this increase was only significant for the *M. longissimus dorsi.* This increased deposition occurred although the Control diet contained a higher concentration of this particular fatty acid. This might be explained by the fact that the supplemental CLA was rumen-protected, and demonstrate some resistance to ruminal biohydrogenation.
The cholesterol content of both the *M. longissimus dorsi* and *M. biceps femoris* was not significantly influenced by the inclusion of rumen-protected CLA in the diet.

Regarding the sensory attributes, measured on the *M. longissimus dorsi,* all the sensory quality characteristics were similar ($P > 0.05$) for the lambs fed the Control diet and the diet containing the rumen-protected CLA. Although no significant differences occurred, the lambs fed the Control diet tended to have the highest scores for aroma, tenderness and flavour, and the lowest for initial and sustained juiciness. When comparing the mean values

for the attributes measured, it is clear that the samples from both treatments were perceived as being high in lamb aroma, high in both initial and sustained juiciness and very tender and flavourful, as all the mean values were above 80 on an unstructured 100 mm line scale.

Based on the results obtained in this investigation, it may be inferred that the inclusion of maize cobs up to 12% in a feedlot finishing diet of lambs did not adversely affect the production efficiency, proximate chemical composition and physical meat quality characteristics of the animals. Maize cobs, therefore, is an excellent and cheap source of roughage for inclusion in diets of finishing lambs.

The inclusion of rumen-protected CLA led to increased CLA content in both muscle investigated, although not significant for the *biceps femoris* muscle. Further scientific investigations in determining the concentrations of the specific isomers of CLA will yield valuable information.

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