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Effect of the production system on lamb meat quality

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Abstract

The main aim of the thesis was to investigate the effects of (i) breed (Bergamasca, Italian Merino and Sopravissana), (ii) slaughter age (40 vs. 60 days) and (iii) rearing season (winter vs. autumn) on growth, slaughter traits and meat quality, including fatty acid (FA) composition of “Agnello del Centro Italia” PGI lambs produced under traditional transhumant sheep system adopted in Marche region (Central Italy).

The lambs of the studied breeds showed similar growth performances, carcass traits and meat quality (i.e., physical and chemical characteristics), despite some differences in the investigated parameters that occurred (e.g. % crude protein, cooking loss, TBARS and ORAC). Breed significantly affected FA profile of meat and adipose tissue with Bergamasca showing less favourable meat FA profile (the lowest values of conjugated linoleic acid and the highest thrombogenic index).

Higher slaughter age in Bergamasca lambs improved some carcass traits and physical characteristics of meat, it did not influence meat chemical composition or strongly affect meat FA profile, but it worsened chemical composition of adipose tissue. These results suggest 60 days as the optimal slaughter age for Bergamasca to produce slightly heavier carcasses without compromising the quality of lamb meat.

Different rearing seasons influenced carcass traits (pluck) and meat quality (pH and drip loss after 6 days of meat storage). FA profile of both meat and adipose tissue was influenced by the rearing season with lambs reared in autumn having adipose tissue with better FA profile for human health.

The research provides additional information filling the present gaps of knowledge regarding the quality of “Agnello del Centro Italia” light lambs produced under traditional systems in Marche region for local breeders and consumers.

Riassunto

Lo scopo principale di questa tesi è stato quello di indagare se gli effetti di: (i) razza (Bergamasca, Merinizzata Italiana e Sopravvissana); (ii) età di macellazione (40 vs 60 giorni) e (iii) stagione di allevamento (inverno vs autunno) abbiano influenzato le performance di allevamento, macellazione, qualità della carne e composizione di Acidi Grassi (FA) degli agnelli leggeri a marchio “Agnello del Centro Italia” IGP, allevati con sistema transumante nella regione Marche (Italia centrale).

Gli agnelli delle razze oggetto di studio hanno mostrato performance di crescita, resa in carcassa e qualità della carne (es. caratteristiche fisiche e chimiche) simili, nonostante si siano evidenziate differenze in alcuni parametri (peso spalle, % proteina, cooking loss, TBARS e ORAC). L'effetto razza ha influenzato significativamente il profilo acidico della carne e del tessuto adiposo ed alcuni parametri legati al valore nutrizionale della carne (PUFA/SFA e indice trombogenico).

Lo studio condotto per valutare l'effetto età di macellazione nella razza Bergamasca ha influenzato le caratteristiche quanti -qualitative della carcassa e della carne (drip loss, colore) e anche la composizione acidica del grasso.

Considerando sempre la razza Bergamasca, l'effetto stagione di macellazione ha influenzato le caratteristiche della carcassa (corata) e della carne (pH e drip loss a 6 d), come anche il profilo FA della carne e del tessuto adiposo.

Questo studio fornisce pertanto ulteriori informazioni sulla qualità di carcassa e carni di agnelli leggeri IGP “Agnello del centro Italia” ottenuti da razze differenti, allevati in stagioni diverse e macellati a differenti età e prodotti con sistema transumante nella regione Marche.

Table of contents

General Introduction.....	1
Preface	1
Aims and structure of the thesis	5
References	6
<u>Chapter 1: Effect of breed on the quality of "Agnello del Centro Italia"</u>	
<u>PGI light lambs</u>	
Effect of breed on quality the of "Agnello del Centro Italia" PGI light lambs: I. Growth, carcass and meat quality traits	10
Abstract	10
1. Introduction	11
2. Materials and methods.....	12
2.1. Experimental design, diet and animal management	13
2.2. Growth performance, slaughter procedure and carcass measurements	13
2.3. Meat quality parameters	14
2.4. Statistical analysis.....	16
3. Results and discussion.....	16
3.1. Growth, slaughter and carcass performances	16
3.2. Meat traits	18
4. Conclusions	24
References	25
Effect of breed on the quality of "Agnello del Centro Italia" PGI light lambs: II. Fatty acid composition of meat and adipose tissue	30
Abstract	30
1. Introduction	31

2. Materials and methods.....	32
2.1. Experimental design, diet and animal management	32
2.2. Slaughter and sampling	33
2.3. Determination of total fatty acid composition	33
2.4. Data analysis.....	34
3. Results and discussion.....	35
3.1. Multivariate analysis.....	35
3.2. Bivariate analysis.....	38
3.2.1. Saturated fatty acids.....	38
3.2.2. Unsaturated fatty acids	40
3.2.3. Trans and conjugated fatty acids	42
3.2.4. Nutritional indices	43
4. Conclusions	48
References	51
Slaughter performance, carcass and meat quality of Bergamasca light lambs as affected by slaughter age.....	58
Abstract	58
1. Introduction	59
2. Materials and methods.....	61
2.1. Experimental design, diet and animal management	61
2.2. Slaughter procedure and assessment of carcass traits.....	61
2.3. Meat quality parameters	62
2.4. Fatty acid analysis	63
2.5. Statistical analysis.....	63
3. Results and discussion.....	64
4. Conclusions	77

References	78
Effect of rearing season on growth, carcass traits and meat quality of Bergamasca light lambs	83
Abstract	83
1. Introduction	84
2. Materials and methods.....	85
2.1. Experimental design, diet and animal management	85
2.2. Slaughter procedure and assessment of carcass traits.....	86
2.3. Meat quality parameters	86
2.4. Fatty acid analysis	87
2.5. Statistical analysis.....	88
3. Results and discussion.....	88
4. Conclusions	100
References	101
General conclusions.....	106
References	108

General Introduction

Preface

Dry weather conditions, irregular topography and cereal based agricultural systems of the Mediterranean Basin make sheep a suitable species to take advantage of the natural resources in this area (Alfonso et al., 2001). Differences between sheep breeding in the Mediterranean and Northern Europe are based on the weather and ecological conditions. Rainfall is one of the main contributors (Sañudo et al., 1998) because it determines pasture land potential and subsequently the productive potential of animals, as well as many productive costs. The characteristics of the sheep breeds in the Mediterranean, compared with Northern countries in Europe, allow them to adapt to their environment perfectly and to transform crude products into top quality meat: rusticity, small or medium size, long reproductive activity and good milk production (Alfonso et al., 2001). In Mediterranean regions lamb meat market demands light carcasses (Sañudo et al., 1998; Santos et al., 2007) that are very specific and differ from most other regions (Santos-Silva et al., 2002). They are usually obtained from two production systems that are common for Mediterranean countries: (i) milk-meat with lambs slaughtered at ultra-light weights and in early ages, around one month (Sañudo et al., 1997); and (ii) meat with lambs slaughtered at light weights (between 40 and 60 days) (Alfonso et al., 2001; Santos-Silva et al., 2002; Juárez et al., 2009).

In Central Italy, a semi-extensive farming system is widely practiced to produce a traditional lamb (Panella and Di Felice, 1996) where rearing system is similar to those of the other regions of Mediterranean Basin. Pasture is used as long as possible for the sheep while lambs, permanently reared into the sheepfold, are fed on their mothers' milk during the whole production, receiving hay and concentrate supplements from 25-30 days to slaughter (Mazzone et al., 2010).

While in some regions of Italy (e.g. Sardinia) suckling lambs (carcasses up to 7 kg) are commonly produced by dairy breeds (Addis et al., 2013), in Central Italy light (7-13 kg carcasses) and heavy (over 13 kg) lambs from meat specialised breeds are mainly produced.

In Central Italy, lamb production is mainly based on extensive grazing systems (Caballero et al., 2009) and transhumance is still of major importance. Most of the flocks (up to 4000-5000 sheep) graze different grazing blocks throughout the year. During summer time, they use upland pastures and in autumn-winter period sheep are progressively transferred to lowlands. In Marche region, itinerant and vertical grazing, continuous stocking and permanent shepherding are common. Lucerne meadows are the main resource used, but the forage balance could include green cereals, crop residues, marginal lands and riverbanks itinerantly grazed (Caballero et al., 2009; D'Ottavio and Santilocchi, 2014).

Sheep meat production should meet quality requirements of industry and consumers, quality being the determining factor in the agro-food chain (Ramírez-Retamal et al., 2013). The concept of quality varies across the world as it is very subjective and defined by the consumer, depending on what flavours they are accustomed to, but also on lamb production system and other factors (Montossi et al., 2013). The quality of lamb carcass can be evaluated by considering weight, the level of fattening, conformation and proportion of different tissue components (lean, bone and fat). The lamb carcass classification system in the EU comprises two different schemes. Carcasses weighing more than 13 kg are evaluated according to conformation (E.U.R.O.P. classification: five classes, from E="good", to P="bad" conformation) and fatness score (5 classes, from 1=lean, to 5=fat). For carcasses weighing less than 13 kg, typical of the Mediterranean area, the conformation score is not considered since they are systematically penalised due to their naturally poor morphology. Thus, in the Mediterranean scheme, carcasses are divided into three categories according to weight (A:7.0 kg, B: 7.1–10.0 kg and C: 10.1–13.0 kg). Each weight category includes two quality classes: quality 1 carcasses have pink meat and a fatness score 2 or 3; quality 2 carcasses have red meat or fatness score 1 or 4 (EEC 2137/92 and 461/93 regulations). Carcass quality is not directly perceived by the consumer because lamb is normally consumed in cut or joints and not as a whole carcass. Therefore, meat quality can be determined more objectively through properties of the meat, such as pH, colour, chemical composition, sensory characteristics and others.

Nowadays, more attention is also paid to fatty acid composition given their importance for human health (Wood et al. 2003).

All these variables are influenced by great diversity of factors and may affect the quality of the product at both levels carcass and meat (Guerrero et al., 2013). According to Beriain et al. (2000) all those factors can be divided into production (biological and production system) and technological factors (slaughter and post-slaughter). While main biological factors such as breed, sex, productivity and susceptibility to stress are not influenced by human, the main factors of production such as environment, management, feeding, weight at slaughter, diseases can be controlled by the farmer and the technician on the farm. However, pre- and post-slaughter, marketing and consumption factors principally affect meat quality and they can be controlled just by the slaughterhouse, marketing chain and consumers. All these factors indicate that in order to obtain a quality product all the production-marketing consumption steps must be carefully attended. Farmers must be involved in the whole process as well as the next links in the product obtaining chain, helping in the achievement of the product tractability. In this sense, the PGI (Protected Geographical Indication) and PDO (Protected Designation of Origin) regulations are gaining importance as methods of promoting carcasses with specified characteristics (Beriain et al., 2000). Those are the best approaches to protect the products obtaining system that can be found as they control the rearing and production at the farm, the slaughtering and dressing procedures and carcass and meat characteristics. To receive the PGI status, the entire product must be traditionally and at least partially manufactured (prepared, processed or produced) within the specific region and thus acquire unique properties. Nowadays it is possible to find three lamb products with PGI quality labels in the Italian market such as “Agnello di Sardegna” PGI (entire area of the island of Sardinia), “Abbacchio Romano” PGI (Lazio region) and “Agnello del Centro Italia” PGI.

The Committee of different Central Italy sheep breeds have made huge effort to create a PGI label named “Agnello del Centro Italia” to promote and to protect local lamb meat market from foreign

competition (Pauselli et al., 2009) as imported lambs are usually sold at cheaper prices. The lamb of Central Italy joined the official list of typical Italian products with the certification “Agnello del Centro Italia” PGI in 2013 (European Union, Commission Regulation No. 475/2013). This label refers to lambs born and raised, through transhumance systems, in the entire territory of Central Italy. The geographical area of breeding the lambs comprises the territories of the following regions: Abruzzo, Lazio, Marche, Tuscany, Umbria and Emilia-Romagna limited to the entire territories of the provinces of Bologna, Rimini, Forlì-Cesena, Ravenna and, partially, to the territories of the provinces of Modena, Reggio Emilia and Parma. Lambs must be born and reared in the above mentioned geographical areas and from the following genetic types, breeds and their crossbreeds: Appenninica, Bergamasca, Biellese, Fabrianese, Italian Merino, Pomarancina, Sopravissana, Zerasca, Comisana, Cornella Bianca, Cornigliese (Corniglio), Garfagnina Bianca, Gentile di Puglia, Massese, Pagliarola, Pecora delle Langhe. These breeds are historically present in Central Italy, and their use, until the middle of the last century, was the triple production of meat, milk and wool and then – due to genetic improvement – many of them have been specialized for meat production. Lambs must be exclusively breastfed until weaning while, subsequently, feeding is based on dry or fresh fodder made up of wild plants from meadows and pastures, cultivated legumes or grasses and with small addition of grain when needed. Finally, for the needs of the market, there are three types of carcasses available: (i) ‘light lamb’ (weighing between 8 and 13 kg), (ii) ‘heavy lamb’ (weighing over 13 kg) and (iii) ‘castrate’ (weight greater than or equal to 20 kg).

“Agnello del Centro Italia” PGI lambs are much appreciated by consumers, both through the circuit of traditional butchers, either through food, especially that became part of the circuit of farmhouses. The product is mainly consumed at Christmas and Easter, as lamb meat is a very popular among Christians during those periods. However, there is an attempt to urge consumers to consider the purchase of “Agnello del Centro Italia” PGI during the other periods of year as well. In the past (in 1988), a campaign with that aim was launched and it had

the slogan “Agnello del Centro Italia, good throughout the year, not only for the holidays.”

Aims and structure of the thesis

Although its reputation and quality are well known among consumers, there is scarcity of published data regarding the quality of “Agnello del Centro Italia” PGI lambs, as well as factors affecting it.

In view of the above, the main aim of this PhD research is to investigate how different biological and production factors influence quality of “Agnello del Centro Italia” PGI light lambs produced under traditional transhumant sheep system adopted in Marche region (Central Italy).

More particular aims are to investigate the effect of:

- i) Breed (Bergamasca, Italian Merino and Sopravissana) on growth, slaughter traits and meat quality with accent on fatty acid composition of meat and adipose tissue.
- ii) Slaughter age (40 vs. 60 days) on slaughter traits and meat quality, including fatty acid composition of Bergamasca light lambs.
- iii) Season (winter vs. autumn) on growth, slaughter traits and meat quality, including fatty acid composition of Bergamasca light lambs.

The thesis contains two chapters, each one composed of two papers written as stand-alone manuscripts to be published in international peer-reviewed journals. Therefore, each of these papers is divided into the subsections: abstract, introduction, materials and methods, results and discussion, conclusions and references.

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Chapter 1

Effect of breed on the quality of “Agnello del Centro Italia” PGI light lambs

Effect of breed on the quality of “Agnello del Centro Italia” PGI light lambs: I. Growth, carcass and meat quality traits

Abstract

The aim of this study was to assess growth performances, slaughter characteristics, carcass and meat quality of Bergamasca (BG), Italian Merino (IM) and Sopravissana (SP) light lambs produced under the “Agnello del Centro Italia” PGI (Protected Geographical Indication) quality label. Effect of storage period (0, 3 and 6 days) on drip loss, colour and TBARS (2-thiobarbituric acid reactive substances) was also investigated. A total of 11 male single born lambs per breed reared on pasture with dams and supplemented with concentrate *ad libitum* from 20 days to slaughtering age (60 days) were used for this purpose. Average daily gain did not differ between breeds, and neither did carcass weight and yield. BG lambs showed higher proportion of full digestive tract. IM lambs had higher proportion of shoulder than SP, while BG had lower proportion of fat but higher proportion of bone in comparison with IM lambs. Other traits of the meat (pH, drip loss and colour) were not affected by breed while IM had higher cooking loss than SP lambs. Lambs differed in meat chemical composition with meat of SP having higher content of crude protein than BG. Antioxidant capacity was the highest for SP and the lowest for BG lambs. BG lambs showed higher lipid oxidation than both the other breeds. Colour, drip loss and TBARS were influenced by storage period.

Key words: light lamb, Bergamasca, Italian Merino, Sopravissana.

1. Introduction

In Mediterranean regions lamb meat market demands light carcasses (Sañudo et al., 1998; Santos et al., 2007) that are very specific and differ from most carcasses of the other regions (Santos-Silva et al., 2002). While in some regions of Italy suckling lambs (carcasses up to 7 kg) are commonly produced by dairy breeds (Oriani et al., 2005), in Central Italy light (7-13 kg carcasses) and heavy (over 13 kg) lambs from meat specialised breeds are mainly produced. Here they are very often raised with mothers on pastures as well as supplemented with concentrate and/or hay and weaned late, or not weaned at all.

Lambs produced in these systems are often not competitive in the global market due to their limited yield and high production costs (Addis et al., 2013). In this sense, the PGI (Protected Geographical Indication) and Protected Designation of Origin (PDO) regulations are gaining importance as methods of promoting production of carcasses with specified characteristics (Beriaín et al., 2000). With this regard, in Central Italy “Agnello del Centro Italia” PGI quality label (European Union, Commission Regulation No. 475/2013) was created to promote and to protect local lamb meat market from foreign competition (Pauselli et al., 2009) of imported lambs usually sold out at cheaper prices. According to “Agnello del Centro Italia” PGI product specifications, lambs produced by a long list of local breeds and cross breeds are admitted. Appenninica, Bergamasca, Fabrianese, Italian Merino and Sopravissana are some of those.

Considering the consumer perspective, concept of quality varies across the world, being very subjective and defined depending on what flavours they are accustomed to, but also on lamb production system and other factors (Montossi et al., 2013). Carcass quality can be evaluated objectively considering weight, the level of fattening, conformation and proportion of different tissue components (lean, bone and fat). As these parameters are not directly perceived by the consumer because lamb is normally consumed in cut or joints, more important properties for determination of meat quality are pH, chemical composition, drip loss and others. Colour is of utmost importance as it is the first impression consumers have of fresh meat and it can be

influenced by oxidation. Lipid oxidation is a major cause of deterioration in the quality of meat as it can occur in either refrigerated or frozen fresh meat (Love and Pearson, 1971) and lead to rapid development of undesirable flavour, texture, nutritional value and acceptability in meat (Buckley et al., 1989). All these variables are influenced by a great diversity of factors such as breed, diet, sex, age (Guerrero et al., 2013) and may affect the quality of the product at both levels- carcass and meat.

Although the breed effect on different productive performance and meat quality aspects of lambs has been widely studied for many breeds and production systems (e.g. D'Alessandro et al., 2013; Marino et al., 2008; Juárez et al., 2009; Sañudo et al., 1997; Vacca et al., 2008), published data on most of the breeds labelled as "Agnello del Centro Italia" PGI are mostly absent. In this sense, the aim of the present study was to assess growth performances, slaughter characteristics, carcass and meat quality of light lambs of three different breeds (Bergamasca, Italian Merino and Sopravissana) produced under the "Agnello del Centro Italia" PGI quality label. The effect of storage period (0, 3 and 6 days) on drip loss, colour and TBARS (2-thiobarbituric acid reactive substances) of meat was also investigated.

2. Materials and methods

The study was conducted under the usual conditions for rearing and management of the transhumant sheep system in Marche region (Central Italy) which is based on extensive grazing. In summer, most of the flocks graze upland pastures. Starting from autumn the sheep are progressively transferred to lowlands where up to the spring time the main forage resources are Lucerne meadows, but winter cereals at vegetative stage, crop residues, marginal lands and riverbanks are sometimes also used (Caballero et al., 2009; D'Ottavio and Santilocchi, 2014). Itinerant and vertical grazing, continuous stocking and permanent shepherding are common here. Mainly, lamb production is performed in lowlands for Easter and Christmas market. This study was performed in winter to produce lambs for Easter market.

The climate of the studied area is characterised by mean annual temperature of 13.7 °C and mean annual precipitation of 792 mm. The animal handling followed the recommendations of the European Union directive 2010/63/EU and Italian law (D. Lgs. n. 26/2014) regarding animal care.

2.1. Experimental design, diet and animal management

At lambing, among lambs of single parturition which occurred within a week, total of 11 male lambs per each breed (Bergamasca, Italian Merino and Sopravissana) were chosen and reared together with their dams. The flock (ewes and lambs of all the monitored breeds) grazed four alfalfa dominated pasture plots from mid-February to mid-April in 2015. Dams had free access to hay and were supplemented by corn grain (0.5 kg head⁻¹ day⁻¹) while lambs were given concentrate (50% corn, 50% barley) *ad libitum* in creep feeders from 20 days of age. All animals had free access to water. Feed samples were collected to determine their chemical composition (Table 1) according to Martillotti et al. (1987).

2.2. Growth performance, slaughter procedure and carcass measurements

In order to calculate average daily gain (ADG), individual lamb weights were recorded at birth and each 20 days until the slaughter performed at the average of 60 days of age. All lambs were transported on the same day to a commercial slaughterhouse, stunned and slaughtered by cutting the jugular vein. After slaughter, non-carcass components such as skin, head, feet, pluck (heart, lungs, liver and spleen), kidneys and digestive tract were removed, weighed and presented as proportions of pre-slaughter weight (PSW). Hot carcasses were weighed and left for chilling at 4 °C for 24 h. Dressing percentages were calculated as hot carcass weight/live weight. After chilling, each carcass was split along the vertebral column into left and right halves. Right half was then split to the three main commercial joints. The weight of each joint (shoulder, whole loin with flank and leg) was recorded and expressed as proportion of half carcass weight. From each right halve of carcass the

samples for analysis of ORAC and TBARS (two thoracic vertebrae) and steak made of muscle *longissimus dorsi* (LD) including bones between the 1st and 6th lumbar vertebrae were taken. The steak was dissected into lean, fat, and bone and afterwards used for further analysis.

Table 1

Dry matter (DM) percentage and chemical composition (% of DM) of the diet.

	Pasture	Hay	Corn grain	Concentrate
DM	21.51	88.31	83.52	85.89
Ashes	15.51	7.95	1.39	1.72
Crude protein	15.58	7.34	5.66	7.68
Ether extract	1.65	1.05	2.88	2.73
Crude fibre	21.46	31.28	2.65	2.94
NDF	39.65	63.45	16.30	21.99
ADF	30.11	41.02	3.03	4.90
ADL	6.37	6.15	0.76	1.52

2.3. Meat quality parameters

Carcass pH was measured 45 min and 24 h *post mortem*, in the *longissimus thoracis* muscle between the 10th and 13th thoracic vertebrae, with a penetrating glass electrode connected to a portable pH-meter (Eutech Instruments, mod. XS pH 110, Singapore).

After the removal from bone and external fat, LD muscle was divided into 3 slices. Two slices were immediately used for drip loss and cooking loss determination (ASPA, 1996) while colour measurements were performed on the last one.

In order to measure the drip loss, meat samples were weighed and wrapped in polyethylene bags. After 24 h storage period at 4 °C, the samples were gently dried with paper towels and reweighed. This procedure was done in two replications and repeated on the 3rd and the 6th day of storage.

For cooking loss determination, meat samples were firstly weighed, then placed in polyethylene bags and cooked in a water bath until reaching internal temperature of 65 °C. Bags with cooked samples were then cooled under cold running water for 30 minutes and then removed from their respective bags, dried with paper towels and reweighed.

Meat colour was measured on fresh meat (first measurement), after 3 days of storage (second measurement) and finally after 6 days of storage (third measurement). During the storage period, samples were vacuum packed and kept at 4 °C. Colour was evaluated after 40 min of blooming using the CIELAB colour space where L* (lightness), a* (redness) and b* (yellowness) values were obtained using a Minolta CR 200 with illuminant D65 as the light source. Chroma (C*, the square root of (a*² + b*²)) and hue angle (H°, tan⁻¹(b*/a*)) were also calculated where final conversion of hue from radians to degrees was achieved by multiplying tan⁻¹(b*/a*) by 180/p (Liu et al., 1996).

In order to monitor meat colour changes over storage, total colour change (ΔE^* , $[(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]$) was calculated where Δ represents difference between colour parameters measured in different days of storage (Iacurto, 2005). On each meat sample, three colour measurements and calculations were performed for each parameter and reported as mean value.

After colour measurements were performed, meat samples were dried by lyophilisation (dry matter), ground (1 mm screen) and analysed for chemical determination of crude protein (Kjeldahl nitrogen x 6.25), fat (extraction with petroleum ether) and ash (incineration in muffle furnace at 550 °C) content. Analyses were performed in duplicate for each sample and investigated parameter, and results corrected for moisture content.

The antioxidant capacity of meat was determined using the oxygen radical absorbance capacity method (ORAC_{FL}) as described by Branciarri et al. (2015). The data are expressed as micromoles of Trolox equivalents (TE) per gram of sample ($\mu\text{mol TE g}^{-1}$).

Lipid oxidation was measured on fresh meat and after the 3rd and the 6th day of storage by TBARS (2-thiobarbituric acid reactive substances) method described by Tarladgis et al. (1960). During the storage period, meat samples were kept in polyethylene bags at 4 °C. Analyses were performed using spectrophotometer with 538 nm (Ultrospec 2100 pro, Amersham pharmacia biotech, Cambridge, UK) and results expressed as mg MDA (malondialdehyde) kg⁻¹ muscle.

2.4. Statistical analysis

A one-way analysis of variance (ANOVA) was performed to test the effect of breed or storage days, followed by the Tukey's HSD post-hoc comparison means test in case of significance. For effect of breed, storage days, and their interaction on drip loss and TBARS, a two-way ANOVA was also performed. All statistical analyses were performed by JMP[®] software (JMP, Version 10. SAS Institute Inc., Cary, NC, 1989-2007).

3. Results and discussion

3.1. Growth, slaughter and carcass performances

Data on growth performances presented in Table 2 indicate that no significant differences were found between breeds, both for live weight (LW) and ADG, although ADG tended to be greater for Italian Merino during the last 20 days of growing period. Lambs of all the breeds had higher birth weight, as well as LW and ADG than those reported by ASSONAPA (2015) as the standard breed characteristics. ADGs were also higher than those reported by Santos-Silva et al. (2002) for Merino Branco and crossbred Ile de France x Merino Branco light lambs, reared with their mothers on grass and with concentrate *ad libitum*.

Table 2
Growth performances of the lambs slaughtered at 60 days of age according to the breed (mean \pm SD).

	Bergamasca	It. Merino	Sopravissana
<i>Live Weight (kg)</i>			
At birth	6.30 \pm 1.15	5.50 \pm 0.89	5.90 \pm 0.92
20 days	13.32 \pm 2.30	11.80 \pm 2.31	12.70 \pm 1.52
40 days	19.47 \pm 3.66	16.91 \pm 3.96	18.55 \pm 2.34
60 days	26.07 \pm 4.16	24.71 \pm 5.29	25.22 \pm 2.38
<i>Average Daily Gain (g d⁻¹)</i>			
0-20 days	336 \pm 81.82	345 \pm 73.93	366 \pm 67.62
20-40 days	293 \pm 100.10	255 \pm 85.88	292 \pm 62.41
40-60 days	314 \pm 58.76	371 \pm 79.02	318 \pm 59.37
0-60 days	323 \pm 64.64	324 \pm 73.16	324 \pm 39.56

The effect of the breed on carcass weight and dressing percentages as well as on non-carcass components is presented in Table 3. No significant differences in carcass weight and dressing percentages between breeds were recorded. All three breeds had similar carcass weight and were classified as class C in the European carcass classification system for light lambs (10.1-13.0 kg; EU, 1994). The carcass weights were higher than those reported for light lambs by Cifuni et al. (2010) slaughtered at 45 days (9.3 kg) and similar to those slaughtered at 90 days of age (12.4 kg). Higher dressing percentages are reported for Apulian light lambs reared in similar conditions (Cifuni et al.; 2010) or Leccese light lambs (D'Alessandro et al., 2013) slaughtered at same age as lambs in the present study. Regarding non-carcass components, breed influenced proportion of the full digestive tract with Bergamasca having the highest ($P < 0.05$) proportion. As digestive tracts content was not removed before weighing, it cannot be concluded that there would be a statistical difference in the size or weight of the empty digestive tracts between breeds.

Table 3
Slaughtering performance of lambs slaughtered at 60 days of age according to the breed (mean \pm SD).

	Bergamasca	It. Merino	Sopravissana
Carcass weight (kg)	12.12 \pm 2.87	12.45 \pm 2.77	12.63 \pm 1.45
Dressing (%)	49.51 \pm 2.59	50.32 \pm 2.14	50.01 \pm 1.83
<i>Proportion (%) on PSW</i>			
Head	4.05 \pm 0.23	3.81 \pm 0.38	3.87 \pm 0.20
Skin	10.96 \pm 0.76	11.81 \pm 1.01	11.63 \pm 0.79
Feet	0.81 \pm 0.25	0.74 \pm 0.07	0.76 \pm 0.03
Pluck	6.19 \pm 1.33	6.83 \pm 0.55	6.76 \pm 0.57
Kidneys	0.29 \pm 0.06	0.32 \pm 0.06	0.30 \pm 0.04
Full digestive tract	15.78 ^a \pm 2.57	12.98 ^b \pm 1.79	13.81 ^{ab} \pm 1.97

PSW: pre-slaughter weight.

^{a, b} Within the same row, means with different letters differ significantly at $P < 0.05$.

Half carcass joints and tissue proportions results are shown in Table 4. No significant differences in half carcass weight, whole loin with flank and leg proportions were observed between the breeds while Italian Merino lambs had higher shoulder proportion ($P < 0.05$) than Sopravissana lambs. Obtained values for leg joint, presented as

proportion of half carcass weight, are comparable with those reported by Vacca et al. (2008) for Mouflon x Sarda and Sarda x Sarda suckling lambs slaughtered at 40 days of age.

Steak (from the 1st to 6th lumbar vertebrae) of all the breeds had similar tissue proportion of lean while Bergamasca breed had lower proportion of fat and higher proportion of bone ($P < 0.05$) compared to Italian Merino. Even lean/fat ratio was the highest for Bergamasca breed, significant differences between breeds for lean/fat or lean/bone ratio were not found. Comparison with other studies is limited as tissue proportion is usually obtained by dissection of one or more main commercial joints (e.g. Carrasco et al., 2009; D'Alessandro et al., 2013; Vacca et al., 2008).

Table 4

Half carcass joints and tissue proportions obtained by dissection of steak of lambs slaughtered at 60 days of age according to the breed (mean \pm SD).

	Bergamasca	It. Merino	Sopravissana
CHCW (kg)	6.28 \pm 1.43	6.49 \pm 1.41	6.60 \pm 0.74
<i>Proportions of joints (%)</i>			
Shoulder	40.05 ^{ab} \pm 1.62	40.87 ^a \pm 2.11	38.84 ^b \pm 1.38
Whole loin with flank	22.33 \pm 2.53	22.24 \pm 2.11	24.13 \pm 1.61
Leg	37.61 \pm 1.49	36.89 \pm 0.75	37.03 \pm 2.24
<i>Tissue proportion of steak (%)</i>			
Lean	61.28 \pm 2.90	61.56 \pm 4.63	61.78 \pm 4.06
Fat	7.78 ^b \pm 3.90	11.74 ^a \pm 3.57	10.33 ^{ab} \pm 2.79
Bone	30.27 ^a \pm 4.06	25.85 ^b \pm 3.53	26.99 ^{ab} \pm 2.34
Losses	0.67 \pm 0.36	0.85 \pm 0.26	0.90 \pm 0.38
Lean/Fat ratio	8.15 \pm 3.29	5.85 \pm 2.23	6.54 \pm 2.39
Lean/Bone ratio	2.07 \pm 0.36	2.44 \pm 0.53	2.31 \pm 0.31

CHCW: cold half carcass weight.

^{a, b} Within the same row, means with different letters differ significantly at $P < 0.05$.

3.2. Meat traits

The main factor determining the quality of meat is its pH, which is related to biochemical processes during the transformation of muscle to meat. Significant differences in carcass pH value (Table 5) were not found between breeds, although pH decreased during the storage period of carcasses as a result of lactic acid accumulation in post-mortem muscle (Nagaraj et al., 2006).

The final pH (24 h) ranged from 5.29 to 5.36, indicating that lambs were not stressed at the time of slaughter. The recorded pH values were acceptable and within the normal ranges, assuming that a final pH greater than 5.8 is regarded as undesirable (Tejeda et al., 2008). In the present study, carcass pH values after slaughter (pH 45') were below the range (6.15-6.80) reported by the “Agnello del Centro Italia” PGI product specifications.

Table 5
Carcass pH, drip loss and cooking loss of the meat of lambs slaughtered at 60 days of age according to the breed (mean \pm SD).

	Bergamasca	It. Merino	Sopravissana
<i>pH</i>			
45'	5.98 \pm 0.20	5.73 \pm 0.36	5.83 \pm 0.16
24 h	5.29 \pm 0.16	5.36 \pm 0.30	5.23 \pm 0.17
<i>Drip loss (%)</i>			
1 days	0.88 \pm 0.23	0.97 \pm 0.44	0.94 \pm 0.27
3 days	4.59 \pm 1.07	4.93 \pm 1.40	5.65 \pm 0.98
6 days	8.85 \pm 1.55	9.47 \pm 2.59	10.93 \pm 2.77
<i>Cooking loss (%)</i>	10.29 ^{ab} \pm 3.40	12.95 ^a \pm 3.92	8.36 ^b \pm 3.53

^{a, b} Within the same row, means with different letters differ significantly at $P < 0.05$.

Contrary, the final pH was inside the range (5.15-5.80) indicated by those specifications. Obtained pH (both 45' and 24 h) values were lower compared to those of light lambs of some Italian breeds such as Altamura (della Malva et al., 2016), Appenninica (Mazzone et al., 2010) and Leccese (D'Alessandro et al., 2013) slaughtered at similar age but at different weight.

No significant differences (Table 5) were found for the drip loss between breeds. When data were analysed with two way ANOVA with fixed effects of breed, storage time and their interactions (Table 9) differences in drip loss between Sopravissana and Bergamasca lambs (5.84 vs 4.90, respectively) emerged ($P < 0.05$). After one day of meat storage, Mazzone et al. (2010) reported higher values of drip loss (1.66 vs. 0.93%) than those obtained in this study for light lambs from similar rearing system.

As expected, a significant increase in drip loss of LD muscle ($P < 0.001$; Table 9) within 1, 3 and 6 days of storage was evident. According to Nagaraj et al. (2006) drip loss increase could be related to different

factors, such as protein denaturation, sarcomere shortening and myosin denaturation.

In the present study, Italian Merino had higher values for cooking loss compared to Sopravissana breed ($P < 0.05$; Table 5) and similar to those of Italian Merino Derived lambs slaughtered at similar age after raising on ewes' milk, organic hay and concentrate (Morbidini et al., 2001). Cooking loss values of Bergamasca, Italian Merino and Sopravissana lambs were lower than those reported for light lambs of Appenninica (Mazzone et al., 2010) and Rasa Aragonesa (Sañudo et al., 2000) breeds.

Table 6

Chemical composition of the *longissimus dorsi* muscle of lambs slaughtered at 60 days of age according to the breed (mean \pm SD).

	Bergamasca	It. Merino	Sopravissana
Moisture	75.08 \pm 1.31	74.39 \pm 1.17	73.70 \pm 1.70
Crude protein	19.31 ^b \pm 0.77	19.77 ^{ab} \pm 0.68	20.37 ^a \pm 1.09
Fat	1.56 \pm 0.66	1.97 \pm 0.55	1.93 \pm 0.57
Ash	1.22 \pm 0.05	1.26 \pm 0.05	1.23 \pm 0.05

^{a, b} Within the same row, means with different letters differ significantly at $P < 0.05$.

Considering the chemical composition of the meat, the influence of breed was statistically significant just for the crude protein content that was higher in Sopravissana compared to Bergamasca ($P < 0.05$; Table 6). In general, the values were found to be within the range reported by other authors (D'Alessandro et al., 2013; della Malva et al., 2016; Juárez et al., 2009; Marino et al., 2008; Vacca et al., 2008) for meat of light lambs of different breeds.

Meat colour is one of the main aspects affecting consumer's purchase decision, as it is used to evaluate its freshness and quality (Faustman and Cassens, 1990). No significant differences in any colour parameters were found between the breeds (Table 7). Differences between breeds in muscle colour depend essentially on the precocity of the breed during the development stage (Hajji et al., 2016). According to Hajji et al. (2016) no differences in muscle colour between the three studied breeds could be detected due to the same age and stage of maturity. In that regard, with animals of the same age significant differences in meat colour were also not found between Awassi and Morkaraman lambs

(Esenbuga et al., 2009) or between Rasa Aragonesa, Lacaune and German Merino light lambs (Sañudo et al., 1993).

Khliji et al. (2010) suggest that when the L^* value of the meat is equal to or exceeds 34, on average, consumer will consider meat colour acceptable. Furthermore, according to Hopkins (1996) and Khliji et al. (2010), a^* values may be more appropriate to assess consumer colour acceptability. When it is equal to or above 9.5, meat colour will be acceptable by consumers. In the present study, average L^* and a^* values for all three breeds indicated a light-coloured and acceptable meat. The chroma (C^*) and hue angles (H°) are both a good indicators of meat discoloration (Young and West, 2001) as they are more closely related to visual appearance. In the present study, values for C^* were much higher while values for H° were much lower than those reported by other authors (della Malva et al., 2016; Mazzone et al., 2010; Russo et al., 2003) for meat of light lambs of different breeds.

Storage period had no effect on lightness (L^*) and redness (a^*) of LD muscle (Table 7). The trend of L^* value is in accordance with the findings of Abdullah and Qudsieh (2009) while they found effect of storage on a^* value. In the present study, within the storage period significant differences ($P < 0.01$) occurred just in terms of yellowness (b^*) that increased in samples after 3 and 6 days of storage compared to fresh meat in Italian Merino and Sopravissana. In the case of Bergamasca, yellowness increased significantly ($P < 0.05$) just after 6 days of storage. Colour changes after storage were also recorded in terms of chroma (C^*) (Bergamasca and Sopravissana) and hue angle (H°) (all the breeds).

No significant changes were observed in terms of LD muscle total colour differences (ΔE^*). This parameter is used to understand if any change in colour between two different measurements is detected by human eye. Even though no statistical differences between storage periods were recorded and colour variations in few days can be little distinguishable, it can be stated that such variations are clear to a human eye from 0 to 6 days of storage (Iacurto, 2005).

Table 7

Colour characteristics of *longissimus dorsi* muscle at different storage period (0, 3 and 6 days) in lambs slaughtered at 60 days of age according to the breed.

	Bergamasca	It. Merino	Sopravissana
<i>Lightness (L*)</i>			
0 days	42.88	41.70	41.04
3 days	42.71	42.98	40.78
6 days	43.37	43.27	41.81
<i>Redness (a*)</i>			
0 days	19.24	19.57	19.30
3 days	18.10	18.55	19.30
6 days	18.48	19.43	19.59
<i>Yellowness (b*)</i>			
0 days	3.13 ^b	3.07 ^B	2.61 ^B
3 days	3.84 ^{ab}	4.47 ^A	4.11 ^A
6 days	4.51 ^a	4.88 ^A	4.15 ^A
<i>Chroma value (C*)</i>			
0 days	29.76 ^b	29.33	27.10 ^b
3 days	33.64 ^{ab}	40.58	37.55 ^a
6 days	41.14 ^a	45.72	38.27 ^a
<i>Hue angle (H°)</i>			
0 days	9.37 ^b	8.92 ^B	7.58 ^B
3 days	12.01 ^{ab}	13.50 ^A	11.91 ^A
6 days	13.67 ^a	13.85 ^A	11.84 ^A
<i>Colour difference (ΔE*)</i>			
0-3 days	2.57	3.81	2.75
0-6 days	3.52	3.46	2.61
3-6 days	2.53	2.34	2.00

a, b; A, B Within the same column, means with different letters differ significantly (^{a, b}: P < 0.05; ^{A, B}: P < 0.01).

Meat lipid oxidation is presented in Table 8 through the evolution of TBARS values according to the breed over the storage period. The effects of breed and storage period were significant for TBARS (P < 0.001; Table 9) as well as their interaction (P < 0.001) that reveals different rates of development of lipid oxidation between breeds during 6 days of storage. Fresh meat samples (0 day) of Bergamasca breed showed the highest (P < 0.001; Table 8) TBARS values compared to the other two breeds and are similar to those observed for Apulian light lambs by Cifuni et al. (2000). Higher lipid oxidation is often associated with higher content of unsaturated fatty acid (UFA) (Cheng, 2016). This is not in accordance with the present study as fresh meat of

Bergamasca lambs had the highest TBARS values but the lowest content of UFA (results not shown). Thus, higher lipid oxidation in fresh meat of Bergamasca lambs could be explained by the lowest ($P < 0.001$; Table 9) antioxidant capacity (ORAC) compared to the other two breeds. However, Italian Merino and Sopravissana showed similar meat lipid oxidation ($P > 0.05$) but differed in antioxidant capacity. According to Cheng (2016) many other factors, such as heat and light, conditions of pre-slaughter and pH can be associated with lipid oxidation in meat. As all three breeds had the same treatment and did not differ in terms of pH values, further research is needed to understand the observed difference between breeds.

Table 8
TBARS at different storage period (0, 3 and 6 days) and ORAC values in lambs slaughtered at 60 days of age according to the breed (mean \pm SD)

	Bergamasca	It. Merino	Sopravissana
TBARS (mg MDA kg ⁻¹)			
0 day	0.13 ^a \pm 0.02	0.08 ^b \pm 0.01	0.09 ^b \pm 0.01
3 days	0.15 \pm 0.01	0.16 \pm 0.03	0.14 \pm 0.02
6 days	0.20 \pm 0.06	0.20 \pm 0.04	0.16 \pm 0.03
ORAC (μ mol TE g ⁻¹)	7.12 ^c \pm 0.90	10.00 ^b \pm 0.87	11.32 ^a \pm 0.57

^{a, b, c} Within the same row, means with different letters differ significantly at $P < 0.001$.

Table 9
P-values ($\alpha \leq 0.05$) for fixed effects of breed, storage time and their interactions on drip loss and TBARS values in lambs slaughtered at 60 days of age.

	Breed	Storage time	Breed x Storage time
Drip Loss	0.0205	<0.001	0.2989
TBARS	0.0011	<0.0001	0.0076

Any TBARS difference was not recorded after the 3rd day of storage. Opposite results were reported by Hajji et al. (2016) who observed significant differences among three North African lamb breeds after the 3rd day of storage. In this study, lambs of similar slaughter weight but older than in ours showed TBARS values much higher (> 0.8 mg MDA kg⁻¹) after 3 and 6 days of storage. Our study showed that even though TBARS increased for all the breeds during 6 days of storage period, mean values were very low (< 0.21 mg MDA kg⁻¹) and under the

threshold accepted value of 1 mg MDA kg⁻¹ (Ripoll et al., 2011) indicating a low degree of lipid oxidation.

4. Conclusions

A slight effect of breed on “Agnello del Centro Italia” PGI light lambs under the traditional lamb production system in Central Italy was recorded. Bergamasca, Italian Merino and Sopravissana lambs showed similar growth performances and high ADG and did not differ in terms of carcass weight and dressing percentages. Found differences between lambs in terms of slaughter, carcass and meat characteristics were minor. Meat of Bergamasca lambs had lower protein content than Sopravissana and the lowest antioxidative stability as well as the highest lipid oxidation than both the other breeds. Effect of storage period on colour, drip loss and TBARS was evident.

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Effect of breed on the quality of “Agnello del Centro Italia” PGI light lambs: II. Fatty acid composition of meat and adipose tissue

Abstract

The study aimed to evaluate the effect of breed (Bergamasca, Italian Merino and Sopravissana) on fatty acid (FA) composition of intramuscular (IM) (*longissimus dorsi*, LD) and subcutaneous (SC) fats of light lambs. Total of 33 male single born lambs, labelled as “Agnello del Centro Italia” Protected Geographical Indication (PGI) were reared according to the traditional extensive management and slaughtered at 60 days of age. Breed had effect just on some of the FAs, both in LD muscle and in SC adipose tissue. However, GC data, in combination with chemometric approach (canonical discriminant analysis), effectively discriminated between breeds and correctly predicted them. In general, meat from Bergamasca lambs presented less favourable nutritional quality, because of the lowest conjugated linoleic acid (CLA) and the highest thrombogenic index (TI) in LD muscle compared to Italian Merino and Sopravissana as well as lower values of unsaturated fatty acids (UFA) compared to Italian Merino.

Keywords: fatty acids, light lamb, Agnello del Centro Italia, PGI.

1. Introduction

In Mediterranean countries consumers prefer light lambs, weighing (carcass) around 8-13 kg (Sañudo, Sanchez, & Alfonso, 1998), giving importance to quantity but also to quality of fat, which is defined by the levels and proportions of fatty acids (FAs). In fact, the recognition of the role of food in improvement and preservation of human health is getting more and more attention among consumers. Fats from red meats (visible and invisible) are considered quite unhealthy, because of their high levels of cholesterol and saturated fatty acids (SFA) and their very low levels of n-3 polyunsaturated ones (PUFA) (Fernandez and West, 2005; Salter, 2011; Schaefer, 2002; Siri-Tarino et al., 2010). Moreover, *trans*-FAs, that can be found in milk and meat fat of ruminants as a result of rumen bacterial biohydrogenation of feed PUFA, are gaining increased attention because of their adverse influence on the risk of cardiovascular diseases, but not limited to it (Gebauer et al., 2011). However, ruminant meats may also be a dietary source of nutritionally valuable FAs (Wahle et al., 2004), such as conjugated linoleic acid (CLA), a group of positional and geometric isomers of linoleic acid in which double bonds are conjugated.

Feeding system has been shown to be one of the main factors influencing FA composition of lamb fats (Dervishi et al., 2010) and consequently a main tool to manipulate it through the use of grass (Cividini et al., 2014; Nuernberg et al., 2008) or different supplements (Jaworska et al., 2016; Urrutia et al., 2015) rich in PUFA. Grass feeding and grazing can lead to an increase in the concentrations of CLA, n-3 PUFA and monounsaturated FAs (MUFA) in lamb meat (Aurousseau et al., 2004; Nuernberg et al., 2008; Popova et al., 2015). In addition to feeding system, FA composition in lamb tissues can be influenced by other factors, such as breed, sex, live weight, environment, degree of fatness and interaction between these factors (Juárez et al., 2008; Nuernberg et al., 2008; Wood and Enser, 1997). According to Juárez et al. (2008), the production system (i.e., combination of breed and diet) is the main factor to explain variations in FA composition of light lambs. It is also known that FA composition varies between different lamb fat depots (Vacca et al., 2008), especially

between intramuscular and subcutaneous fat (Gallardo, Dannenberger, Rivero, Pulido, & Nuernberg, 2014; Guler, Aktumsek, & Karabacak, 2011; Juárez et al., 2009; Osorio, Zumalacárregui, Figueira, & Mateo, 2007).

In Central Italy, lamb meat is the main product of transhumant farms where lambs are reared with their mothers on pastures and are supplemented with concentrate and/or hay from day 25-30 until slaughter. Lambs born and reared in Central Italy can be labelled as “Agnello del Centro Italia” Protected Geographical Indication (PGI) (European Union, Commission Implementing Regulation No. 475/2013). The label refers to lambs from a permitted list of local breeds and crossbreeds, some of which are Appenninica, Bergamasca, Fabrianese, Italian Merino and Sopravissana. Lambs are mainly produced as light lambs (carcass weight up to 13 kg) or heavy lambs (carcass weight > 13 kg) for Easter and Christmas market. Few data are however available on FA composition of unweaned lambs reared in extensive systems typical for Central Italy (e.g. Mazzone et al., 2010). Furthermore, there is scarcity of published data on the FA composition of “Agnello del Centro Italia” PGI light lambs. The aim of this study was to evaluate the effect of breed (Bergamasca, Italian Merino and Sopravissana) on FA composition of intramuscular (IM) fat of *longissimus dorsi* muscle and of subcutaneous (SC) adipose tissue of “Agnello del Centro Italia” PGI light lambs, with special reference to their nutritional properties.

2. Materials and methods

The animal handling followed the recommendations of European Union directive 2010/63/EU and Italian law (D. Lgs. n. 26/2014) regarding animal care.

2.1. Experimental design, diet and animal management

The study was conducted under the usual conditions for rearing and management of the transhumant sheep system in Marche region (Caballero et al., 2009). The experiment started in February 2015 with

a total of 33 male single born lambs of 3 breeds, Bergamasca (n=11), Italian Merino (It. Merino; n=11) and Sopravissana (n=11), produced for Easter market. After lambing all lambs were gathered with their dams in an experimental flock that grazed alfalfa-dominated pasture plots from mid-February to mid-April 2015. Dams had free access to hay and were supplemented by corn grain (0.5 kg head⁻¹ day⁻¹) while lambs were given concentrate (50% corn, 50% barley) *ad libitum* in creep feeders from 20 days of age. All animals had free access to water. Feeding samples were collected to determine their chemical composition (Table 1) according to Martillotti et al. (1987).

Table 1
Dry matter (DM) percentage and chemical composition (% of DM) of the diet.

	Pasture	Hay	Corn grain	Concentrate
DM	21.51	88.31	83.52	85.89
Ashes	15.51	7.95	1.39	1.72
Crude protein	15.58	7.34	5.66	7.68
Ether extract	1.65	1.05	2.88	2.73
Crude fibre	21.46	31.28	2.65	2.94
NDF ^a	39.65	63.45	16.30	21.99
ADF ^b	30.11	41.02	3.03	4.90
ADL ^c	6.37	6.15	0.76	1.52

^aNDF, neutral detergent fibre.

^bADF, acid detergent fibre.

^cADL, acid detergent lignin.

2.2. Slaughter and sampling

The lambs were slaughtered at the average of 60 days of age in a commercial slaughterhouse. All lambs were slaughtered on the same day by exsanguinations after stunning by the same procedures. Carcasses were chilled at 4°C for 24 h, and after that the samples of *longissimus dorsi* (LD) muscle and adipose SC tissue were taken between the 1st and 6th lumbar vertebrae of the right side. The samples were frozen at -20 °C until the analytical procedure.

2.3. Determination of total fatty acid composition

Total lipids from freeze-dried muscle samples and from adipose tissue were extracted by SoxtecTM system with the use of petroleum ether. The

liquefied fat (approximately 0.1 mL) was collected and dissolved in hexane (1.0 mL). Fatty acids methyl esters (FAMES) were prepared by “rapid” KOH-catalysed transesterification, according to method ISO 12966-2:2011. The FAME mixture was analysed on a gas chromatograph (HRGC MEGA 2 series, Fisons Instruments, Milano, Italy) equipped with a Rt-2560[®] column (length = 100 m, internal diameter = 0.25 mm, and film thickness = 0.2 µm, Restek, PA, USA), a flame-ionization detector and He as carrier gas (1mL/min flow rate). The initial column temperature of 150 °C was held for 3 min, then it was increased to 240 °C at the rate of 4 °C/min and the final temperature was kept for 15 min. The injector and detector temperatures were set at 250 and 280 °C, respectively. The injection volume was 1.0 µL and the split ratio was 1:50.

FAMES were identified by comparison of their retention times with those of commercial standards (Supelco[®] 37 FAMES Mix; Supelco, Bellefonte, PA, USA) analysed under the same conditions and quantified (% of total FAs) by the peak area normalisation method.

To assess the nutritional quality of the studied IM and SC fats, the PUFA/SFA and the PUFA n-6/n-3 ratios were calculated. Moreover, Atherogenic Index (AI) and Thrombogenic Index (TI) were calculated according to the formulas suggested by Ulbricht and Southgate (1991), and hypocholesterolemic/Hypercholesterolemic ratio (h/H) was calculated according to (Fernández et al., 2007).

2.4. Data analysis

Statistical analyses were conducted using the software JMP[®] Version 10 (SAS Institute Inc., Cary, NC). Measured variables (FAs), which global mean value was above 0.1% (of total FAs) and nutritional indices were included in multivariate analysis. Correlation between variables was determined by mean centred data matrix by the Pearson product-moment correlation coefficient (r). Variable reduction was achieved by PCA (Principal Component Analysis) on variance-covariance matrix. Mean centred data were used to optimally describe the orientation of scores and loadings. A linear discriminant analysis (DA) was also performed on the same set of variables to evaluate their ability to discriminate between breeds and to predict the correct breed

assignment. A one-way analysis of variance (ANOVA) was carried out to evaluate between breed differences for each of the variables. Multiple comparisons among means were carried out through the Tukey-Kramer's Honest Significant Difference (HSD) test and level of significance was set to 0.05.

3. Results and discussion

3.1. Multivariate analysis

Total FA composition of LD muscle and SC adipose tissue of Bergamasca, Italian Merino and Sopravissana lambs is presented in Table 2 and 4, respectively. In general, intramuscular FA percentages of the present study were similar to those of Italian Merino light lambs reared according to similar production system (Oriani et al., 2005), and to those of some other Italian sheep breeds slaughtered at similar age, such as Apulian (Cifuni et al., 2000), Leccese and Comisana (D'Alessandro et al., 2015), Apenninica (Mazzone et al., 2010). Ratios and indices useful to evaluate the nutritional quality of LD muscle and SC adipose tissue are presented in Tables 3 and 5, respectively.

Experimental data (individual FAs and ratios) were explored by means of PCA to evaluate the structure of variables (measured and derived from measured parameters) and objects (i.e., samples).

The first two PCs globally explained 86% of the total variability of the samples in term of their FA composition. Loadings plot in Figure 1 showed that oleic acid (C18:1) and a cluster of longer SFA (C18:0, C17:0, C20:0) had positive loadings on PC1, while shorter SFA (C12:0, C14:0, C15:0, C16:0) had negative loadings, as well as all PUFA and *trans*-FAs (18:1t, 18:2t, CLA). On the contrary, PC2 was more affected by nutritional indices (TI and AI with high positive loadings; h/H with negative loading) than by measured variables, except for C18:1. The loadings plot clearly reflected the biochemical closeness inside the n-3 (C18:3, C20:5, C22:5, C22:6) and n-6 (C18:2, C20:4) clusters. The biological precursors of the families, linoleic (C18:2) and α -linolenic (C18:3) acids, were positively correlated as well.

As expected, PUFA/SFA ratio was positively correlated with all the PUFAs cited above but it was negatively correlated only with the SFA C18:0. Positive linear correlations (vectors oriented in the same directions) were also found between AI and C14:0 and between h/H and C18:1. The strongest inverse correlations (variables positioned in opposite direction with respect to the axes origin and far from the plot origin) were found between the pairs AI vs. h/H, h/H vs. C16:0, TI vs. PUFA/SFA, TI vs. C18:3, AI vs. C18:1. No correlation (variables positioned in approximately orthogonal directions between them) was found between TI and n-6/n-3 ratio, and between AI and h/H ratio. The matrix of correlation coefficients (Table 6) summarizes the strength of the linear relationships between each pair of variables.

The scores plot in Figure 1 shows the distribution of the samples on the plane defined by PC1 and PC2. A biplot should characterise the objects in terms of their values of the original variables; however, to avoid excessive number of points on the same plot, we simply compared the scores plot with the corresponding loadings plot for the same PCs in Figure 1. Lamb fats were well differentiated depending on the body part (IM vs. SC): SC samples had positive relationships with variables having positive loadings in PC1 (mainly C18:0 and C18:1, but also with C10:0, C17:0, C20:0), while IM fats had positive relationships with variables having negative PC1 loadings (shorter SFA, PUFA, *trans*-FAs, and CLA). Hence, PC1 was able to distinguish between IM and SC fat, the latter being characterized by higher values of C18:1 (28.27-32.79 vs. 27.69-28.88%), C18:0 (18.86-20.35 vs. 11.47-15.90%), C17:0 (1.50-1.64 vs. 1.21-1.34%), C20:0 (0.19-0.20 vs. 0.08-0.14%), and lower values of C16:0 (20.23-21.18 vs. 23.46-24.79%), n-6 PUFA (2.17-2.30 vs. 3.72 - 4.46%), n-3 PUFA (1.24-1.28 vs. 2.39-3.00%), and CLA (1.45-1.78 vs. 1.81-2.20%). PC2 was somewhat useful for differentiating between breeds. In fact, Bergamasca samples were generally characterized by positive loadings on PC2, while Sopravissana and Italian Merino samples, which are genetically correlated, mainly lay undifferentiated on the negative PC2 space. Higher values of TI characterized SC fats of Bergamasca, while h/H values and C18:1 levels “drove” the differentiation of Italian Merino

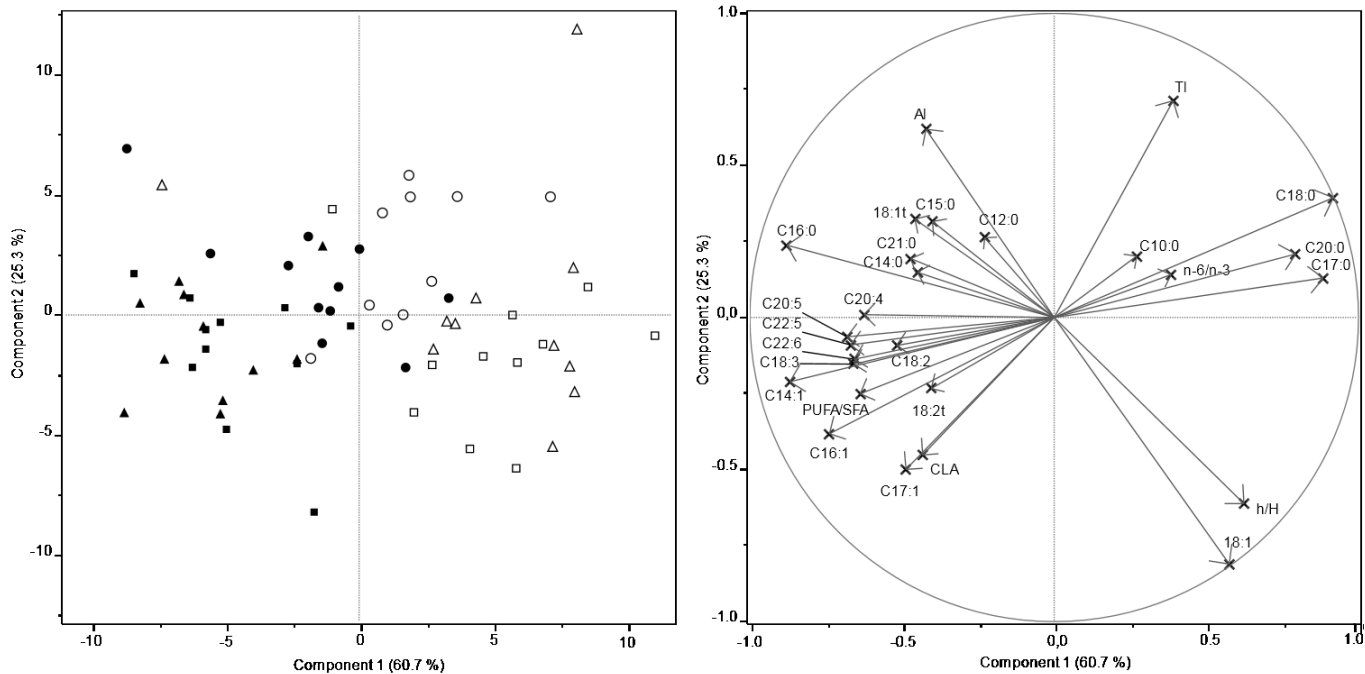


Figure 1 - Left: PCA scores plot of intramuscular (IM) and subcutaneous (SC) fat samples. Symbols are: ● Bergamasca, IM; ▲ Sopravissana, IM; ■ Italian Merino, IM; ○ Bergamasca, SC; △ Sopravissana SC; □ Italian Merino, SC. Right: PCA loadings plot of variables (individual and ratios).

and Sopravissana SC fat composition (lower right quarter) away from that of Bergamasca breed (upper right quarter). Higher AI values characterized the FAs composition of IM fat of Bergamasca, while IM samples of Italian Merino and Sopravissana were “pulled” towards negative scores (lower left quarter) mainly by unsaturated fatty acids (C14:1, C17:1, C16:1, n-3 PUFA, C18:2, CLA).

A linear discriminant analysis was developed to check if the same set of variables could efficiently discriminate breeds. Figure 2 shows the scores of the samples on the plane defined by the two canonical functions obtained, selecting “breed” as category. The plot shows that these two coordinates separate the three breeds in a way that is correlated to the genetic distance between them (Sopravissana and Italian Merino closer to each other than to Bergamasca), no matter what the type of fat is (IM or SC). Since there was no Validation set, the entire data set is considered the Training set: out of 66 observations, only 4 were misclassified (3 Italian Merino, predicted as Sopravissana, and 1 Sopravissana, predicted as Italian Merino).

3.2. Bivariate analysis

Based on the results of PCA and DA, one-way ANOVA was carried out on breeds, for IM and SC fat samples separately (Tables 2 - 4).

3.2.1. Saturated fatty acids

Bergamasca lambs showed higher value ($0.65 \pm 0.11\%$) for capric acid (C10:0) than Italian Merino and the highest value ($15.90 \pm 1.75\%$) for stearic acid (C18:0) in LD muscle. These results are probably due to higher content of these acids in the Bergamasca ewe’s milk (Atti et al., 2006). Lambs of the three breeds did not differ in levels of lauric (C12:0), myristic (C14:0) and palmitic (C16:0) acids, for which LDL cholesterol-raising effect is reported. Higher content of arachidic acid (C20:0) in LD muscle was also detected in Bergamasca lambs, compared to the other breeds (0.14 vs. 0.08%). As a result of significantly higher values of previously mentioned FAs, Bergamasca lambs had the highest amount of total SFA in LD muscle compared to other two breeds (53.90 vs. 48.87-50.47%).

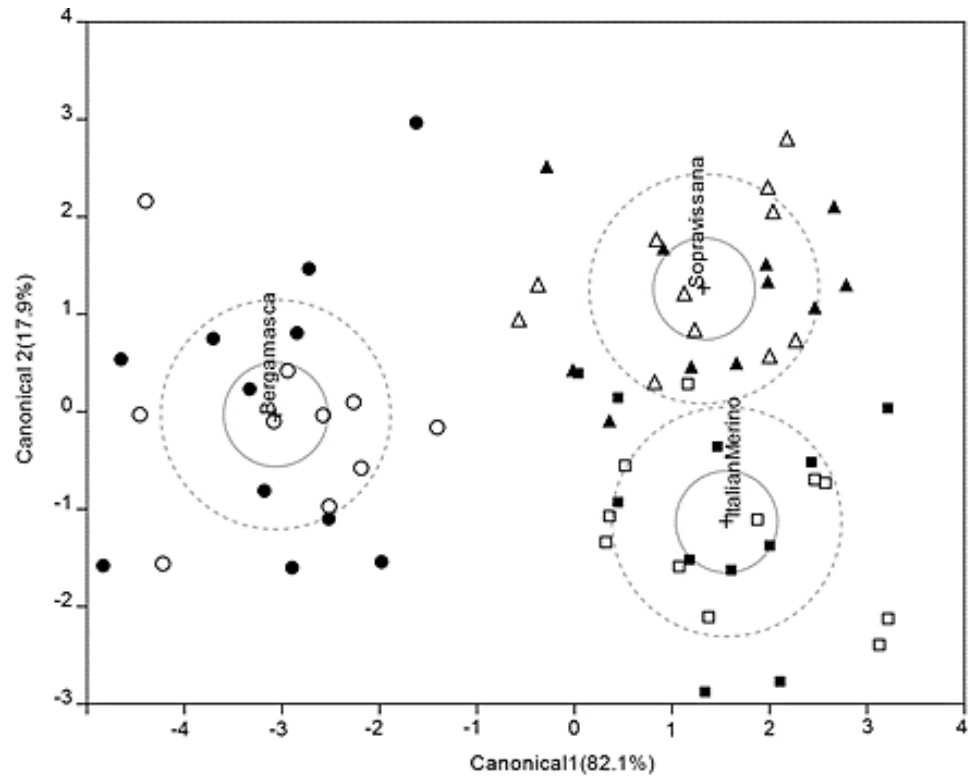


Figure 2 - Scores of the samples on the plain of the canonical variables obtained to predict the breed. Continuous line = centroid confidence limits (95%); dashed lines = normal 50% contours.

High tissue levels of SFAs are characteristics of unweaned lambs, as they are not considered ruminants from metabolic point of view and their FA composition primary depends on that of ewe's milk (Vacca et al., 2008). However, obtained percentages for total SFAs were higher than those reported for Sarda (Addis et al., 2013) and Mouflon × Sarda and Sarda × Sarda (Vacca et al., 2008) suckling lambs.

Regarding SC adipose tissue, C21:0 was the only SFA affected by breed, with Bergamasca lambs having higher values than the other two breeds (0.26 vs. 0.08-0.10%). Consequently, breeds did not differ in total SFA content, which was similar to that of SC fat of Grazalema Merino light lambs reported by Juárez et al. (2009).

3.2.2. *Unsaturated fatty acids*

MUFA profile showed the lower percentage of palmitoleic acid (C16:1) in LD muscle (1.46 vs. 2.01-2.05%) and in SC adipose tissue (1.06 vs. 1.26-1.38%) of Bergamasca lambs compared to the other two breeds. Effect of breed on C16:1 is also reported by Juárez et al. (2008) for light lambs of five Spanish sheep breeds. Lower content of C14:1 (0.22 vs. 0.32-0.36%) and C17:1 (0.47 vs. 0.61-0.62%) in LD muscle (Table 2) was also observed in Bergamasca lambs with respect to the other breeds. The major MUFA was \sum C18:1c (sum of oleic and *cis*-vaccenic acids), both in the LD muscle (27.69-28.88%) and SC adipose tissue (28.27-32.79%). Oleic acid is also reported to be the major MUFA for meat of unweaned lambs by other authors (Addis et al., 2013; Oriani et al., 2005; Vacca et al., 2008). While it did not differ significantly between breeds in LD muscle, Italian Merino had higher percentages (32.79 vs. 28.27%) of \sum C18:1c in SC adipose tissue than Bergamasca lambs. Content of \sum C18:1c is comparable with reported values of oleic acid for meat of light lambs of different breeds (della Malva et al., 2016; Mazzone et al., 2010; Santos-Silva et al., 2002). Total MUFA content in LD muscle from all breeds (29.84-31.84%) is lower to that in Leccese light lambs (36.55%) slaughtered at same age as in the present study (D'Alessandro et al., 2015). Total MUFA content in SC adipose tissue from Bergamasca (29.94%) was significantly lower than in Italian Merino lambs (34.76%). Observed values were lower than reported for SC fat of Grazalema Merino and Churra Lebrijana light lambs (40.10

and 41.38%, respectively) by Juárez et al. (2009) or for perennial and pelvic fat of Mouflon × Sarda and Sarda × Sarda suckling lambs (41.0 and 45.3%, respectively) by Vacca et al. (2008).

PUFAs were also influenced by breed. Italian Merino lambs showed significantly higher values of α -linolenic acid (C18:3 n-3) in LD muscle, compared to Bergamasca (1.63 vs. 1.40%), while no differences between breeds were observed for this acid in SC adipose tissue. The obtained mean value for α -linolenic acid in LD muscle (1.51%) is similar to those reported for IM fat of Muflon × Sarda and Sarda × Sarda suckling lambs (Vacca et al., 2008) or Merino Branco and Ile de France × Merino Branco light lambs (Santos-Silva et al., 2002), while lower values are reported for Leccese (0.91%) and Comisana (1.0%) light lambs slaughtered at same age (D'Alessandro et al., 2015). Bergamasca lambs had higher values of eicosapentaenoic acid (EPA; C20:5 n-3) in SC adipose tissue than Italian Merino (0.06 vs. 0.02%) while other long chain n-3 PUFAs were not affected by breed, both in LD muscle and SC adipose tissue.

Marino et al. (2008) and Santos-Silva et al. (2002) reported higher values of EPA in IM fat of light lambs reared with ewes on pasture and supplemented with hay and concentrate (0.41 vs. 0.66 and 0.74%, respectively) as well as higher values of docosahexaenoic acid (DHA; C22:6 n-3) (0.23 vs. 0.67% and 0.42%, respectively). However, no significant differences between breeds were evident concerning the total n-3 PUFA, both in LD muscle and SC adipose tissue. Other authors also found minor or no effect of breed on n-3 PUFA proving that differences in n-3 PUFA are more influenced by feeding system and presence/absence of dry/fresh herbage than by breed (Hajji et al., 2016; Santos-Silva et al., 2002).

No differences between breeds in total n-6 PUFA were observed in LD muscle and SC adipose tissue of lambs. Regarding single n-6 PUFA, breed had effect just on C20:4 in SC adipose tissue, with Bergamasca lambs having higher value than other breeds (0.22 vs. 0.04 - 0.07%). Obtained values for linoleic acid (18:2 n-6) in LD muscle were much lower than reported for Appenninica lambs (Mazzone et al., 2010) and Italian Merino (Oriani et al., 2005) slaughtered at the similar age.

Total PUFA values in LD muscle and SC adipose tissue did not differ between breeds. Mean value in LD muscle (8.71%) was similar to the one reported for Leccese (8.91%) and lower than reported for Comisana (10.94%) light lambs slaughtered at the same age (D'Alessandro et al., 2015), Altamura lambs (17.24%) (della Malva et al., 2016), and Italian Merino lambs (16.09%) (Oriani et al., 2005) slaughtered at similar age (70 days). Mean value of total PUFA in SC adipose tissue (5.13%) was similar to those reported for Grazalema Merino (4.97%) and Churra Lebrijana (5.91%) light lambs (Juárez et al., 2009). Bergamasca breed showed lower content of total UFA in LD muscle (37.76 vs. 41.50%) and therefore less favourable meat for human health than Italian Merino.

3.2.3. *Trans and conjugated fatty acids*

C18:1 *trans*-isomers in SC adipose tissue significantly differed between breeds, where Bergamasca had the highest (4.90%) and Italian Merino the lowest (1.31%) values. In the same time, this is the only difference in FA composition found between Italian Merino and Sopravissana lambs, both in LD muscle and SC adipose tissue. This is in accordance with the study of Santos-Silva et al. (2002) where FA composition was not significantly affected by breed, when breeds were similar in origin. Meat from ruminants has higher levels of CLA than meat from non-ruminants, as product of bacterial isomerisation or/and biohydrogenation of PUFA in the rumen (Griinari and Bauman, 1999). Rumenic acid (C18:2 *cis*-9, *trans*-11) is the main isomer found in meat and milk of ruminants and anticarcinogenic, antidiabetic and other positive effects to human health are attributed to it (Schmid et al., 2006). Bergamasca lambs displayed significantly lower (1.81 vs. 2.18-2.20%) value of CLA *cis*-9, *trans*-11 in LD muscle than Italian Merino and Sopravissana, while no differences were observed in SC adipose tissue. In any case, these levels were higher than average value (1.02%) reported for lambs under “Agnello di Sardegna” quality label, where Sarda lambs were fed with maternal milk as they followed their mother at pasture and were slaughtered at 30–40 days of age (Addis et al., 2013). It is known that grass-fed lambs tend to have higher amounts of rumenic acid than concentrate-fed (Cividini et al., 2014; Guler et al.,

2011; Nuernberg et al., 2008). Furthermore, CLA *cis*-9, *trans*-11 can differ between lambs grazing different pastures (Willems et al., 2014). Values for rumenic acid from present study were higher than reported for Apenninica (Mazzone et al., 2010) or Merino Branco and Ile de France × Merino Branco (Santos-Silva et al., 2002) light lambs with similar rearing system. High values (1.62%) of rumenic acid were also observed in SC adipose tissue, but they were not influenced by breed.

3.2.4. Nutritional indices

Significant difference occurred in PUFA/SFA ratio of LD muscle, with Bergamasca lambs having lower (0.15 vs. 0.20) ratio than Italian Merino. However, the PUFA/SFA ratio of all three breeds was relatively low (0.15-0.20 and 0.09-0.10 in LD and SC adipose fats, respectively) and did not reach the recommended value for human health (above 0.4) (Wood et al., 2003). Obtained PUFA/SFA ratio was comparable with those of Italian Merino (0.25) (Oriani et al., 2005) and Comisana (0.21) (D'Alessandro et al., 2015) light lambs slaughtered at similar age, while higher values are reported for Sarda suckling (0.55) by Addis et al. (2013) and Appenninica light lambs (0.42) by Mazzone et al. (2010).

The high proportion of PUFAs is not necessarily healthy in itself, if it is not balanced in relation to the n-6/n-3 ratio. According to Enser et al. (2001), the n-6/n-3 ratio is a risk factor in cancer and coronary heart disease, especially in the formation of blood clots that can lead to a heart attack. Although the differences between breeds were not observed in terms of n-6/n-3 ratio, observed values ranged from 1.38 to 1.52 and from 1.72 to 1.83 for IM and SC fat respectively, and were within the nutritional recommendations for human diet, as this ratio should vary from 1 to 4 (Simopoulos, 2004; Wood et al., 2003). Higher n-6/n-3 ratio was found for lambs exclusively milk fed (Vacca et al., 2008) as well as for light lambs reared with their dams on pasture and supplemented with hay and concentrate (della Malva et al., 2016; Santos-Silva et al., 2002).

The hypocholesterolemic/Hypercholesterolemic (h/H) ratio is used as an index of the cholesterolemic effect of the fat source (Santo-Silva et al., 2002) and it can be influenced by genetic factors, as fat deposition

differs between breeds (Raes et al., 2003; Sinanoglou et al., 2013). However, the breed did not influence h/H ratio in the present study. Observed values (1.01-1.23) were lower than reported for different breeds of suckling (Sinanoglou et al., 2013) or light lambs (Santos-Silva et al., 2002).

Atherogenic (AI) and thrombogenic (TI) indices are used for evaluation of lipid quality, as they consider the different effects that single FAs might have on human health. Ulbricht and Southgate (1991) recommended replacing the ratio PUFA/SFA by these indices as a measure of the influence of dietary lipids on the incidence of coronary heart disease. In the present study, higher TI was found in Bergamasca lambs LD muscle compared to other breeds (1.99 vs. 1.62-1.75). According to Sinanoglou et al. (2013), the appropriate values of AI and TI for a healthy diet are under 1.0 and values obtained in present study are above this recommended value, both in LD muscle and SC adipose tissue (see Tables 3 and 5). However, obtained AI and TI values in LD muscle are similar to those from Italian Merino suckling lambs (Oriani et al., 2005), while lower values are reported for Altamurana light lamb (della Malva et al., 2016). Vacca et al. (2008) also reported lower values for AI and TI for IM fat of suckling lambs while values for these indices reported in adipose tissue are comparable with the present study.

Table 2

Fatty acid composition (% of total fatty acid as methyl esters) of the total lipids of the *longissimus dorsi* muscle from “Agnello del Centro Italia” PGI light lambs (mean \pm SD; n = 11).

Fatty acid	Bergamasca	It. Merino	Sopravissana
C10:0	0.65 ^a \pm 0.11	0.49 ^b \pm 0.12	0.54 ^{ab} \pm 0.09
C11:0	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01
C12:0	1.45 \pm 0.25	1.30 \pm 0.29	1.37 \pm 0.21
C13:0	0.09 \pm 0.01	0.09 \pm 0.02	0.10 \pm 0.01
C14:0	9.38 \pm 1.09	9.20 \pm 1.13	9.77 \pm 0.82
C15:0	0.87 \pm 0.09	0.82 \pm 0.10	0.86 \pm 0.05
C16:0	23.72 \pm 1.97	23.46 \pm 1.55	24.79 \pm 1.46
C17:0	1.34 \pm 0.15	1.23 \pm 0.12	1.21 \pm 0.12
C18:0	15.90 ^a \pm 1.75	11.89 ^b \pm 1.85	11.47 ^b \pm 2.45
C20:0	0.14 ^a \pm 0.05	0.08 ^b \pm 0.03	0.08 ^b \pm 0.01
C21:0	0.31 \pm 0.09	0.26 \pm 0.12	0.23 \pm 0.06
C14:1	0.22 ^b \pm 0.08	0.32 ^a \pm 0.06	0.36 ^a \pm 0.08
C16:1 n-7	1.46 ^b \pm 0.37	2.01 ^a \pm 0.27	2.05 ^a \pm 0.37
C17:1	0.47 ^b \pm 0.11	0.62 ^a \pm 0.07	0.61 ^a \pm 0.11
Σ C18:1t	3.89 \pm 1.45	4.84 \pm 0.83	4.72 \pm 0.82
Σ C18:1c	27.69 \pm 3.08	28.88 \pm 2.97	28.46 \pm 2.00
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA)	1.81 ^b \pm 0.25	2.20 ^a \pm 0.36	2.18 ^a \pm 0.42
Σ 18:2t	1.56 ^b \pm 0.21	1.87 ^a \pm 0.24	1.92 ^a \pm 0.30
C18:2 n-6	2.96 \pm 1.08	3.49 \pm 1.02	2.90 \pm 1.09
C18:3 n-3	1.40 ^b \pm 0.19	1.63 ^a \pm 0.19	1.49 ^{ab} \pm 0.23
C20:2 n-6	0.08 \pm 0.03	0.08 \pm 0.01	0.08 \pm 0.01
C20:4 n-6	0.69 \pm 0.30	0.89 \pm 0.65	0.74 \pm 0.25
C20:5 n-3 (EPA)	0.34 \pm 0.13	0.50 \pm 0.29	0.40 \pm 0.15
C22:5 n-3 (DPA)	0.46 \pm 0.11	0.61 \pm 0.32	0.52 \pm 0.15
C22:6 n-3 (DHA)	0.19 \pm 0.06	0.27 \pm 0.15	0.24 \pm 0.06
Other fatty acids	2.89 \pm 0.22	2.92 \pm 0.28	2.88 \pm 0.20
SFA	53.90 ^a \pm 2.25	48.87 ^b \pm 2.40	50.47 ^b \pm 3.12
MUFA	29.84 \pm 2.83	31.84 \pm 2.89	31.47 \pm 2.22
PUFA	7.92 \pm 1.60	9.66 \pm 2.49	8.54 \pm 1.92
UFA	37.76 ^b \pm 2.37	41.50 ^a \pm 2.86	40.02 ^{ab} \pm 2.68
n-6 PUFA	3.72 \pm 1.29	4.46 \pm 1.67	3.72 \pm 1.27
n-3 PUFA	2.39 \pm 0.39	3.00 \pm 0.90	2.65 \pm 0.51

Values in a row with different letters are significantly different ($P < 0.05$).

Σ C18:1t = sum of unidentified C18:1 *trans* isomers; Σ C18:1c = C18:1 Δ 9c + Δ 11c; Σ C18:2t = 18:2 n-6 c,t + t,c + t,t; SFA, Saturated Fatty Acids = Σ (C10:0, C11:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, C21:0); MUFA, Monounsaturated Fatty Acids = Σ (C14:1, C16:1 n-7, C17:1, Σ 18:1c); PUFA, Polyunsaturated Fatty Acids = (C18:2 n-6, C18:3 n-3, C18:2 c9t11, C20:2 n-6, C20:4 n-6, C20:5 n-3, C22:5 n-3, C22:6 n-3); UFA, Unsaturated Fatty Acids = (MUFA + PUFA); n-6 PUFA = Σ (C18:2 n-6, C20:2 n-6, C20:4 n-6); n-3 PUFA = Σ (C18:3 n-3, C20:5 n-3, C22:5 n-3, C22:6 n-3).

Table 3

Ratios and indices of fatty acids determined in the total lipids of the *longissimus dorsi* muscle from “Agnello del Centro Italia” PGI light lambs (mean \pm SD; n = 11).

	Bergamasca	It. Merino	Sopravissana
PUFA/SFA	0.15 ^b \pm 0.03	0.20 ^a \pm 0.06	0.17 ^{ab} \pm 0.04
n-6/n-3	1.52 \pm 0.39	1.48 \pm 0.15	1.38 \pm 0.34
h/H	1.03 \pm 0.15	1.12 \pm 0.17	1.01 \pm 0.10
AI	1.76 \pm 0.30	1.58 \pm 0.26	1.73 \pm 0.19
TI	1.99 ^a \pm 0.19	1.62 ^b \pm 0.22	1.75 ^b \pm 0.20

Values in a row with different letters are significantly different (P < 0.05).

SFA, saturated fatty acids = Σ (C10:0, C11:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, C21:0); MUFA, Monounsaturated Fatty Acids = Σ (C14:1, C16:1 n-7, C17:1, Σ 18:1c); PUFA, Polyunsaturated fatty acids = (C18:2 n-6, C18:3 n-3, C18:2 c9t11, C20:2 n-6, C20:4 n-6, C20:5 n-3, C22:5 n-3, C22:6 n-3); n-6 PUFA = Σ (C18:2 n-6, C20:2 n-6, C20:4 n-6); n-3 PUFA = Σ (C18:3 n-3, C20:5 n-3, C22:5 n-3, C22:6 n-3); h/H, hypocholesterolemic/Hypercholesterolemic ratio = (C18:1 n-9 + C18:2 n-6 + C18:3 n-3 + C20:2 n-6 + C20:4 n-6 + C20:5 n-3 + C22:5 n-3 + C22:6 n-3)/(C14:0 + C16:0); AI, Atherogenic Index = (C12:0 + 4 \times C14:0 + C16:0)/(MUFA + n-6 PUFA + n-3 PUFA); TI, Thrombogenic Index = (C14:0 + C16:0 + C18:0)/[0.5 \times MUFA + 0.5 \times n-6 PUFA + 3 \times n-3 PUFA + (n-3/n-6)].

Table 4

Fatty acid composition (% of total fatty acid as methyl esters) of the total lipids of the subcutaneous adipose tissue from “Agnello del Centro Italia” PGI light lambs (mean \pm SD; n = 11).

Fatty acid	Bergamasca	It. Merino	Sopravissana
C10:0	0.81 \pm 0.18	0.73 \pm 0.18	0.67 \pm 0.13
C11:0	0.04 \pm 0.01	0.04 \pm 0.01	0.04 \pm 0.01
C12:0	1.68 \pm 0.27	1.50 \pm 0.47	1.38 \pm 0.35
C13:0	0.10 \pm 0.02	0.10 \pm 0.03	0.10 \pm 0.02
C14:0	10.08 \pm 1.02	9.58 \pm 1.69	9.38 \pm 1.83
C15:0	0.95 \pm 0.08	0.85 \pm 0.17	0.83 \pm 0.15
C16:0	21.18 \pm 1.86	20.39 \pm 2.04	20.23 \pm 3.07
C17:0	1.50 \pm 0.13	1.57 \pm 0.10	1.64 \pm 0.15
C18:0	18.86 \pm 3.06	19.63 \pm 2.89	20.35 \pm 3.63
C20:0	0.19 \pm 0.04	0.19 \pm 0.01	0.20 \pm 0.03
C21:0	0.26 ^a \pm 0.11	0.08 ^b \pm 0.05	0.10 ^b \pm 0.05
C14:1	0.15 \pm 0.04	0.14 \pm 0.03	0.15 \pm 0.04
C16:1 n-7	1.06 ^{b±} 0.23	1.38 ^a \pm 0.17	1.26 ^{ab} \pm 0.20
C17:1	0.47 \pm 0.06	0.45 \pm 0.08	0.46 \pm 0.04
Σ C18:1t	4.90 ^{a±} 1.37	1.31 ^c \pm 0.50	2.92 ^b \pm 1.82
Σ C18:1c	28.27 ^b \pm 1.69	32.79 ^a \pm 2.72	30.82 ^{ab} \pm 4.40
C18:2 <i>cis</i> -9, <i>trans</i> -11 (CLA)	1.45 \pm 0.55	1.78 \pm 0.29	1.65 \pm 0.29
Σ 18:2t	1.55 \pm 0.18	1.67 \pm 0.19	1.75 \pm 0.26
C18:2 n-6	1.96 \pm 0.21	2.08 \pm 0.19	2.16 \pm 0.29
C18:3 n-3	0.99 \pm 0.11	1.07 \pm 0.13	1.04 \pm 0.18
C20:2 n-6	0.08 \pm 0.04	0.04 \pm 0.02	0.07 \pm 0.03
C20:4 n-6	0.22 ^{a±} 0.14	0.04 ^b \pm 0.02	0.07 ^b \pm 0.05
C20:5 n-3 (EPA)	0.06 ^a \pm 0.05	0.02 ^b \pm 0.02	0.03 ^{ab} \pm 0.02
C22:5 n-3 (DPA)	0.15 \pm 0.05	0.14 \pm 0.06	0.15 \pm 0.04
C22:6 n-3 (DHA)	0.05 \pm 0.02	0.04 \pm 0.03	0.05 \pm 0.02
Other fatty acids	3.02 ^a \pm 0.16	2.39 ^b \pm 0.23	2.47 ^b \pm 0.22
SFA	55.64 \pm 3.24	54.65 \pm 3.17	54.93 \pm 3.55
MUFA	29.94 ^b \pm 1.91	34.76 ^a \pm 2.85	32.70 ^{ab} \pm 4.49
PUFA	4.95 \pm 0.72	5.22 \pm 0.62	5.22 \pm 0.64
UFA	34.89 ^b \pm 2.55	39.98 ^a \pm 3.30	37.92 ^{ab} \pm 4.71
n-6 PUFA	2.26 \pm 0.34	2.17 \pm 0.21	2.30 \pm 0.31
n-3 PUFA	1.24 \pm 0.20	1.28 \pm 0.22	1.27 \pm 0.22

Values in a row with different letters are significantly different ($P < 0.05$).

Σ C18:1t = sum of unidentified C18:1 *trans* isomers; Σ C18:1c = C18:1 Δ 9c + Δ 11c; Σ C18:2t = 18:2 n-6 c,t + t,c + t,t; SFA, Saturated Fatty Acids = Σ (C10:0, C11:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, C21:0); MUFA, Monounsaturated Fatty Acids = Σ (C14:1, C16:1 n-7, C17:1, Σ 18:1c); PUFA, Polyunsaturated Fatty Acids = (C18:2 n-6, C18:3 n-3, C18:2 c9t11, C20:2 n-6, C20:4 n-6, C20:5 n-3, C22:5 n-3, C22:6 n-3); UFA, Unsaturated Fatty Acids = (MUFA + PUFA); n-6 PUFA = Σ (C18:2n-6, C20:2n-6, C20:4n-6); n-3 PUFA = Σ (C18:3n-3, C20:5n-3, C22:5n-3, C22:6n-3).

Table 5

Ratios and indices of fatty acids determined in the total lipids of the subcutaneous adipose tissue from “Agnello del Centro Italia” PGI light lambs (mean \pm SD; n = 11).

	Bergamasca	It. Merino	Sopravissana
PUFA/SFA	0.09 \pm 0.02	0.10 \pm 0.02	0.10 \pm 0.02
n-6/n-3 PUFA	1.83 \pm 0.15	1.72 \pm 0.13	1.82 \pm 0.21
h/H	1.02 \pm 0.11	1.23 \pm 0.21	1.19 \pm 0.26
AI	1.90 \pm 0.21	1.60 \pm 0.36	1.68 \pm 0.50
TI	2.48 \pm 0.33	2.20 \pm 0.34	2.33 \pm 0.46

Values in a row with different letters are significantly different ($P < 0.05$).

SFA, saturated fatty acids = Σ (C10:0, C11:0, C12:0, C13:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, C21:0); MUFA, Monounsaturated Fatty Acids = Σ (C14:1, C16:1 n-7, C17:1, Σ 18:1c); PUFA, Polyunsaturated fatty acids = (C18:2 n-6, C18:3 n-3, C18:2 c9t11, C20:2 n-6, C20:4 n-6, C20:5 n-3, C22:5 n-3, C22:6 n-3); n-6 PUFA = Σ (C18:2 n-6, C20:2 n-6, C20:4 n-6); n-3 PUFA = Σ (C18:3 n-3, C20:5 n-3, C22:5 n-3, C22:6 n-3); h/H, hypocholesterolemic/Hypercholesterolemic ratio = (C18:1 n-9 + C18:2 n-6 + C18:3 n-3 + C20:2 n-6 + C20:4 n-6 + C20:5 n-3 + C22:5 n-3 + C22:6 n-3)/(C14:0 + C16:0); AI, Atherogenic Index = (C12:0 + 4 \times C14:0 + C16:0)/(MUFA + n-6 PUFA + n-3 PUFA); TI, Thrombogenic Index = (C14:0 + C16:0 + C18:0)/[0.5 \times MUFA + 0.5 \times n-6 PUFA + 3 \times n-3 PUFA + (n-3/n-6)].

4. Conclusions

The present study, to our knowledge, is the first one reporting comparison of detailed FA profiles of light lambs reared according to the traditional extensive management of Central Italy, recently labelled with the “Agnello del Centro Italia” PGI.

With respect to IM fat, SC tissue of the analysed samples was characterized by higher values of monounsaturated FAs (C18:1), and longer SFAs (C18:0, C17:0, C20:0), and by lower values of C16:0, n-6 and n-3 PUFA, and CLA.

Differences between breeds occurred just for some of the FAs and ratios (C10:0, C18:0, C20:0, C21:0, C14:1, C16:1, C17:1, CLA, Σ 18:1t, Σ 18:2t, C18:3, C20:4, C20:5, SFA, MUFA, PUFA/SFA), but always as differences between Bergamasca compared with other breeds, both in LD muscle and adipose tissue. Therefore, probably due to the similar genotype, Italian Merino and Sopravissana (both Merino derived breeds) had very similar FA composition. GC data in combination with chemometric approach (canonical discriminant analysis) was effective to discriminate between breeds and correctly predict them.

In general, meat from Bergamasca lambs presented less favourable nutritional quality, because of lower CLA as well as higher TI, compared to Italian Merino and Sopravissana. Interestingly, PCA described a characteristic behaviour of nutritional quality indices, each of them “driving” the differentiation between fat type and breed: the highest values of TI (2.48) characterized SC adipose tissue of Bergamasca lambs while the highest h/H ratios (1.19-1.23) were a feature of Merino derived breeds; Bergamasca IM fat had the highest AI (1.76), whereas Sopravissana and Italian Merino IM fat had the highest PUFA/SFA (0.17-0.20).

Levels of *trans*-acids (sum of 18:1t and 18:2t) ranged from 5.45 to 6.64 % in IM fat, and from 2.98 to 6.45% in SC tissue. Meat and meat products frequently represent the main sources of total fat in diets of Western European countries (Hulshof et al., 1999), hence the contribution of lamb meat to the daily intake of *trans* FAs may not be negligible.

Table 6 - Pearson correlation matrix (n = 66). The most significant (P < 0.0001) coefficients are highlighted in boldface black (positive correlations) and italic boldface grey (negative correlations).

	C10:0	C12:0	C14:0	C14:1	C15:0	C16:0	C16:1	C17:0	C17:1	C18:0	18:1t	18:1	18:2t	C18:2	C20:0	C18:3	CLA	C21:0	C20:4	C20:5	C22:5	C22:6	n-6/ n-3	PUFA/ SFA	h/H	AI	TI
C10:0	1,0000																										
C12:0	0,7239	1,0000																									
C14:0	0,5228	0,8584	1,0000																								
C14:1	-0,4414	-0,0093	0,2808	1,0000																							
C15:0	0,5250	0,8783	0,8463	0,1150	1,0000																						
C16:0	-0,1517	0,2847	0,5086	0,7202	0,4572	1,0000																					
C16:1	-0,5325	-0,1060	0,1741	0,9288	-0,0146	0,5907	1,0000																				
C17:0	0,3480	-0,1066	-0,3225	<i>-0,8691</i>	-0,2046	<i>-0,7248</i>	<i>-0,7931</i>	1,0000																			
C17:1	-0,5618	-0,3129	-0,1394	0,6575	-0,1895	0,2113	0,6717	-0,4642	1,0000																		
C18:0	0,3255	-0,1108	-0,3476	<i>-0,8763</i>	-0,2453	-0,6854	<i>-0,8192</i>	0,8627	-0,6572	1,0000																	
18:1t	-0,2617	-0,1195	-0,0816	0,3318	0,0286	0,2391	0,1180	-0,3590	0,3436	-0,3296	1,0000																
18:1	0,0133	-0,3370	-0,3525	-0,3309	-0,4739	-0,6629	-0,1227	0,4138	0,1090	0,2134	-0,5415	1,0000															
18:2t	-0,1921	0,1371	0,1796	0,4694	0,1669	0,1888	0,5130	-0,2530	0,4565	-0,4800	0,0742	-0,0883	1,0000														
C18:2	-0,5523	-0,2456	-0,1935	0,4777	-0,1436	0,2892	0,4679	-0,5679	0,3359	-0,5266	0,2351	-0,2898	0,3231	1,0000													
C20:0	0,3848	0,0421	-0,1830	<i>-0,8263</i>	-0,0838	-0,6597	<i>-0,7965</i>	0,7545	-0,5925	0,8069	-0,3922	0,2805	-0,3764	-0,3045	1,0000												
C18:3	-0,5507	-0,2103	-0,1256	0,6458	-0,1112	0,4525	0,6386	-0,6684	0,4697	-0,6725	0,2770	-0,3043	0,5389	0,7723	-0,6343	1,0000											
CLA	-0,4633	-0,3071	-0,1458	0,5462	-0,2053	0,2185	0,6270	-0,4142	0,6291	-0,5800	0,1403	0,0913	0,5228	0,3611	-0,5712	0,5599	1,0000										
C21:0	-0,3151	-0,0612	-0,0934	0,3002	0,0841	0,2996	0,1839	-0,5532	0,1697	-0,3779	0,5130	-0,4705	-0,1220	0,5804	-0,3032	0,4852	0,1025	1,0000									
C20:4	-0,5147	-0,1841	-0,1558	0,5439	-0,0718	0,4332	0,5026	<i>-0,7032</i>	0,3687	-0,5819	0,3808	-0,4175	0,1531	0,8177	-0,4927	0,7509	0,2710	0,7732	1,0000								
C20:5	-0,5605	-0,2158	-0,1379	0,6356	-0,1046	0,4846	0,6111	<i>-0,7546</i>	0,4298	-0,6591	0,3556	-0,3883	0,2294	0,8273	-0,5872	0,8180	0,3877	0,7018	0,9730	1,0000							
C22:5	-0,5561	-0,1954	-0,1300	0,6124	-0,0853	0,4585	0,6066	<i>-0,7337</i>	0,4575	-0,6596	0,3267	-0,3634	0,2723	0,8302	-0,5812	0,8205	0,4082	0,6818	0,9641	0,9865	1,0000						
C22:6	-0,5892	-0,2553	-0,1650	0,6631	-0,1405	0,4460	0,6513	<i>-0,7257</i>	0,5713	-0,6671	0,3401	-0,3135	0,2853	0,7734	-0,6160	0,7684	0,4352	0,6461	0,9246	0,9538	0,9581	1,0000					
n-6/n-3	0,1271	-0,0098	-0,0881	-0,4083	-0,0243	-0,3922	-0,4336	0,3826	-0,3132	0,3949	-0,0862	0,0878	-0,2101	0,1501	0,6642	-0,4220	-0,3618	-0,1086	-0,2295	-0,3109	-0,3273	-0,3481	1,0000				
PUFA/SFA	-0,6439	-0,3203	-0,2261	0,6350	-0,1976	0,3548	0,6441	-0,6924	0,5822	-0,6997	0,3362	-0,2229	0,3990	0,9067	-0,5603	0,8717	0,5832	0,6052	0,8938	0,9276	0,9356	0,9119	-0,1946	1,0000			
h/H	-0,2588	-0,5950	<i>-0,7006</i>	-0,3798	<i>-0,7102</i>	<i>-0,8065</i>	-0,1770	0,4025	0,1270	0,3170	-0,3143	0,8195	-0,0845	0,0708	0,3546	-0,0796	0,1081	-0,1695	-0,0911	-0,0975	-0,0691	-0,0469	0,2345	0,0780	1,0000		
AI	0,4580	0,7528	0,8338	0,1738	0,8080	0,6205	-0,0120	-0,2215	-0,2837	-0,1322	0,1581	<i>-0,7085</i>	-0,0034	-0,2341	-0,1289	-0,1665	-0,2771	0,0039	-0,1250	-0,1382	-0,1475	-0,1830	-0,0722	-0,2993	<i>-0,9102</i>	1,0000	
TI	0,5778	0,4272	0,3353	-0,5275	0,3944	-0,0683	-0,6354	0,5314	-0,6386	0,6506	-0,0894	-0,3118	-0,3917	-0,6149	0,5301	<i>-0,7263</i>	-0,6218	-0,3552	-0,6130	-0,6802	-0,6943	<i>-0,7009</i>	0,3432	<i>-0,7942</i>	-0,4292	0,6264	1,0000

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Chapter 2

Effect of slaughter age and rearing season on
Bergamasca light lambs

Slaughter performance, carcass and meat quality of Bergamasca light lambs as affected by slaughter age

Abstract

The present study aims to evaluate the effect of slaughter age (40 vs. 60 days) on the slaughter performance, carcass and meat quality (including fatty acid (FA) composition of intramuscular (IM) and subcutaneous (SC) fat) of Bergamasca lambs reared according to the traditional transhumant system adopted in Central Italy. Twenty-two male single born lambs were reared with dams on pastures, fed with maternal milk and supplemented with hay and concentrate from the 20th day to the slaughter. The increase of the slaughter age from 40 to 60 days resulted in an improvement in the carcass weight, lower dressing percentage, a higher proportion of non-carcass components and leg commercial cut, a lower content of bones and a better lean/bone ratio. Furthermore, older lambs had lower drip loss with increase of meat storage period compared to meat of younger lambs. While meat chemical composition did not differ between lambs of the two slaughter ages, meat colour was influenced both by slaughter age and storage period. FA composition of IM fat was slightly influenced by slaughter age, while SC fat of lambs slaughtered at 40 days of age showed better FA profile in terms of lower content of saturated fatty acids (SFA) and higher content of monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), n-6 and n-3 PUFA. Furthermore, SC fat of lambs slaughtered at 40 days of age had better PUFA/SFA and hypocholesterolemic/hypercholesterolemic ratio and lower atherogenic and thrombogenic indices more favourable for human health. For the light lamb production in the traditional rearing system, instead of 40 days, farmers should consider 60 days as optimal slaughter age for Bergamasca lambs to produce slightly heavier carcasses without compromising the quality of meat.

Keywords: light lamb, slaughter age, carcass trait, meat quality.

1. Introduction

Traditional lamb production in Mediterranean countries is based on light lambs slaughtered at early ages (between 30 and 60 days) just after the weaning or short fattening period (Juárez et al., 2009; Santos-Silva et al., 2002b). Specific carcasses (weight up to 13 kg) are characterised by pale pink colour, reduced amount of fat and good flavour (Beriaín et al., 2000) compared to heavier carcasses produced in other production systems and countries (Ekiz et al., 2013; Hajji et al., 2016; Jacques et al., 2011; Lind et al., 2009; Piasentier, 2003; Priolo et al., 2002).

In recent years, the demand for lean carcasses is growing due to the awareness of consumers of healthy meat with accent on quantity and quality of fat (Sendim et al., 1997). Scientific research and nutritional guidelines recommend not only a reduction in total fat intake (with accent on saturated fatty acids (SFA)) but also high consumption of polyunsaturated fatty acids (PUFA) and especially n-3 PUFA (Calder and Yaqoob, 2009; World Health Organisation, 2003).

Age and weight at slaughter are among the main factors affecting lamb meat quality at both levels, carcass and meat. Usually greater weight implies a higher age, except when feed is manipulated or the animal has periods of strong alimentary restrictions (Guerrero et al., 2013). Although, higher slaughter age of lambs results in heavier carcasses, greater levels of adiposity, as well as better carcass conformation (D'Alessandro et al., 2013; Juárez et al., 2009) it may also result in higher amount of intramuscular fat (Abdullah and Qudsieh, 2009; della Malva et al., 2016; Pérez et al., 2002). Furthermore, lambs slaughtered at higher age can display fatty acid (FA) composition of meat less favourable for human health as a result of higher content of SFA, lower content of PUFA and n-3 PUFA as well as higher atherogenic (AI) and thrombogenic (TI) indices (Cifuni et al., 2000; Marino et al., 2008; Santos-Silva et al., 2002b).

Those aspects are of the utmost importance for the definition of strategies for enhancing the production, taking the market demand into consideration as well. In this sense, there are significant gaps of knowledge for lambs produced under the quality labels, such as "Agnello del Centro Italia" PGI (European Union, Commission

Regulation No. 475/2013), in which, together with some other breeds, Bergamasca is used.

Bergamasca sheep is autochthonous Italian breed which originates from Lombardy region (northwest Italy) traditionally raised in transhumant systems (Piasentier, 2003). Nowadays, it is raised principally for meat in most parts of continental Italy (Bigi and Zanon, 2008) and often used for cross-breeding with other breeds to improve meat yield. Males and females can reach adult weights of 105 kg and 82 kg, respectively. In Lombardy region, Bergamasca is still the most important breed used to produce castrate, heavy and light lambs as traditional products of transhumant management (Piasentier et al., 2003).

In Marche region (central Italy), lamb production is mainly based on extensive grazing and transhumance is still of major importance. In summer, most of the flocks graze upland pastures. Starting from autumn, the sheep are progressively transferred to lowlands where up to the spring time the main forage resources are Lucerne meadows, but green cereals, crop residues, marginal lands and riverbanks are sometimes also used (Caballero et al., 2009; D'Ottavio and Santilocchi, 2014). Mainly, lamb production is performed in lowlands for Easter and Christmas markets when meat is traditionally consumed in Italy (Cifuni et al., 2000). Lambs are reared on pastures with ewes until they reach the optimal slaughter weight that, according to local practices, is obtained starting from a period of 40 days of age. Lambs are not weaned so their diet is mostly based on milk while later (from 20 days on) they are supplemented with concentrate and/or hay when needed.

Although the effect of slaughter age on quality of light lamb meat of some other Italian breeds such as Altamura (della Malva et al., 2016; Marino et al., 2008), Apulian (Cifuni et al., 2000), Leccese (D'Alessandro et al., 2013), Trimeticchio (Marino et al., 2008) and Italian Merino (Oriani et al., 2005) has been studied, there is no information about such effect on lamb meat quality of Bergamasca breed.

The aim of this study was to evaluate the effect of slaughter age on slaughter performance, carcass traits and meat quality including fatty acid composition of intramuscular (IM) and subcutaneous (SC) fat of

Bergamasca light lambs reared in transhumant system traditional for Marche region. The effect of storage period (0, 3 and 6 days) on meat colour parameters was also investigated.

2. Materials and methods

2.1. *Experimental design, diet and animal management*

The experiment was carried out from September to October 2015 in Marche region (Central Italy) under the usual conditions for rearing and management of the transhumant sheep system, characteristic for this region. Twenty-two male, single born Bergamasca lambs were included into the study. All animals stayed with their dams on alfalfa dominated-grasslands (10.3 MJ ME kg⁻¹ dry matter (DM), 15.7% crude protein DM, 28.49 % crude fiber DM), and suckled their dams throughout the whole experimental period. Dams grazed and had free access to alfalfa hay (11.8 MJ ME kg⁻¹ DM; 15.2% crude protein DM; 22.3% crude fiber DM) and were supplemented by corn grain (0.5 kg head⁻¹ day⁻¹; 16.5 MJ ME kg⁻¹ DM, 7.9% crude protein DM, 4.98% crude fiber DM) while lambs were given corn grain *ad libitum* in creep feeders from 20 days of age and had access to hay. Chemical composition of collected feed samples was determined according to Martillotti et al. (1987). Lambs were slaughtered at two different ages: eleven lambs at the average of 40 days while the other eleven lambs at the average of 60 days. The lambs grouped per slaughter age did not differ in birth weight, weight at 20 and 40 days of age as well as in average daily gain (ADG).

2.2. *Slaughter procedure and assessment of carcass traits*

To obtain pre-slaughter weight (PSW), lambs were weighed on the farm and soon after transferred to commercial slaughterhouse where they were stunned and slaughtered by cutting the jugular vein. After the slaughter, non-carcass components, namely skin, head, feet, pluck (heart, lungs, liver and spleen) and digestive tract, were removed and weighed. After chilling at 4 °C for 24 hours, cold carcass weight was recorded and dressing percentages were calculated. The right side of the carcass was dissected into three main commercial cuts: shoulder, whole

loin with flank and leg. The weight of each commercial cut was recorded and expressed as proportion of half carcass weight. The steak of the muscle *longissimus dorsi* (LD) between the 1st and 6th lumbar vertebrae was then dissected into its main tissue components (lean, fat and bone) as a possible index of the carcass tissues' proportion. The lean/bone and lean/fat ratios were determined.

2.3. Meat quality parameters

The pH of the *longissimus thoracis* muscle (between the 10th and 13th thoracic vertebrae) was measured 45 minutes and 24 hours (final pH) *post mortem* using a portable pH meter (Eutech Instruments, mod. XS pH 110, Singapore) equipped with a penetrating glass electrode.

Drip loss and cooking loss were determined on LD muscle according to ASPA (1996). In order to measure the drip loss, meat samples were weighed and wrapped in polyethylene bags. After 24 hours of storage period at 4 °C, the samples were gently dried with paper towels, and reweighed. This procedure was done in two replications and repeated for the 3rd and the 6th day of storage. For cooking loss determination, meat samples were firstly weighed, and then placed in polyethylene bags and cooked in a water bath until they reached internal temperature of 65 °C. Bags with cooked samples were then cooled under cold running water for 30 minutes and then the cooked samples were removed from their respective bags, dried with paper towels and reweighed.

The colorimetric indices (lightness, L*; redness, a*; yellowness, b*) of the LD muscle were performed on fresh meat and after 3 and 6 days of storage period using a Minolta CR 200 with illuminant D 65 as the light source. During the storage period, samples were vacuum packed and kept at 4 °C and their colour was evaluated after 40 minutes of blooming. Chroma (C*, the square root of (a*² + b*²)) and hue angle (H°, $\tan^{-1}(b^*/a^*)$) were also calculated as well as colour difference (ΔE^* , $[(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]$) where Δ represents difference between colour parameters measured on different days of storage (Iacurto, 2005). On each meat sample, three colour measurements and calculations were performed for each parameter and reported as mean value. After colour measurements were performed, meat samples were

dried by lyophilisation (DM), ground and analysed for chemical determination of crude protein (Kjeldhal nitrogen x 6.25), fat (extraction with petroleum ether) and ash (incineration in muffle furnace at 550 °C) content. Analyses were performed in duplicate for each sample and investigated parameter and results were corrected for moisture content.

2.4. Fatty acid analysis

Fatty acid (FA) composition was performed on lyophilised LD muscle (intramuscular, IM) and subcutaneous (SC) fat samples. Total lipids from the samples were extracted by Soxtec™ system with the use of petroleum ether. Fatty acids methyl esters (FAME) were prepared by transmethylation, using KOH 2 mol/L in methanol and n-heptane, according to method 5509 of the ISO. The individual FAME were analysed on a gas chromatograph (HRGC MEGA 2 series, Fisons Instruments, Milano, Italy) equipped with a Rt®-2560 column (L = 100 m, ID = 0.25 mm and df = 0.2 µm, RESTEK, PA, USA), a flame-ionization detector and He as carrier gas. Identification of fatty acids was based on the comparison of retention time with a 37-component FAME standard (Supelco™) and Linolenic acid methyl ester isomer mix (Supelco™). The concentration of individual FAME was expressed as the percentage of total FAME identified and grouped as follows: saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA). PUFA/SFA and n-6/n-3 ratios were determined as well as atherogenic (AI) and thrombogenic (TI) indices (Ulbricht and Southgate, 1991) and hypocholesterolemic/hypercholesterolemic ratio (h/H; Fernández et al., 2007).

2.5. Statistical analysis

Data were analysed using JMP® software (JMP, Version 10. SAS Institute Inc., Cary, NC, 1989-2007). Student's t-test was utilized to evaluate effect of two slaughter ages on all parameters except for colour. Colour parameters were examined by two-way analysis of

variance (ANOVA). The model with fixed effects involved slaughter age (two levels), storage periods (three levels) and their interaction.

3. Results and discussion

The effect of slaughter age on carcass traits and non-carcass components is presented in Table 1. As expected, older lambs had higher pre-slaughter and carcass weight ($P < 0.001$ and $P < 0.05$, respectively). According to EU carcass classification system, the average carcass weight obtained in the two age categories can be classified as class C for light lambs (10.1 – 13 kg; EU, 1994).

Table 1
Carcass traits and non-carcass components of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
PSW (kg)	19.84 \pm 3.07	25.95 \pm 3.34	***
Cold carcass weight (kg)	10.36 \pm 1.75	12.44 \pm 2.14	*
Cold dressing (%)	52.16 \pm 2.50	47.68 \pm 2.65	***
<i>Proportion (%) on PSW</i>			
Head	2.63 \pm 0.23	3.92 \pm 0.32	***
Skin	9.09 \pm 0.60	11.54 \pm 0.85	***
Feet	1.01 \pm 0.11	0.83 \pm 0.10	**
Pluck	5.39 \pm 0.93	7.69 \pm 0.44	***
Full digestive tract	13.86 \pm 2.30	17.96 \pm 2.94	***

PSW: pre-slaughter weight.

* $P < 0.05$.

** $P < 0.01$.

*** $P < 0.001$.

In agreement with previous studies (Cifuni et al., 2000; Morbidini et al., 1987), slaughter age significantly influenced dressing percentages of light lambs with decrease ($P < 0.001$) at higher slaughter ages. Skapetas et al. (2006) reported the highest dressing percentages for lambs slaughtered at 45 days, with a significant decrease at 60 days and a further increase at 75 and 90 days of age. Results were explained by the highest growth rate of the lambs at 45 days compared to those at 60 days when the weaning stress caused decrease in weight gains.

According to our results, D'Alessandro et al. (2013) found significant differences in carcass weight between Leccese lambs slaughtered at 45 and 60 days of age, but no variation in dressing percentages were reported. Dressing percentage of lambs slaughtered at 60 days of age was similar to that (46.3%) reported for heavy Bergamasca lambs (Piasentier et al., 2001). However, dressing percentages of lambs from both age groups were much lower than those reported for Leccese breed slaughtered at 45 and 60 days (D'Alessandro et al., 2013) or Apulian lambs slaughtered at 45 and 90 days of age (Cifuni et al., 2000). Regarding non-carcass components, slaughter age influenced all investigated parameters (Table 1). Lambs slaughtered at 60 days of age had higher ($P < 0.001$) proportions of head, skin, pluck and full digestive tract and lower ($P < 0.01$) proportion of feet than lambs slaughtered at 40 days of age. As expected, older lambs had higher ($P < 0.01$) half carcass weight than the younger (Table 2). Furthermore, lambs slaughtered at 60 days of age had higher ($P < 0.01$) proportion of leg than lambs slaughtered at 40 days of age while other commercial cuts did not differ between lambs of two slaughter ages.

Table 2
Half carcass joints and tissue proportions obtained by dissection of steak of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
CHCW (kg)	4.80 \pm 0.85	6.06 \pm 1.07	**
<i>Proportions of commercial cuts (%)</i>			
Shoulder	19.81 \pm 0.88	19.53 \pm 1.36	NS
Whole loin with flank	44.69 \pm 1.71	42.65 \pm 2.84	NS
Leg	35.49 \pm 1.06	37.82 \pm 1.85	**
<i>Tissue proportion of steak (%)</i>			
Lean	42.17 \pm 4.39	43.77 \pm 3.19	NS
Fat	27.53 \pm 7.18	31.55 \pm 4.46	NS
Bone	29.55 \pm 5.43	24.16 \pm 3.19	*
Lean/Fat ratio	1.72 \pm 0.88	1.42 \pm 0.28	NS
Lean/Bone ratio	1.48 \pm 0.34	1.84 \pm 0.27	*

CHCW: cold half carcass weight

NS: not significant, $P > 0.05$.

* $P < 0.05$.

** $P < 0.01$.

The percentage of commercial leg cut had a significant difference due to age as revealed also by D'Alessandro et al. (2013). However, our results are opposite to those indicated by the aforementioned author. Indeed, in the study of D'Alessandro et al. (2013) the proportion of leg was higher for lambs slaughtered at 45 days compared to those slaughtered at 60 days of age.

From the dissection of the steak, lambs slaughtered at 40 days showed higher ($P < 0.05$, Table 2) proportion of bone. This can be explained by the fact that skeleton growth occurs earlier than muscle and fat growth (Taylor et al., 1989). Thus, with animal growth, proportion of bones decreases, followed by muscle proportion, while fat proportions increase in their carcasses (Irshad et al., 2013). According to our results, higher proportion of bone and lower of fat in leg joint were reported for younger lambs in study of Cifuni et al. (2000). The carcass value is influenced by the lean/bone ratio where higher ratio is better since it equates to more lean meat and better carcass conformation (Irshad et al., 2013). Lambs slaughtered at 60 days of age showed higher ($P < 0.05$) and therefore better lean/bone ratio while lean/fat ratio did not differ between slaughter age groups.

The age at slaughter did not affect meat pH values at 45' and 24 h post-mortem (Table 3). The results are in accordance with other studies where obtained pH values did not differ between light lambs slaughtered at different ages/weights (D'Alessandro et al., 2013; della Malva et al., 2016; Juárez et al., 2009). The final pH values are within the normal ranges (Tejeda et al., 2008) and are similar to values reported for light lambs by other authors (D'Alessandro et al., 2013; della Malva et al., 2016; Juárez et al., 2009; Mazzone et al., 2010).

No differences could be evidenced between the lambs slaughtered at 40 and 60 days of age in drip loss measured after one day of meat storage (Table 3). The values were lower than those reported for Appenninica light lambs (Mazzone et al., 2010). The differences in drip loss between lambs of two slaughter ages started to occur after the 3rd day and continued through the 6th day of storage with meat of younger lambs having higher ($P < 0.001$) drip loss than meat from older lambs. Results

are in agreement with Russo et al. (2003) who reported better water-holding capacity for heavier carcasses due to lower drip loss after the 2nd day of storage.

Table 3

pH, drip loss and cooking loss of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
<i>pH</i>			
45'	6.41 \pm 0.14	6.54 \pm 0.35	NS
24 h	5.71 \pm 0.11	5.69 \pm 0.14	NS
<i>Drip loss (%)</i>			
1 day	0.77 \pm 0.17	1.01 \pm 0.53	NS
3 day	6.16 \pm 1.41	3.69 \pm 0.97	***
6 day	9.36 \pm 1.45	5.73 \pm 1.16	***
<i>Cooking loss (%)</i>	12.22 \pm 3.85	10.53 \pm 4.94	NS

NS: not significant, $P > 0.05$.

*** $P < 0.001$.

Cooking loss did not differ between lambs slaughtered at 40 and 60 days of age (Table 3) and it was much lower than that reported for light Appenninica lambs of similar slaughter age (Mazzone et al., 2010). On the contrary, although another method for cooking loss determination was used, Russo et al. (2003) reported increase of cooking loss with increase of carcass weight. Absence of differences in cooking loss between lambs of the two slaughter age groups in the present study could be explained with the fact that, contrary to Russo et al. (2003), differences in carcass weights between groups of light lambs were much lower.

Meat chemical composition of lambs slaughtered at 40 and 60 days of age is reported in Table 4. Obtained values are comparable with those reported for light lambs of different Italian breeds (D'Alessandro et al., 2013; della Malva et al., 2016; Mazzone et al., 2010). Taking into consideration intramuscular fat content, meat analysed in the present study is of good quality, assuming that 2-3% intramuscular fat ensures the desirable organoleptic properties of meat (Wood, 1990). There were no significant differences for moisture, crude protein, fat and ash of LD muscle in relation to the slaughter age.

These results are in accordance with other studies highlighting that meat chemical composition was not affected by slaughter age (D'Alessandro et al., 2013; Marino et al., 2008) or carcass weight (Russo et al., 2003). In general, higher values of intramuscular fat are reported for older lambs (Abdullah and Qudsieh, 2009; della Malva et al., 2016; Juárez et al., 2009; Pérez et al., 2002).

Table 4
Chemical composition (%) of *longissimus dorsi* muscle of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
Moisture	73.45 \pm 1.35	74.55 \pm 1.56	NS
Crude Protein	19.98 \pm 0.50	19.68 \pm 0.62	NS
Fat	2.70 \pm 1.11	2.15 \pm 0.91	NS
Ash	1.31 \pm 0.07	1.39 \pm 0.25	NS

NS: not significant, $P > 0.05$.

Table 5 reports the colour parameters of the lamb meat considering the effect of age, storage time and their interaction. Differing from D'Alessandro et al. (2013), who observed no effect of slaughter age on any meat colour parameter of light lambs, in the present study some colour differences were recorded. Contrary to different authors (Abdullah and Qudsieh, 2009; della Malva et al., 2016; Juárez et al., 2009; Santos-Silva et al., 2002a; Sañudo et al., 1996), in the present study the increase of age/weight of lambs did not result in darker meat. Indeed, no differences were found in the lightness (L^*), yellowness (b^*) as well as in Hue and Chroma between meat of lambs slaughtered at 40 and 60 days of age. Observed higher ($P < 0.01$) value of redness (a^*) in meat of older/heavier lambs are in accordance with other studies (della Malva et al., 2016; Juárez et al., 2009; Sañudo et al., 1996) and could be explained by higher myoglobin content in meat of older lambs (Juárez et al., 2009). Furthermore, colour differences of meat from lambs slaughtered at different ages could be due to higher consumption of milk by the younger lambs compared to the older ones that utilised proportionally more concentrate until the slaughter (Sañudo et al., 1996).

Table 5

Effect of slaughter age (SA), storage period (SP) and SA x SP interaction on colour parameters of Bergamasca lambs meat (mean values).

	Slaughter age (d)		Storage period (d)			Effect			
	40	60	0	3	6	SEM	SA	SP	SA x SP
L*	42.61	41.85	41.31	42.43	42.95	0.33	NS	NS	NS
a*	19	20.2	19.18	19.81	19.82	0.21	**	NS	NS
b*	4.29	4.63	3.37 ^b	5.05 ^a	4.98 ^a	0.16	NS	***	NS
Hue	12.8	12.84	9.99 ^b	14.37 ^a	14.10 ^a	0.47	NS	***	NS
Chroma	39.02	43.51	31.16 ^b	46.78 ^a	45.85 ^a	1.57	NS	***	NS

SEM: standard error of the mean.

^{a, b} Within the same row, means with different letters differ significantly.

NS: not significant, $P > 0.05$.

** $P < 0.01$

*** $P < 0.001$

Storage period influenced meat colour. Indeed, after 3 and 6 days of storage the samples had higher values ($P < 0.001$, Table 5) for b*, Hue and Chroma compared to fresh meat while no interactions of slaughter age and storage period for any colour parameter were evident. In agreement with our results, in the study conducted by Abdullah and Qudsieh (2009), parameter L* of LD muscle was not influenced by the storage period. Contrary to what we obtained, they reported no effect of storage period also on b*, Hue and Chroma values of lamb meat, while storage period increased its redness (a*).

Total colour difference (ΔE^*) is a parameter used to understand if any change in meat colour between two measurements is detected by human eye. In our study, there was no difference between the meat of the lambs slaughtered at different ages (Table 6). Variations between colour of fresh meat and meat colour after 3 and 6 days of storage were quite evident for human eye while the meat colour change from the 3rd to the 6th day of storage was little distinguishable (Iacurto, 2005).

Table 6

Changes of meat colour (ΔE^*) of Bergamasca lambs slaughtered at 40 and 60 days of age.

	Age (d)		Significance
	40	60	
0-3 days	3.30	3.53	NS
0-6 days	3.39	3.77	NS
3-6 days	1.99	2.55	NS

NS: not significant, $P > 0.05$.

The FA composition of IM and SC fat is presented in Table 7 and Table 8, respectively. In general, IM FA composition of this study was similar to those reported by other authors (D'Alessandro et al., 2015; Cifuni et al., 2000; Oriani et al., 2005) for light lambs with high content of milk in the diet. Total SFA were the most abundant FA followed by MUFA and PUFA, both in IM and SC fat.

Slaughter age slightly influenced FA composition of LD muscle with lambs slaughtered at 60 days of age having higher ($P < 0.05$) value of tridecylic (C13:0) and palmitic (C16:0) SFA than lambs slaughtered at 40 days of age (Table 7). In agreement with other authors (Cifuni et al., 2000; D'Alessandro et al., 2015; della Malva et al., 2016; Santos-Silva et al., 2002b), our results confirmed higher amount of C16:0 in IM fat of older/heavier lambs. The same authors reported better fatty acid profile for meat of younger (less heavy) lambs as an increase in proportion of SFA occurred with lamb growth. The reported trend for IM fat was not evident in the present study as slaughter age did not affect any other single SFA, MUFA or PUFA in lamb IM fat. Consequently, nutritional indices of the meat such as ratios PUFA/SFA, n-6/n-3 and h/H as well as AI and TI were similar between Bergamasca lambs slaughtered at 40 and 60 days of age.

Regarding FA composition of SC fat, effect of slaughter age was much more evident (Table 8).

Due to paucity of published data on effect of slaughter age on SC fat of lambs, trend of changes was compared with those reported for IM fat. In accordance with the results reported for IM fat by different authors (Cifuni et al., 2000; Marino et al., 2008; Santos-Silva et al., 2002b), SC fat of lambs slaughtered at 40 days of age showed better FA composition in terms of lower ($P < 0.01$) content of SFA and higher ($P < 0.01$) content of MUFA than lambs slaughtered at 60 days of age. In contrast to our study, Juárez et al. (2009) reported FA composition of SC fat of suckling and light lambs where weight/age influenced just some of single SFA while total SFA, MUFA and PUFA, as well as PUFA/SFA did not differ between lambs of the two slaughter weights.

Table 7

Fatty acid composition (%) of *longissimus dorsi* muscle of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
C10:0	0.64 \pm 0.24	0.75 \pm 0.18	NS
C11:0	0.04 \pm 0.01	0.04 \pm 0.01	NS
C12:0	1.53 \pm 0.57	1.84 \pm 0.50	NS
C13:0	0.10 \pm 0.03	0.12 \pm 0.02	**
C14:0	9.78 \pm 1.90	10.77 \pm 2.05	NS
C15:0	0.97 \pm 0.17	1.01 \pm 0.15	NS
C16:0	24.43 \pm 1.84	26.09 \pm 0.90	**
C17:0	1.20 \pm 0.17	1.12 \pm 0.10	NS
C18:0	11.98 \pm 1.72	11.18 \pm 1.79	NS
C20:0	0.10 \pm 0.04	0.08 \pm 0.03	NS
C21:0	0.15 \pm 0.07	0.16 \pm 0.03	NS
C14:1	0.29 \pm 0.11	0.34 \pm 0.11	NS
C16:1 Δ 9cis	1.78 \pm 0.40	1.90 \pm 0.32	NS
C17:1	0.55 \pm 0.14	0.55 \pm 0.10	NS
Σ 18:1trans ¹	3.31 \pm 1.59	3.05 \pm 1.21	NS
Σ 18:1cis ¹	30.14 \pm 3.68	28.12 \pm 3.29	NS
Σ 18:2trans ¹	1.58 \pm 0.26	1.65 \pm 0.27	NS
C18:2n-6	4.2 \pm 1.36	3.56 \pm 0.83	NS
C18:2 cis9 trans 11; CLA	1.50 \pm 0.30	1.64 \pm 0.40	NS
C18:3n-3	1.57 \pm 0.32	1.53 \pm 0.24	NS
C20:2n-6	0.09 \pm 0.03	0.07 \pm 0.01	NS
C20:4n-6	0.53 \pm 0.39	0.63 \pm 0.29	NS
C20:5n-3	0.26 \pm 0.19	0.29 \pm 0.12	NS
C22:5n-3	0.34 \pm 0.23	0.36 \pm 0.14	NS
C22:6n-3	0.14 \pm 0.10	0.14 \pm 0.07	NS
OFA	2.79 \pm 0.26	2.99 \pm 0.19	NS
SFA	50.91 \pm 3.89	53.17 \pm 2.35	NS
MUFA	36.07 \pm 3.20	33.96 \pm 2.83	NS
PUFA	10.22 \pm 2.46	9.88 \pm 1.30	NS
n-6 PUFA	4.82 \pm 1.67	4.27 \pm 1.01	NS
n-3 PUFA	2.32 \pm 0.82	2.33 \pm 0.50	NS
n-6/n-3 PUFA	2.13 \pm 0.59	1.88 \pm 0.53	NS
PUFA/SFA	0.20 \pm 0.06	0.19 \pm 0.02	NS
h/H	1.11 \pm 0.21	0.95 \pm 0.15	NS
AI	1.53 \pm 0.36	1.77 \pm 0.32	NS
TI	1.68 \pm 0.29	1.81 \pm 0.15	NS

¹ Σ C18:1trans includes sum of C18:1 trans isomers; Σ C18:1cis includes C18:1 Δ 9c + Δ 11c;

Σ C18:2trans isomers includes 18:2 c,t + t,c + t,t.

NS: not significant; P > 0.05.

* P < 0.05.

** P < 0.01.

*** P < 0.001.

OFA: other non-identified fatty acids

SFA = (C10:0 + C11:0 + C12:0 + C13:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C21:0).

MUFA = (C14:1 + C16:1 + \sum 18:1t + 18:1n-9).

PUFA = (C18:2n-6 + C18:3n-3 + C18:2c9t11 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3).

n-6 PUFA = (C18:2n-6 + C20:2n-6 + C20:4n-6).

n-3 PUFA = (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3).

h/H: the hypocholesterolemic/hypercholesterolemic ratio = [(C18:1n-9 + C18:2n-6 + C18:3n-3 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3)/(C14:0 + C16:0)].

AI: atherogenic index = (C12:0 + 4 x C14:0 + C16:0)/ [MUFA + PUFA (n-6) and (n-3)].

TI: thrombogenic index = (C14:0 + C16:0 + C18:0)/ [0.5 x MUFA + 0.5 x PUFA (n-6) + 3 x PUFA (n-3) + (n-3)/(n-6)].

In the present study, slaughter age affected the proportion of almost all investigated SFA. In particular, SC fat from lambs slaughtered at 60 days showed higher proportions of capric (C10:0; $P < 0.01$), lauric (C12:0; $P < 0.05$), tridecyllic ($P < 0.01$), myristic (C14:0; $P < 0.01$), pentadecanoic (C15:0; $P < 0.01$), palmitic ($P < 0.001$) and lower values of heptadecanoic (C17:0; $P < 0.01$) and stearic (C18:0; $P < 0.001$) FA than lambs slaughtered at 40 days of age.

It is reported that short chain SFA C12:0, C14:0 and C16:0 have high LDL (“bad”) cholesterol-raising effect (Siri-Tarino et al., 2010). Therefore, SC fat from Bergamasca lambs slaughtered at 60 days showed less favourable level of SFA than the SC fat of lambs slaughtered at 40 days of age.

Older lambs also had higher ($P < 0.01$) values of myristoleic (C14:1) and palmitoleic (C16:1) acid and lower ($P < 0.01$) value of $\Sigma 18:1$ cis (sum of oleic and cis-vaccenic acid) MUFA in SC fat than lambs slaughtered at 40 days of age.

Furthermore, slaughter age affected proportions of single PUFA in SC fat with lambs slaughtered at 60 days having higher ($P < 0.05$) values of C18:2 trans and C20:2 n-6 and lower values of linoleic (C18:2 n-6; $P < 0.001$) and linolenic (C18:3 n-3; $P < 0.05$) acid. Lower values of linoleic acid in SC fat of heavier lambs are in accordance with Juárez et al. (2009).

With increase of slaughter age, a decrease in total PUFA of both series n-6 ($P < 0.001$) and n-3 ($P < 0.05$) occurred. However, ratios between them (n-6/n-3) did not differ between Bergamasca lambs of the two slaughter ages and they were within the recommended value for human diet (less than 4) (Wood et al., 2003).

Lambs slaughtered at 40 days had SC fat with more desirable PUFA/SFA ratio as it was higher ($P < 0.01$) compared to lambs slaughtered at 60 days of age. However, the PUFA/SFA ratio of both slaughter age groups was low and did not reach the recommended value for human health (above 0.4) (Wood et al., 2003).

The overall changes in FA composition of SC fat with increase of slaughter age determined a decline ($P < 0.01$) in h/H affecting negatively the nutritional value of the meat. This finding is in accordance with the study of Santos-Silva et al. (2002b) for IM fat

where the lower h/H is reported for light lambs slaughtered at higher live weight.

SC fat of lambs slaughtered at 40 days of age had more favourable atherogenic (AI) and thrombogenic (TI) indices as they had lower values ($P < 0.01$) than the values observed for lambs slaughtered at 60 days of age.

Table 8

Fatty acid composition (%) of subcutaneous adipose tissue of Bergamasca lambs slaughtered at 40 and 60 days of age (mean \pm SD).

	Age (d)		Significance
	40	60	
C10:0	0.63 \pm 0.18	0.86 \pm 0.11	**
C11:0	0.03 \pm 0.01	0.04 \pm 0.01	**
C12:0	1.09 \pm 0.39	1.50 \pm 0.30	*
C13:0	0.06 \pm 0.02	0.08 \pm 0.02	**
C14:0	7.42 \pm 1.55	9.28 \pm 1.24	**
C15:0	0.72 \pm 0.18	0.90 \pm 0.09	**
C16:0	19.58 \pm 2.13	22.99 \pm 1.51	***
C17:0	1.73 \pm 0.17	1.51 \pm 0.06	**
C18:0	20.94 \pm 2.23	18.23 \pm 1.68	**
C20:0	0.17 \pm 0.03	0.17 \pm 0.05	NS
C21:0	0.09 \pm 0.03	0.10 \pm 0.01	NS
C14:1	0.10 \pm 0.02	0.14 \pm 0.02	**
C16:1 Δ 9cis	0.80 \pm 0.09	0.94 \pm 0.12	**
C17:1	0.42 \pm 0.06	0.39 \pm 0.06	NS
Σ 18:1trans ¹	2.84 \pm 2.14	3.82 \pm 1.79	NS
Σ 18:1cis ¹	32.74 \pm 2.93	28.69 \pm 2.51	**
Σ 18:2trans ¹	1.59 \pm 0.15	1.72 \pm 0.14	*
C18:2n-6	2.92 \pm 0.33	2.44 \pm 0.14	***
C18:2 cis9 trans 11; CLA	1.71 \pm 0.29	1.71 \pm 0.20	NS
C18:3n-3	1.57 \pm 0.26	1.37 \pm 0.11	*
C20:2n-6	0.03 \pm 0.01	0.04 \pm 0.01	*
C20:4n-6	0.05 \pm 0.01	0.05 \pm 0.02	NS
C20:5n-3	0.02 \pm 0.01	0.02 \pm 0.01	NS
C22:5n-3	0.12 \pm 0.02	0.13 \pm 0.03	NS
C22:6n-3	0.04 \pm 0.01	0.03 \pm 0.02	NS
OFA	2.59 \pm 0.26	2.83 \pm 0.10	*
SFA	52.46 \pm 2.60	55.68 \pm 2.33	**
MUFA	36.90 \pm 2.57	33.98 \pm 2.12	**
PUFA	8.05 \pm 0.73	7.51 \pm 0.44	NS
n-6 PUFA	3.00 \pm 0.33	2.52 \pm 0.13	***
n-3 PUFA	1.75 \pm 0.27	1.56 \pm 0.09	*
n-6/n-3 PUFA	1.73 \pm 0.15	1.62 \pm 0.09	NS
PUFA/SFA	0.15 \pm 0.02	0.13 \pm 0.01	**
h/H	1.42 \pm 0.29	1.03 \pm 0.15	**
AI	1.22 \pm 0.28	1.63 \pm 0.26	**
TI	1.87 \pm 0.19	2.15 \pm 0.19	**

¹ Σ C18:1trans includes sum of C18:1 trans isomers; Σ C18:1cis includes C18:1 Δ 9c + Δ 11c;

Σ C18:2trans isomers includes 18:2 c,t + t,c + t,t.

NS: not significant; P > 0.05.

* P < 0.05.

** P < 0.01.

*** P < 0.001.

OFA: other non-identified fatty acids

SFA = (C10:0 + C11:0 + C12:0 + C13:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C21:0).

MUFA = (C14:1 + C16:1 + \sum 18:1t + 18:1n-9).

PUFA = (C18:2n-6 + C18:3n-3 + C18:2c9t11 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3).

n-6 PUFA = (C18:2n-6 + C20:2n-6 + C20:4n-6).

n-3 PUFA = (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3).

h/H: the hypocholesterolemic/hypercholesterolemic ratio = [(C18:1n-9 + C18:2n-6 + C18:3n-3 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3)/(C14:0 + C16:0)].

AI: atherogenic index = (C12:0 + 4 x C14:0 + C16:0)/ [MUFA + PUFA (n-6) and (n-3)].

TI: thrombogenic index = (C14:0 + C16:0 + C18:0)/ [0.5 x MUFA + 0.5 x PUFA (n-6) + 3 x PUFA (n-3) + (n-3)/(n-6)].

4. Conclusions

The increase of the slaughter age in Bergamasca lamb from 40 to 60 days resulted in an improvement in the carcass weight, a higher proportion of commercial leg cut, a lower content of bones and a better lean/bone ratio. Furthermore, older lambs had lower drip loss with increase of meat storage period compared to meat of younger lambs. Meat colour was influenced both by slaughter age and storage period indicating higher a^* values for older lambs compared to younger ones and higher b^* , Hue and Chroma values for meat after storage period compared to the fresh ones. FA composition of IM fat was not strongly affected by slaughter age while SC fat from lambs slaughtered at 40 days of age showed better fatty acid profile in terms of lower content of SFA and higher content of MUFA, n-6 and n-3 FA. Furthermore, SC fat of lambs slaughtered at 40 days of age had better PUFA/SFA and H/h ratios and lower AI and TI indices more favourable for human health. For the light lamb production in the traditional rearing systems, instead of 40 days, farmers should consider 60 days as optimal slaughter age for Bergamasca lambs to produce slightly heavier carcasses without compromising the quality of meat.

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Effect of rearing season on growth, carcass traits and meat quality of Bergamasca light lambs

Abstract

Twenty-two male, single born lambs of Bergamasca breed were used to investigate the effect of the rearing season (winter *vs.* autumn) on growth performances, carcass traits and meat quality, including fatty acid (FA) profile of intramuscular (IM) and subcutaneous (SC) fat. All lambs were reared according to the traditional transhumant sheep system adopted in Central Italy. In each season, 11 unweaned lambs were reared with their dams on alfalfa-dominated pastures and had *ad libitum* access to concentrate in creep feeders from 20 days to slaughter performed at 60 days of age. Rearing season did not affect average daily gain, carcass weight, dressing percentage, cooking loss, colour and chemical composition of lamb meat. Lambs reared in winter had i) higher birth weight and drip loss after 6 days of meat storage and ii) lower proportion of pluck and pH values than lambs reared in autumn. FA composition from both IM and SC fat was influenced by the rearing season. In particular, lambs differed in some saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) in IM fat. On the contrary, SC fat of lambs reared in autumn showed better FA profile as a result of the higher content of total n-6, n-3 and polyunsaturated fatty acids (PUFA), as well as more favourable PUFA/SFA and n-6/n-3 ratios for human health.

Keywords: Bergamasca, rearing season, carcass traits, meat quality.

1. Introduction

Lamb meat production systems vary across the world from very extensive (e.g. Erasmus et al., 2016; Gallardo et al., 2014; Komprda et al., 2012; Ramírez-Retamal et al., 2013) to very intensive (e.g. Ekiz et al., 2009; Rodríguez et al., 2011; Vergara et al., 1999). The increasing importance of lamb meat quality for consumers has been an objective of many studies in the recent years (de Andrade et al., 2016; Font i Furnols et al., 2009; Hersleth et al., 2012; Montossi et al., 2013; Sepúlveda et al., 2011). When informed on the lamb rearing system, consumers would prefer meat from lambs of pasture-fed rather than of stall-fed mothers as it is perceived more natural and thus healthier (D'Alessandro et al., 2012). However, very often lambs produced in extensive systems are not competitive on the global market due to their limited yield and high production costs (Addis et al., 2013). For this reason, the “Agnello del Centro Italia” PGI quality label (European Union, Commission Regulation No. 475/2013) was created in Central Italy to promote and to protect local lamb meat market from foreign competition (Pauselli et al., 2009).

Consumers expect PGI products to have unique and consistent quality. Furthermore, they require meat which is safe, healthy and convenient (Nuernberg et al., 2008). This represents a challenge for breeders and other professionals involved in the production chain as lamb meat quality is influenced by many factors. Breed (Esenbuga et al., 2009; Juárez et al., 2009; Sañudo et al., 1997; Teixeira et al., 2005), slaughter age/weight (Cifuni et al., 2000; D'Alessandro et al., 2013; Oriani et al., 2005; Santos-Silva et al., 2002b) and sex (Alexandridis et al., 2014; Santos et al., 2007; Vergara et al., 1999) are the main. Some other factors could also influence lamb meat quality, such as rearing season (D'Alessandro et al., 2013; Mazzone et al., 2010). Although seasonal variation in meat quality is widely investigated for other species, e.g. cattle (Pestana et al., 2012; Sobczuk-Szul et al., 2013; Węglarz, 2010) there is paucity of data on the effect of rearing season on lamb meat quality, especially in extensive rearing systems as also highlighted by Mazzone et al. (2010).

In Central Italy, lamb production is mainly based on extensive grazing and transhumance is still of major importance. In summer, most of the flocks graze upland pastures. Starting from autumn the sheep are progressively transferred to lowlands where up to the spring time different forages are used to produce lambs for Easter and Christmas when meat is traditionally consumed in Italy.

The aim of this study was to evaluate the effect of rearing season (winter and autumn) on growth performances, carcass and meat traits (including fatty acid composition) of Bergamasca light lambs traditionally produced in Marche region under the “Agnello del Centro Italia” PGI quality label.

2. Materials and methods

2.1. *Experimental design, diet and animal management*

The trial was carried out in Marche region (Central Italy) on 22 Bergamasca lambs in winter and autumn to produce lambs for Easter and Christmas market. All the lambs had been reared according to the traditional transhumant sheep system. In the winter and autumn, 11 male single born lambs per season and from the same breeder were considered. In order to calculate average daily gain (ADG), lambs were weighed at birth and each 20 days until the slaughter. All animals were reared with their dams on cultivated pastures, suckled throughout the whole experimental period by their dams and had *ad libitum* access to concentrate in creep feeders from 20 days to slaughter. Particularly, lambs born in winter and slaughtered at Easter (winter lambs) were reared on alfalfa-dominated pastures (11.8 MJ ME kg⁻¹ dry matter (DM), 15.6% crude protein DM, 21.5% crude fibre DM) and were given concentrate (50% corn, 50% barley; 17.1 MJ ME kg⁻¹ DM, 7.7 % crude protein DM, 2.9 % crude fibre DM) while dams had free access to hay (9.5 MJ ME kg⁻¹ DM, 7.3 % crude protein DM, 31.3 % crude fibre DM) and were supplemented with corn grain (0.5 kg head⁻¹ day⁻¹; 17.1 MJ ME kg⁻¹ DM, 5.7% crude protein DM, 2.7% crude fibre DM). Conversely, lambs slaughtered at Christmas (autumn lambs) were reared on alfalfa dominated pastures (10.3 MJ ME kg⁻¹ DM, 15.7% crude protein DM, 28.49 % crude fibre DM) and were supplemented by

corn grain (16.5 MJ ME kg⁻¹ DM, 7.9% crude protein DM, 5.0 % crude fibre DM) while dams were supplemented with the same corn grain (0.5 kg head⁻¹ day) and had access to alfalfa hay (11.8 MJ ME kg⁻¹ DM; 15.2% crude protein DM; 22.3% crude fibre DM).

2.2. Slaughter procedure and assessment of carcass traits

In both rearing seasons, lambs were slaughtered at the average of 60 days of age. To obtain pre-slaughter weight (PSW) in both rearing season, lambs were firstly weighed on the farm and soon after transferred to the same commercial slaughterhouse where they were stunned and slaughtered by cutting the jugular vein. After slaughter, non-carcass components: skin, head, feet, pluck (heart, lungs, liver and spleen) and digestive tract were removed and weighed. Hot carcass weight was recorded and dressing percentages were expressed as hot carcass weight/PSW. Carcasses were left to chill at 4°C for 24 h after which samples of *longissimus dorsi* (LD) muscle and subcutaneous (SC) adipose tissue were taken from each half carcass between the 1st and the 6th lumbar vertebrae.

2.3. Meat quality parameters

Carcass pH was recorded in *longissimus thoracis* muscle (between the 10th and 13th thoracic vertebrae) 45 min (45') and 24 h (final pH) *post mortem* using a portable pH meter (Eutech Instruments, mod. XS pH 110, Singapore) equipped with a penetrating glass electrode. Other meat quality parameters were performed on LD muscle. Drip loss was determined according to the method of ASPA (1996) where meat samples were firstly weighed, wrapped in polyethylene bags and, after 24 h storage period at 4 °C, they were gently dried with paper towels and reweighed. This procedure was done in two replications and repeated for the 3rd and 6th day of storage.

For cooking loss determination, meat samples were firstly weighed, and then placed in polyethylene bags and cooked in a water bath until reaching internal temperature of 65 °C. Bags with cooked samples were then cooled under cold running water for 30 minutes and then the

cooked samples were removed from their respective bags, dried with paper towels and reweighed.

Colour on freshly cut surface of LD muscle samples was evaluated using the $L^* a^* b^*$ system and a Minolta colorimeter (Chroma Meter CR 200, Minolta Camera, Osaka, Japan). Chroma (C^* , the square root of $(a^{*2} + b^{*2})$) and hue angle (H° , $\tan^{-1}(b^*/a^*)$) were calculated (Iacurto, 2005). On each meat sample, three colour measurements and calculations were performed for each parameter and reported as mean value. Afterwards, samples were dried by lyophilisation (DM), and ground and crude protein (Kjeldahl, nitrogen \times 6.25), fat (extraction with petroleum ether) and ash (incineration in muffle furnace at 550 °C) content were determined. Analyses were performed in duplicate for each sample and investigated parameter and results were corrected for moisture content.

2.4. Fatty acid analysis

Fatty acid composition was performed on lyophilised LD muscle (intramuscular, IM) and subcutaneous (SC) fat samples. Total lipids from the samples were extracted by SoxtecTM system with the use of petroleum ether. Fatty acids methyl esters (FAME) were prepared by transmethylation, using KOH 2mol/L in methanol and n-heptane, according to method 5509 of the ISO. Chromatographic analysis of FAMEs was performed using HRGC MEGA 2 series gas chromatograph (Fisons Instruments, Milano, Italy) equipped with a Rt[®]-2560 column (L = 100 m, ID = 0.25 mm and df = 0.2 μ m, RESTEK, PA, USA), a flame-ionization detector and He as the carrier gas. Identification of fatty acids was based on the comparison of retention time with a 37-component FAME standard (SupelcoTM) and Linolenic acid methyl ester isomer mix (SupelcoTM). The concentration of individual FAME was expressed as a percentage of total FAME identified and grouped as follows: saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA). PUFA/SFA and n-6/n-3 ratios were determined as well as atherogenic (AI) and thrombogenic (TI) index (Ulbricht and Southgate, 1991) and hypocholesterolemic/hypercholesterolemic ratio (h/H; Fernández et al., 2007).

2.5. Statistical analysis

Data were analysed using JMP[®] software (JMP, Version 10. SAS Institute Inc., Cary, NC, 1989-2007). Student's t-test was utilized to evaluate effect of two rearing seasons on all parameters investigated in the study.

3. Results and discussion

Growth performances of Bergamasca lambs reared in two seasons are presented in Table 1. Lambs born in winter had higher ($P < 0.05$) birth weight than lambs born in autumn while they did not differ in live weights as well as in ADGs recorded in different periods. By contrast, in study of D'Alessandro et al. (2013), at the age of 60 days, Lecce lambs showed greatest final weight when slaughtered at Easter.

Table 1
Growth performances of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
Live Weight (kg)			
At birth	6.30 \pm 1.15	5.11 \pm 0.83	*
20 days	13.32 \pm 2.30	11.77 \pm 1.22	NS
40 days	19.47 \pm 3.66	19.59 \pm 2.35	NS
60 days (PSW)	26.07 \pm 4.16	25.95 \pm 3.34	NS
Average Daily Gain (g d ⁻¹)			
0-20 days	336 \pm 81.82	337 \pm 72.57	NS
20-40 days	293 \pm 100.08	373 \pm 63.01	NS
40-60 days	314 \pm 58.76	303 \pm 67.36	NS
0-60 days	323 \pm 64.46	337 \pm 61.93	NS

* $P < 0.05$.

PSW: pre-slaughter weight.

Table 2 summarises the effect of rearing season on carcass weight, dressing percentages and non-carcass components of Bergamasca lambs. No significant differences in average carcass weight were found between lambs of two rearing seasons, although carcass weights of lambs slaughtered at Christmas (autumn lambs) tended to be slightly higher than those of lambs slaughtered at Easter (winter lambs), even

though the differences were not significant ($P > 0.05$). The results are in accordance with Mazzone et al. (2010) who found the same trend for Appenninica suckling lambs. On the contrary, D'Alessandro et al. (2013) reported heavier carcasses for lambs slaughtered at Easter than Christmas. In accordance with results of D'Alessandro et al. (2013), rearing season did not affect dressing percentages of lambs in the present study. Regarding non-carcass components, lambs differed ($P < 0.001$) just in proportion of pluck that was higher for lambs slaughtered at Christmas.

Table 2
Slaughtering performance of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
Carcass weight (kg)	12.12 \pm 2.87	12.78 \pm 2.15	NS
Dressing (%)	49.51 \pm 2.59	48.98 \pm 2.53	NS
<i>Proportion (%) on PSW</i>			
Head	4.05 \pm 0.23	3.92 \pm 0.32	NS
Skin	10.96 \pm 0.76	11.54 \pm 0.85	NS
Feet	0.81 \pm 0.25	0.83 \pm 0.10	NS
Pluck	6.19 \pm 1.33	7.69 \pm 0.44	**
Full digestive tract	15.78 \pm 2.57	17.96 \pm 2.94	NS

NS: not significant.

** $P < 0.01$.

The physical characteristics of lamb meat are presented in Table 3. The pH at slaughter (45') and final pH were both affected ($P < 0.001$) by the rearing season, showing greater values in autumn compared to winter rearing season. These findings are partly in agreement with D'Alessandro et al (2013) as they did not find differences in pH at slaughter between lambs of two rearing season but they reported higher final pH for lambs reared in autumn and slaughtered at Christmas just as we did. In general, pH values (both at 45' and 24 h) of lambs reared in winter were much lower than those reported by other authors (D'Alessandro et al., 2013; della Malva et al., 2016; Mazzone et al., 2010) for some other Italian breeds while lambs reared in autumn had similar pH values to those reported by those authors. Furthermore, pH values at slaughter of lambs reared in winter were below the range (6.15

- 6.80) reported by the “Agnello del Centro Italia” PGI product specifications while lambs reared in autumn had pH values inside the reported range. However, lambs from both rearing seasons had acceptable final pH values that were within normal range (5.15 - 5.80) indicated by those specifications.

Lambs reared in winter or autumn did not differ in drip loss measured in meat samples after 1 or 3 days of storage (Table 3). The differences occurred after the 6th day of storage indicating higher ($P < 0.001$) drip loss for lambs reared in winter. Mazzone et al. (2010) reported opposite results where meat drip loss measured after 1 day of storage was higher for lambs reared in autumn.

In agreement with results of Mazzone et al. (2010), no differences could be found in cooking loss between lambs slaughtered in two seasons. However, the values were much lower (10.29 and 10.53 vs. 24.01) than those reported by Mazzone et al. (2010) for Appenninica suckling lambs slaughtered at the same age.

Table 3
Carcass pH, drip loss and cooking loss of the meat of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
<i>pH</i>			
45'	5.98 \pm 0.20	6.54 \pm 0.35	***
24h	5.29 \pm 0.16	5.69 \pm 0.14	***
<i>Drip loss (%)</i>			
1 day	0.88 \pm 0.23	1.01 \pm 0.53	NS
3 day	4.59 \pm 1.07	3.69 \pm 0.97	NS
6 day	8.85 \pm 1.55	5.73 \pm 1.16	***
<i>Cooking loss (%)</i>	10.29 \pm 3.40	10.53 \pm 4.94	NS

NS: not significant.

*** $P < 0.001$.

Table 4 presents the effect of rearing season on meat colour parameters. Meat colour is influenced by many factors, where final pH is one of the most important ones (Priolo et al., 2001).

Although they differed in pH, lambs showed similar colour parameters independent from the rearing season. On the contrary, Mazzone et al.

(2010) reported darker meat and higher Chroma (C*) values for autumn lambs.

It is reported that meat of unweaned lambs has high L* (40-49) and low a* (7-17) values of meat colour with b* value in range from 6 to 13 (D'Alessandro et al., 2013; Juárez et al., 2009, Russo et al., 2003; Santos-Silva et al, 2002a). L* values in the present study (42.88 and 41.11) were inside the reported range and were very similar to that (41.95) of Appenninica suckling lambs reported by Mazzone et al. (2010). However, a* values were higher (19.24 and 19.43) and b* lower (3.13 and 3.43) than previously mentioned range. In general, colour of Bergamasca lambs was similar to that reported for LD muscle of Istrian sheep lambs with similar slaughter age and weight (Vnučec et al. 2014) or to Sarda suckling lambs (Addis et al., 2013). Hue values of the present study (9.37 and 10.06) were much lower than values reported by Mazzone et al. (2010) and Russo et al. (2003) for Appenninica light lambs (17.34 and 24.10, respectively) while Chroma values were much higher (29.76 and 32.13, respectively) than the ones observed in those studies (14.83 and 19.68, respectively).

Table 4

Colour parameters of *longissimus dorsi* muscle of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean ± SD).

	Rearing season		Significance
	Winter	Autumn	
L*	42.88	41.11	NS
a*	19.24	19.43	NS
b*	3.13	3.43	NS
Hue°	9.37	10.06	NS
Chroma*	29.76	32.13	NS

NS: not significant.

The effect of rearing season on meat chemical composition of Bergamasca lambs is reported in Table 5. The values are within the range reported by other authors (Addis et al., 2013; della Malva et al., 2016; Marino et al., 2008; Vacca et al., 2008) for meat of light lambs. There were no statistical differences ($P > 0.05$) for the content of moisture, crude protein, fat and ash in the LD muscle between the lambs reared in winter and autumn. On the contrary, higher amount of fat in

the meat of lambs reared in autumn was reported by Mazzone et al. (2010). Authors attributed differences to a different feeding management as lambs reared in autumn were suckling their dams that were permanently pastured (better milk performances due to high fat content) while winter lambs received milk of dams that never acceded pasture. In agreement with our study, when lambs were reared with dams on pastures in both seasons, no differences were found in meat chemical composition of Leccese light lambs (D'Alessandro et al., 2013).

Table 5
Chemical composition of the *longissimus dorsi* muscle of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
Moisture (%)	75.08 \pm 1.31	74.55 \pm 1.56	NS
Crude Protein (%)	19.31 \pm 0.77	19.68 \pm 0.62	NS
Fat (%)	1.56 \pm 0.66	2.15 \pm 0.91	NS
Ash (%)	1.22 \pm 0.05	1.39 \pm 0.25	NS

NS: not significant.

The fatty acid composition of intramuscular fat (IM) is shown in Table 6. The rearing season had a significant effect on the levels of C12:0 ($P < 0.05$), C13:0 ($P < 0.01$), C15:0 ($P < 0.05$), C16:0 ($P < 0.01$) SFA. Higher concentrations of those acids were noted in the meat of lambs reared in autumn and slaughtered at Christmas. From the other side, meat of lambs reared in winter showed higher levels of C17:0 ($P < 0.001$), C18:0 ($P < 0.001$), C20:0 ($P < 0.01$) and C21:0 ($P < 0.001$). The C12:0, C14:0 and C16:0 SFA are known to have great LDL cholesterol-raising effect (Siri-Tarino et al., 2010). Therefore, meat of lambs reared in winter is characterised by more desirable levels of these SFA than meat of lambs reared in autumn. However, total SFA content did not differ between lambs reared in two seasons and values were similar to those reported for unweaned Grazalema Merino and Churra Leberijana light lambs (Juárez et al., 2009). The high levels of total SFA are common for the meat of unweaned lambs as their FA profile depends essentially on FA composition of ewes' milk that is characterised by prevalence of C16:0, C14:0, C18:0 and C10:0 SFA (Atti et al., 2006).

MUFA profiles showed higher levels of C14:1 ($P < 0.05$) and C16:1 ($P < 0.01$) in meat of lambs reared in autumn. No differences were observed in total MUFA between lambs reared in two rearing seasons. Furthermore, lambs from both rearing seasons showed similar PUFA profile and consequently did not differ in n-6, n-3 or total PUFA content. The PUFA/SFA together with n-6/n-3 ratio is a good indicator of the nutritional value of dietary fat (Ulbricht and Southgate, 1991). According to the current nutritional recommendations, the PUFA/SFA ratio in human diet should be above 0.4, and the n-6/n-3 PUFA should not exceed 4.0 (Wood et al., 2003). In our study, PUFA/SFA ratio did not differ between meat of lambs reared in two seasons and obtained values (0.18 and 0.19) were below the recommended ones. The results are in accordance with other authors (Juárez et al., 2009; Oriani et al., 2005; Vacca et al., 2008) where PUFA/SFA values in meat of unweaned lambs did not reach the recommended values. However, they reported slightly higher values for PUFA/SFA ratio than obtained in this study. Rearing season did not influence n-6/n-3 ratio while obtained values were inside recommended ones. Contrary to our results, Mazzone et al. (2010) reported higher content of total PUFA, n-6 and n-3 as well as higher PUFA/SFA and lower n-6/n-3 ratio for meat of lambs reared in autumn. The differences were attributed to the traditional feeding system where lambs born and reared in autumn received milk from permanently pastured dams while those reared in winter were suckled by permanently stall-fed dams.

Table 6

Fatty acids (% of total analysed fatty acid methyl esters) determined in the total lipids of the *longissimus dorsi* muscle of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
C10:0 capric	0.65 \pm 0.11	0.75 \pm 0.18	NS
C11:0 undecanoic	0.04 \pm 0.01	0.04 \pm 0.01	NS
C12:0 lauric	1.45 \pm 0.25	1.84 \pm 0.50	*
C13:0 tridecanoic	0.09 \pm 0.01	0.12 \pm 0.02	**
C14:0 myristic	9.39 \pm 1.09	10.77 \pm 2.05	NS
C15:0 pentadecanoic	0.87 \pm 0.09	1.01 \pm 0.15	*
C16:0 palmitic	23.72 \pm 1.97	26.09 \pm 0.90	**
C17:0 heptadecanoic	1.34 \pm 0.15	1.12 \pm 0.10	***
C18:0 stearic	15.90 \pm 1.75	11.18 \pm 1.79	***
C20:0 arachidic	0.14 \pm 0.05	0.08 \pm 0.03	**
C21:0 heneicosanoic	0.31 \pm 0.09	0.16 \pm 0.03	***
C14:1 myristoleic	0.22 \pm 0.08	0.34 \pm 0.11	*
C16:1 palmitoleic	1.46 \pm 0.37	1.90 \pm 0.32	**
C17:1 heptadecanoic	0.47 \pm 0.11	0.55 \pm 0.10	NS
C18:1 trans tot.	3.89 \pm 1.45	3.05 \pm 1.21	NS
C18:1 cis tot.	27.69 \pm 3.08	28.12 \pm 3.29	NS
C18:2 c,t + t,c + t,t.	1.56 \pm 0.21	1.65 \pm 0.27	NS
C18:2 n-6 linoleic	2.96 \pm 1.08	3.56 \pm 0.83	NS
C18:3 n-3 linolenic	1.40 \pm 0.19	1.53 \pm 0.24	NS
C18:2 cis-9, trans-11 (CLA)	1.81 \pm 0.25	1.64 \pm 0.40	NS
C20:2 n-6 eicosadienoic	0.08 \pm 0.03	0.07 \pm 0.01	NS
C20:4 n-6 arachidonic	0.69 \pm 0.30	0.63 \pm 0.29	NS
C20:5 n-3 eicosapentoic	0.34 \pm 0.13	0.29 \pm 0.12	NS
C22:5 n-3 docosapentaenoic	0.46 \pm 0.11	0.36 \pm 0.14	NS
C22:6 n-3 docosehexanoic	0.19 \pm 0.06	0.14 \pm 0.07	NS
Other fatty acids	2.89 \pm 0.22	2.99 \pm 0.19	NS
<i>Sums and ratio</i>			
SFA	53.90 \pm 2.25	53.17 \pm 2.35	NS
MUFA	33.74 \pm 2.64	33.96 \pm 2.83	NS
PUFA	9.48 \pm 1.63	9.88 \pm 1.30	NS
Total n-6	3.72 \pm 1.29	4.27 \pm 1.01	NS
Total n-3	2.39 \pm 0.39	2.33 \pm 0.50	NS
n-6/n-3	1.52 \pm 0.39	1.88 \pm 0.53	NS
PUFA/SFA	0.18 \pm 0.03	0.19 \pm 0.02	NS
h/H	1.03 \pm 0.15	0.95 \pm 0.15	NS
AI	1.59 \pm 0.27	1.77 \pm 0.32	NS
TI	1.85 \pm 0.18	1.81 \pm 0.15	NS

NS: not significant; $P > 0.05$.

* $P < 0.05$.

** $P < 0.01$.

*** $P < 0.001$.

SFA = (C10:0 + C11:0 + C12:0 + C13:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C21:0).

MUFA = (C14:1 + C16:1 + \sum C18:1t + C18:1n-9).

PUFA = (C18:2n-6 + C18:3n-3 + C18:2c9t11 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3).

n-6 PUFA = (C18:2n-6 + C20:2n-6 + C20:4n-6).

n-3 PUFA = (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3).

h/H: the hypocholesterolemic/hypercholesterolemic ratio = $[(C18:1n-9 + C18:2n-6 + C18:3n-3 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3)/(C14:0 + C16:0)]$.

AI: atherogenic index = $(C12:0 + 4 \times C14:0 + C16:0) / [MUFA + PUFA (n-6) \text{ and } (n-3)]$.

TI: thrombogenic index = $(C14:0 + C16:0 + C18:0) / [0.5 \times MUFA + 0.5 \times PUFA (n-6) + 3 \times PUFA (n-3) + (n-3)/(n-6)]$.

The fatty acid composition of SC fat is shown in Table 7. C16:0 was higher ($P < 0.05$) and C21:0 lower ($P < 0.01$) in SC fat of lambs reared in autumn than those reared in winter, while other SFA did not show significant differences between lambs of two seasons. Among the MUFA, C17:1 was the only one affected ($P < 0.01$) by the rearing season, with higher level observed in the SC fat of lambs reared in winter. PUFA profile was affected by the rearing season where lambs reared in autumn and slaughtered at Christmas had higher ($P < 0.05$) levels of C18:2t (C18:2 c,t + t,c + t,t.). Much higher ($P < 0.001$) levels of linoleic (C18:2 n-6) and linolenic (C18:3 n-3) PUFA were also observed in SC fat of lambs reared in autumn. Since the above fatty acids have a beneficial influence on human health, it can be stated that SC fat of lambs slaughtered at Christmas delivers more health benefits. The above written is probably caused by seasonal variation in the quality of green forage and its availability to sheep. As demonstrated by Cabiddu et al. (2003) FA composition of ewe's milk can differ according to different botanical composition of grazed pastures. Although in our study animals grazed on alfalfa dominated pastures in both season, differences in overall botanical composition were present and therefore the differences in their FA composition were probably present as well. On the other side, SC fat of lambs reared in winter was characterised by higher levels of long chain fatty acids C20:2 ($P < 0.05$) and C20:4 ($P < 0.01$) from n-6 series. Total PUFA values were higher ($P < 0.01$) in SC fat of lambs reared in autumn due to higher levels of total fatty acids from n-6 ($P < 0.05$) and n-3 ($P < 0.001$) series. This led to a higher and therefore more desirable ($P < 0.05$) PUFA/SFA ratio for human health in SC fat of lambs reared in autumn. However, the values of PUFA/SFA ratio were higher than those reported by Juarez et al. (2009) for SC fat of Grazalema Merino and Churra Lebrijana light lambs (0.09 and 0.11, respectively) or Vacca et al. (2008) for perirenal and pelvic fat of Mouflon x Sarda and Sarda x Sarda suckling lambs (0.1 and 0.1, respectively). Lambs slaughtered at Christmas had also more favourable n-6/n-3 ratio in SC fat that was lower ($P < 0.01$) than observed in SC fat of lambs slaughtered at Easter suggesting that lambs slaughtered at Christmas had a higher nutritional value and more health-promoting properties of SC fat, compared to lambs slaughtered at

Easter. The n-6/n-3 ratios reported by Vacca et al. (2008) for perirenal and pelvic fat (4.5 and 5.7, respectively) for suckling lambs were much higher than those obtained in this study for SC fat of lambs reared in winter and autumn (1.82 and 1.62, respectively). SC fat of lambs reared in two different seasons also differed in levels of other non-identified fatty acids where higher values were observed in the SC fat of lambs slaughtered for Easter.

Table 7

Fatty acids (% of total analysed fatty acid methyl esters) determined in subcutaneous adipose tissue of Bergamasca lambs slaughtered at 60 days of age related to the rearing season (mean \pm SD).

	Rearing season		Significance
	Winter	Autumn	
C10:0 capric	0.81 \pm 0.18	0.86 \pm 0.11	NS
C11:0 undecanoic	0.04 \pm 0.01	0.04 \pm 0.01	NS
C12:0 lauric	1.68 \pm 0.27	1.50 \pm 0.30	NS
C13:0 tridecanoic	0.10 \pm 0.02	0.08 \pm 0.02	NS
C14:0 myristic	10.08 \pm 1.02	9.28 \pm 1.24	NS
C15:0 pentadecanoic	0.95 \pm 0.08	0.90 \pm 0.09	NS
C16:0 palmitic	21.18 \pm 1.86	22.99 \pm 1.51	*
C17:0 heptadecanoic	1.51 \pm 0.13	1.51 \pm 0.06	NS
C18:0 stearic	18.86 \pm 3.06	18.23 \pm 1.68	NS
C20:0 arachidic	0.19 \pm 0.04	0.17 \pm 0.05	NS
C21:0 heneicosanoic	0.26 \pm 0.11	0.10 \pm 0.01	**
C14:1 myristoleic	0.15 \pm 0.04	0.14 \pm 0.02	NS
C16:1 palmitoleic	1.06 \pm 0.23	0.94 \pm 0.12	NS
C17:1 heptadecanoic	0.47 \pm 0.06	0.39 \pm 0.06	**
C18:1 trans tot.	4.90 \pm 1.37	3.82 \pm 1.79	NS
C18:1 cis tot.	28.27 \pm 1.69	28.69 \pm 2.51	NS
C18:2 c,t + t,c + t,t	1.55 \pm 0.18	1.72 \pm 0.14	*
C18:2 n-6 linoleic	1.97 \pm 0.21	2.44 \pm 0.14	***
C18:3 n-3 linolenic	0.98 \pm 0.11	1.37 \pm 0.11	***
C18:2 cis-9, trans-11 (CLA)	1.45 \pm 0.55	1.71 \pm 0.20	NS
C20:2 n-6 eicosadienoic	0.08 \pm 0.04	0.04 \pm 0.01	*
C20:4 n-6 arachidonic	0.22 \pm 0.14	0.05 \pm 0.02	**
C20:5 n-3 eicosapentenoic	0.06 \pm 0.05	0.02 \pm 0.01	NS
C22:5 n-3 docosapentaenoic	0.15 \pm 0.05	0.13 \pm 0.03	NS
C22:6 n-3 docosehexanoic	0.05 \pm 0.02	0.03 \pm 0.02	NS
Other fatty acids	3.02 \pm 0.16	2.83 \pm 0.10	**
<i>Sums and ratio</i>			
SFA	55.64 \pm 3.24	55.68 \pm 2.33	NS
MUFA	34.84 \pm 2.51	33.98 \pm 2.12	NS
PUFA	6.50 \pm 0.80	7.51 \pm 0.45	**
Total n-6	2.26 \pm 0.34	2.52 \pm 0.13	*
Total n-3	1.24 \pm 0.20	1.56 \pm 0.09	***
n-6/n-3	1.82 \pm 0.15	1.62 \pm 0.09	**
PUFA/SFA	0.12 \pm 0.02	0.13 \pm 0.01	*
h/H	1.02 \pm 0.11	1.03 \pm 0.15	NS
AI	1.66 \pm 0.20	1.63 \pm 0.26	NS
TI	2.22 \pm 0.30	2.15 \pm 0.19	NS

NS: not significant; P > 0.05.

* P < 0.05.

** P < 0.01.

*** P < 0.001.

SFA = (C10:0 + C11:0 + C12:0 + C13:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C21:0).

MUFA = (C14:1 + C16:1 + \sum C18:1t + C18:1n-9).

PUFA = (C18:2n-6 + C18:3n-3 + C18:2c9t11 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3).

n-6 PUFA = (C18:2n-6 + C20:2n-6 + C20:4n-6).

n-3 PUFA = (C18:3n-3 + C20:5n-3 + C22:5n-3 + C22:6n-3).

h/H: the hypocholesterolemic/hypercholesterolemic ratio = [(C18:1n-9 + C18:2n-6 + C18:3n-3 + C20:2n-6 + C20:4n-6 + C20:5n-3 + C22:5n-3 + C22:6n-3)/(C14:0 + C16:0)].

AI: atherogenic index = (C12:0 + 4 x C14:0 + C16:0)/ [MUFA + PUFA (n-6) and (n-3)].

TI: thrombogenic index = (C14:0 + C16:0 + C18:0)/ [0.5 x MUFA + 0.5 x PUFA (n-6) + 3 x PUFA (n-3) + (n-3)/(n-6)].

4. Conclusions

Rearing season (winter *vs.* autumn) had light effect on growth, carcass traits and meat quality of Bergamasca light lambs. Lambs reared in winter had higher birth weight and drip loss after the 6th day of meat storage period as well as lower proportion of pluck and pH values than lambs reared in autumn. Rearing season did not affect growth performances, carcass traits and other physical and chemical characteristics of meat while FA composition from both IM and SC fat was influenced by the rearing season. In particular, lambs from the two rearing seasons differed in some SFA and MUFA in IM fat, while SC fat of lambs reared in autumn showed better FA profile as a result of higher content of total n-6, n-3 and PUFA, as well as more favourable PUFA/SFA and n-6/n-3 ratios for human health.

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General conclusions

The research investigated how different biological and production factors influence quality of “Agnello del Centro Italia” PGI light lambs produced under traditional transhumant sheep system adopted in Marche region (Central Italy).

Although it is reported that breed is the main biological factor influencing quality of lambs on both levels, carcass and meat quality (Juárez et al., 2008; Ramírez-Retamal and Morales, 2014; Sañudo et al., 1997), in this research its effect on “Agnello del Centro Italia” PGI light lambs, was slight. Bergamasca, Italian Merino and Sopravissana lambs showed similar growth performances, carcass traits and meat quality (as defined by physical and chemical characteristics), although some differences between them occurred for some of investigated parameters. On the other side, the effect of breed on FA profile of meat and adipose tissue of lambs of investigated breeds was much more evident. Italian Merino and Sopravissana lambs, which are both Merino derived breeds, showed similar FA composition of meat and adipose tissue. Results confirmed previous findings (Santos-Silva et al., 2002), indicating that lambs of similar genotype may not vary in FA composition. Furthermore, GC data in combination with chemometric approach (canonical discriminant analysis) was effective to discriminate between breeds and correctly predict them. Research also revealed that, although meat of Bergamasca lambs presented less favourable quality (on the basis of lowest CLA and highest TI), meat and adipose tissue of all three breeds had a high content of health promoting n-3 PUFA and CLA *cis*-9, *trans*-11. Furthermore, the ratios between n-6/n-3 PUFA of both meat and adipose tissue were low and met the recommended values for human diet.

After feeding, slaughter weight/age is the most important production factor influencing carcass and meat quality (Cifuni et al., 2000; D’Alessandro et al., 2013; Marino et al., 2008; Oriani et al., 2005). Increase of the slaughter age in Bergamasca lamb from 40 to 60 days resulted in an improvement in some carcass traits and physical

characteristics of meat, while meat chemical composition was not influenced. Furthermore, meat FA profile was not strongly affected by slaughter age. On the contrary, adipose tissue showed more favourable FA profile in younger lambs than in the older ones.

The research indicated that for the light lamb production in the traditional rearing systems, instead of 40 days farmers should consider 60 days as optimal slaughter age for Bergamasca lambs to produce slightly heavier carcasses without compromising the quality of meat.

There is a paucity of data on the effect of rearing season on lamb carcass and meat quality of light lambs reared in extensive systems (Mazzone et al., 2010). This research showed that different rearing season (winter vs autumn) did not have much influence on growth, carcass traits and meat quality of Bergamasca light lambs. However, FA profile of both meat and adipose tissue was influenced by the rearing season. In particular, adipose tissue showed better FA profile in lambs reared in autumn.

Finally, it can be concluded that this PhD research is an important contribution for filling the present gaps of knowledge regarding the quality of “Agnello del Centro Italia” light lambs produced under traditional systems in Marche region as it gives useful information both for local breeders and consumers.

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