



The relationship between muscle fiber characteristics and some meat quality parameters in Turkish native sheep breeds



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ABSTRACT

This research was conducted to determine muscle fiber characteristics and its relationship with some meat quality traits in Longissimus dorsi (LD) and Semitendinosus (ST) muscles from lambs of some Turkish native sheep breeds. A total of 36 singleton male lambs were used as experimental pure breed animals of Karayaka (n = 6), Kıvrıkcık (n = 6), Middle Anatolian Merino (n = 6), Awassi (n = 6), Morkaraman (n = 6) and Akkaraman (n = 6) breeds. All experimental animals were fed the same diet until they reach to a target weight of 40 kg weight. After the feeding period all lambs were slaughtered and LD and ST muscle samples were collected for determination of some meat quality traits (tenderness, pH, water holding capacity, and colour) and ATPase staining of muscle fibers. Type IIB muscle fiber numbers of Morkaraman sheep were higher than those of other breeds in LD muscle ($P < 0.05$). Awassi lambs had higher number of ($P < 0.05$) type IIA fibers and total fiber numbers in ST muscle compared to other breeds. Diameter of type I muscle fiber of ST muscle from Kıvrıkcık lambs was higher than those of other breeds ($P < 0.05$). There were positive correlations between diameters of type I ($r = 0.513$; $P < 0.05$), type IIIA ($r = 0.476$; $P < 0.05$) and tenderness in LD and ST muscles of all breeds. Results of the present study showed that muscle fiber characteristics of lambs of different Turkish native sheep breeds differ and muscle fiber characteristics influence some meat quality traits.

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1. Introduction

Meat quality is a very important issue for consumers and it is also important for the meat industry in terms of meat processability and production of meat products (Lee et al., 2010; Joo et al., 2013). In many developed/developing countries, the demand of consumers for high quality meat, which is delicious, nutritious, safe and healthy, is increasing day by day. Therefore, the meat industry has begun to work in order to meet this demand to provide continuous consumption of meat products (Joo et al., 2013). It is essential to understand the skeletal muscle tissue characteristics such as

muscle fiber types, metabolic and contractile features to produce high quality meat and what factors affecting them which influence meat quality. The quality of the meat presented for consumption is affected by factors such as post mortem changes in muscle tissues, muscle structure, stromal tissue content, fat content, muscle fiber composition, chemical composition and interaction of chemical constituents, microbiological contamination level, pre-slaughter stress, product processing and storage (Sen et al., 2011; Joo et al., 2013). The majority of skeletal muscle is composed by muscle fibers and biological characteristics of muscle fibers are directly related to meat quality such as tenderness, pH and colour (Joo et al., 2013). Generally, skeletal muscle fibers are classified based on their major metabolic activities and contractile properties (Lee et al., 2010; Joo et al., 2013). Moreover type, number and diameter of muscle fibers have important effects on quality of the meat (Joo et al., 2013).

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Generally fibers in skeletal muscle tissue are classified into three types as follows; 1) slow-oxidative or type I; 2) fast oxido-glycolytic or type IIA; 3) fast glycolytic or type IIB (type IIX) muscle fiber (Furuichi et al., 2014). Characteristics of skeletal muscle tissue may vary depending on various factors such as breed (Ryu et al., 2008), sex (Ozawa et al., 2000), hormone level (Rehfeldt et al., 2004), growth term (Gondret et al., 2006), nutrition (Sen et al., 2016) and muscle location (Hwang et al., 2010). Muscle fiber composition of the skeletal muscle tissue is one of the intrinsic properties of muscle and this has impact on eating quality characteristics of meat such as colour, odor, flavor, juiciness, tenderness, and texture (Bünger et al., 2009). Also, metabolic and contractile characteristics of skeletal muscle tissue are determined by the composition of muscle fibers (Ryu et al., 2008; Ozawa et al., 2000). Therefore, the understanding of relationship between muscle fiber features and traits of meat quality would increase the high quality meat production. Manipulation of muscle fiber features may increase profitability of the meat industry. Consequently, total muscle fiber numbers in the muscle cross section area, muscle fiber diameter, muscle fiber type and composition, which influence post mortem meat quality, are important features for meat quality (Ryu et al., 2008).

Lamb meat production is a of major importance in meat production in Turkey, with approximately 32 million sheep (TurkStat, 2015). Additionally, Turkey has many local sheep breeds, which are well suited to a variety of geographic and climatic conditions. Although these breeds have low production levels, they constitute an important source of red meat production under harsh climate conditions. The most popular native sheep breeds in Turkey are Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman, and male lambs of these breeds are used as fattening material for meat production. Although, there have been numerous studies on growth performance, carcass characteristics and meat quality of male lambs of these breeds, there is no comparative information about muscle fiber characteristics and its relationship with some meat quality traits. For this reason, detect of skeleton muscle fiber type differences among Turkish native sheep breeds is important; especially, whether muscle fiber differences observed could be exploited to improve meat quality and managing the meat quality through improved breeding strategies (Bünger et al., 2009).

The purpose of the current study was therefore to determine muscle fiber characteristics and its relationship with some meat quality traits in Longissimus dorsi (LD) and Semitendinosus (ST) skeletal muscles from lambs of Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman Turkish native sheep breeds.

2. Materials and methods

2.1. Animals

This experiment was conducted at the Agricultural Research Unit of Gaziosmanpaşa University, Tokat, Turkey (40°31'N, 36°53'E, 650 m above sea level). A total of 36 singleton male lambs were used as experimental pure breed animals of Karayaka (n=6), Kıvrıkcık (n=6), Middle Anatolian Merino (n=6), Awassi (n=6), Morkaraman (n=6) and Akkaraman (n=6) breeds. Weaning age of lambs was at day 90 and they had similar weaning weights (approximately 20 kg). After weaning all lambs were subjected to a fattening period for approximately 125 ± 7 days and they were slaughtered upon reaching a target weight of 40 kg weight. The lambs were fed a diet composed of approximately 100 g/day alfalfa hay and concentrate feed. The nutrient contents of the diet during fattening period are given in Table 1. Water and mineral stone were freely available during fattening period.

Table 1
The nutrient contents of concentrate and alfalfa hay.

Nutrient Content	Concentrate	Alfalfa hay
Dry Matter (%)	93.10	94.02
Crude Protein (%)	15.21	15.01
ADF (%)	29.41	59.75
NDF (%)	30.22	58.22
Crude Oil (%)	2.23	0.74
Crude Ash (%)	8.58	10.30
Metabolisable Energy (kcal/kg)	2690.00	1878.00

2.2. Muscle sample collection

Following slaughter, the carcasses of all lambs were chilled for 24 h at 4 °C. Within 30 min of slaughter muscle samples (approximately 50 g) were isolated from the mid-sections of LD and ST muscles. Fat and connective tissue were removed from muscle samples and immediately frozen in liquid nitrogen. Afterwards all muscle samples were stored at –80 °C until ATPase staining of muscle fibers. For determination of meat quality traits, approximately 100–150 g muscle samples were collected from the central parts of the mid-section of the whole LD and ST muscles from the left side of the carcasses after 24 h chilled storage. These samples were trimmed of subcutaneous fat and fascia before storage at 4 °C for meat quality analysis.

2.3. Meat quality analyses and histochemical determination of muscle fiber type composition

All meat quality parameters (tenderness, pH, water holding capacity, and colour) were analyzed as described by Sen et al. (2011). Muscle fibers characteristics of LD and ST muscles were analyzed using myosin ATPase staining at pH 4.2 described by Broke and Keiser (1970) and Sen et al. (2016). Muscle fibers were counted using a microscope (Nikon Eclipse E600, Nikon Corporation, Tokyo, Japan) linked to an image analysis software (Laica Q Win V3.4 Processing-Analysis Software). Pictures of stained muscle fibers from LD and ST muscles are presented in Fig. 1.

2.4. Statistical analysis

To analyze the data for muscle fiber types and diameter Kolmogorov-Smirnov one sample test was performed to examine the normality. Data did not distribute normally. Levene variance homogeneity test was performed to obtain the information about homoscedasticity and results showed that variances were not homogeneous. Then, non-parametric permutation tests were applied to the data. To compare the means posterior pairwise permutation tests were used (Onder, 2007; Onder and Cebeci, 2009). Data analyses were utilized with NPMANOVA software written by Anderson (2000). Relationships between muscle fiber characteristics and some meat quality traits were determined with a Pearson correlation analysis at the 95% confidence interval.

3. Results

3.1. Muscle fibers number

The numbers of type I, IIA, IIB and total muscle fibers in mm² muscle area of LD and ST muscles from Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman male lambs are presented in Fig. 2. Type I, IIA and total muscle fiber numbers in LD muscle were similar between lambs of different breeds, but type IIB muscle fiber numbers of Morkaraman lambs were higher than those of other breeds in LD muscle (P<0.05). Type IIA and total muscle fiber numbers of ST muscle in Awassi

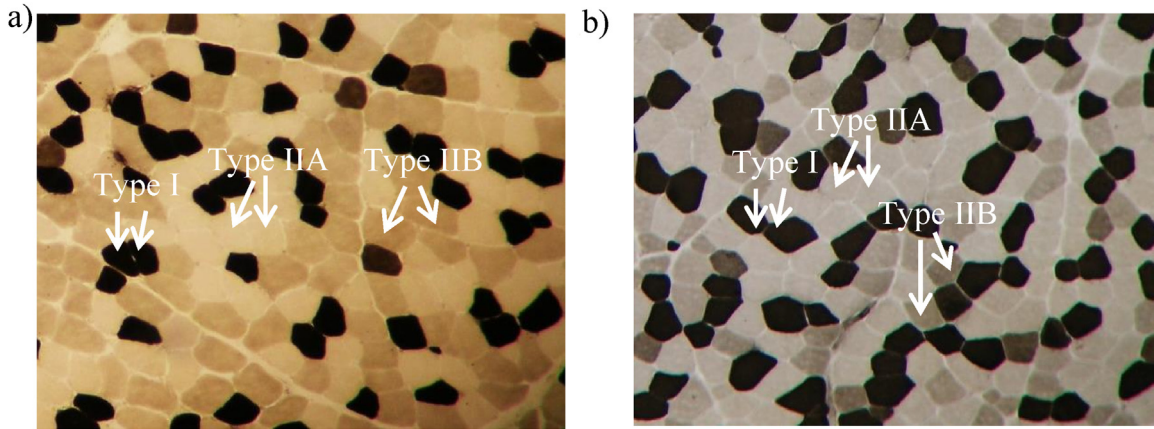


Fig. 1. Pictures of myosin ATPase staining (pH 4.2) of (a) longissimus dorsi and (b) semitendinosus muscles. The darkest muscle fiber is type I, intermediate muscle fiber is type IIB and the lightest muscle fiber is type IIA.

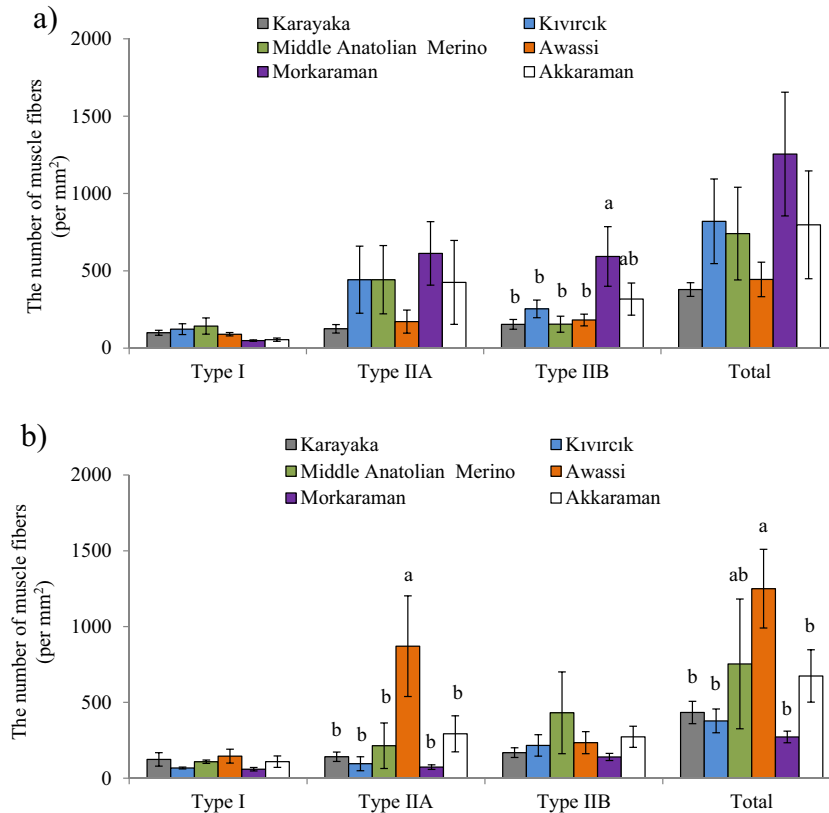


Fig. 2. The mean numbers of type I, IIA, IIB and total fibers in mm² muscle area of longissimus dorsi (a) and semitendinosus (b) muscles from Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman male lambs. The error bars represent the standard error of the mean and bars with different letters are significantly different at $P < 0.05$.

lambs were higher than those of other breeds ($P < 0.05$), while all lambs had similar type I and IIB muscle fiber numbers in ST muscle. Additionally, total muscle fiber numbers in LD muscle were higher than those of ST muscle of Morkaraman lambs, but total muscle fiber numbers in LD muscle were lower ($P < 0.05$) than those of ST muscle of Awassi lambs (data not shown). Type I, IIA, IIB and total muscle fiber numbers were similar between LD and ST muscles of Karayaka, Kıvrıkcık, Middle Anatolian Merino and Akkaraman male lambs (data not shown).

3.2. Muscle fibers diameter

The diameter of type I, IIA and IIB in LD and ST muscles of Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman male lambs are presented in Fig. 3. Diameter of muscle fibers were similar between lambs of all breeds in LD muscle, but Kıvrıkcık lambs had higher diameter of type I muscle fiber in ST muscle than those of other breeds ($P < 0.05$). Additionally, diameter of type IIA muscle fiber in LD muscle was higher than those of ST muscle of Awassi lambs, while the diameter of type I muscle

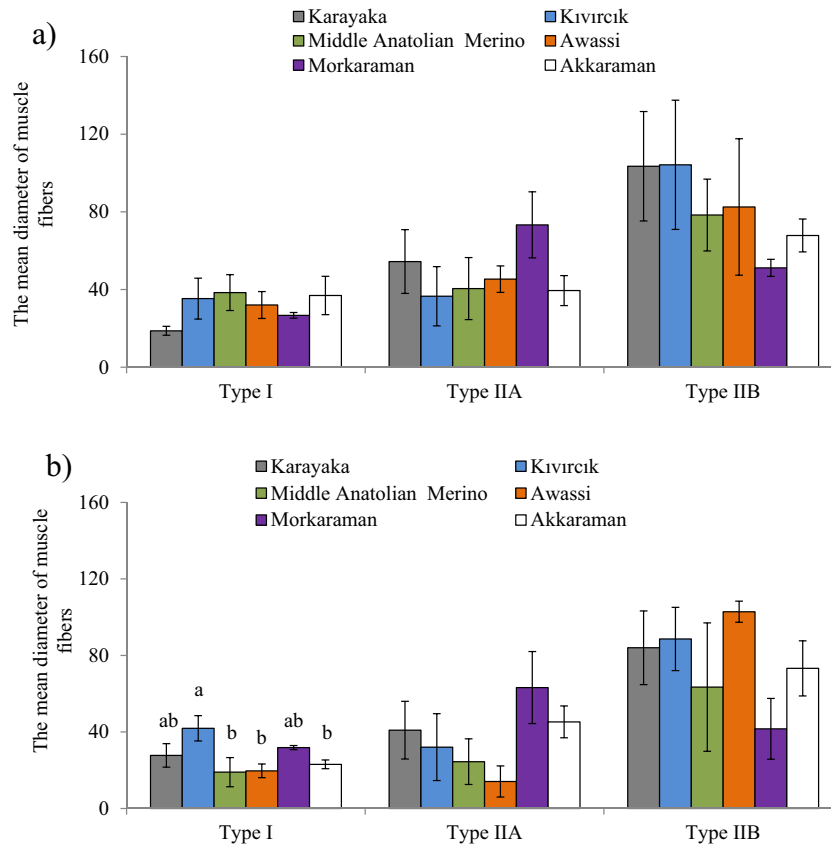


Fig. 3. The mean diameter (μm) of type I, IIA and IIB fibers in (a) longissimus dorsi and (b) semitendinosus muscles of from Karayaka, Kıvrıkcık, Middle Anatolian Merino, Awassi, Morkaraman and Akkaraman male lambs. The error bars represent the standard error.

fiber in LD muscle was lower ($P < 0.05$) than those of ST muscle of Morkaraman lambs (data not shown).

3.3. Correlations between muscle fiber characteristics and meat quality traits

Pearson correlation coefficients between muscle fiber characteristics and some meat quality traits for all lambs are presented in Table 2. The analysis of Pearson correlation coefficients on the pooled data for all breeds showed that there were positive correlation between diameter of type I muscle fibers and pH ($r = 0.604$; $P < 0.05$). On the contrary, a negative relationships were obtained between diameter of type I muscle fibers and WHC ($r = -0.411$; $P < 0.05$) in LD muscle. Positive correlations between diameters of type I and tenderness ($r = 0.513$; $P < 0.05$) in LD muscle, and diameters of type IIA and tenderness ($r = 0.476$; $P < 0.05$) in ST muscle were calculated.

The significant differences for Pearson correlation coefficients between muscle fiber characteristics and some meat quality traits in lambs of Karayaka, Akkaraman, Kıvrıkcık and Morkaraman Turkish native sheep breeds are presented in Table 3. There were positive correlations between meat lightness (L^*) and number of type IIB fibers ($r = 0.822$; $P < 0.01$), L^* and type I fibers ($r = 0.877$; $P < 0.01$) and L^* and diameter of type I fibers ($r = 0.817$; $P < 0.01$) in LD muscle for Karayaka lambs. Also, there were positive correlations between a^* , numbers of type I and type IIA muscle fibers ($r = 0.721$ and $r = 0.813$; $P < 0.01$) in ST muscle for Karayaka lambs. There were positive correlations between meat water holding capacity (WHC) and number of type IIB muscle fibers ($r = 0.874$; $P < 0.01$) and diameter of type IIB fibers and meat pH ($r = 0.792$; $P < 0.01$) in LD muscle for Akkaraman male lambs. There were pos-

itive correlations between WHC and number of type IIB fibers ($r = 0.877$; $P < 0.01$) and meat redness (a^*) and number of type I fibers ($r = 0.898$; $P < 0.01$) in ST muscle for Kıvrıkcık male lambs. There were positive correlations between a^* and diameter of type I fibers ($r = 0.958$; $P < 0.01$) in ST muscle for Morkaraman lambs

4. Discussion

In this study, muscle fiber characteristics and its relationship with some meat quality traits of LD and ST muscles in six Turkish sheep breeds have been identified and the results demonstrate that muscle fiber characteristics of male lambs of six Turkish sheep breeds differ and muscle fiber characteristics influence some meat quality traits.

4.1. Muscle fibers number

Breeds within a species are the most important factor affecting compositions of muscle fiber type, size and total number of fibers in skeletal muscle tissue. Most of previous studies reporting muscle fiber characteristics have been carried out in pigs with different breeds. However, there has been limited comparative information about muscle fiber characteristics in ruminant animals. Previous studies showed that Berkshire pigs has more type I fiber than that of Yorkshire and Landrace pigs in LD muscle (Ryu et al., 2008). Also, Wimmers et al. (2008) reported that high muscularity in different pig breeds is highly correlated with high ratio in myosin heavy chain transcripts of type IIB muscle fiber. Therefore, it should be underlined that an increase in carcass lean percentage of different breeds can be attributed to muscle fiber characteristics. Consequently, there is a strong relationship between muscle fiber

Table 2
Pearson correlation coefficients between muscle fiber characteristics and some meat quality traits for all male lambs born to some Turkish native sheep breeds.

Traits	Tenderness	pH	WHC	Lightness (L*)	Redness (a*)	Yellowness (b*)
LD						
Numbers						
Type I	0.232	-0.211	0.205	-0.310	-0.103	-0.174
Type IIA	0.201	-0.140	0.346	0.100	0.031	-0.047
Type IIB	0.079	0.116	-0.151	-0.111	0.288	0.080
Total	0.214	-0.182	0.334	0.181	0.074	0.013
Diameters						
Type I	0.513*	0.604*	-0.411*	0.119	0.082	-0.018
Type IIA	0.259	0.061	0.108	0.120	0.074	0.209
Type IIB	0.131	-0.162	0.194	0.373	0.164	0.163
Average	0.234	0.235	-0.146	0.020	0.237	0.141
ST						
Numbers						
Type I	0.248	-0.137	0.235	0.082	0.313	0.300
Type IIA	0.296	-0.154	-0.288	-0.087	0.060	-0.035
Type IIB	0.227	-0.181	0.134	-0.329	0.165	0.134
Total	0.093	-0.229	-0.168	-0.125	0.096	-0.048
Diameters						
Type I	0.256	0.152	-0.153	-0.110	-0.062	0.217
Type IIA	0.476*	0.074	0.134	0.005	-0.051	0.142
Type IIB	0.230	-0.209	0.090	-0.157	0.006	-0.146
Average	0.316	-0.135	0.140	-0.259	0.079	0.217

LD = longissimus dorsi muscle. ST = semitendinosus muscle. WHC = water holding capacity.
P < 0.05*.

Table 3
Pearson correlation coefficients between muscle fiber characteristics and some meat quality traits in some Turkish native sheep breeds.

Breeds	Muscle fiber traits	WHC	Lightness (L*)	Redness (a*)
Karayaka	Numbers of LD Type I		0.877**	
	Numbers of LD Type IIB		0.822**	
	Numbers of ST Type I			0.721**
	Numbers of ST Type IIB			0.813**
Akkaraman	Diameters of LD Type I		0.817**	
	Numbers of LD Type IIA	0.874**		
	Diameters of LD Type IIB	0.792**		
Kıvrıcık	Numbers of ST Type I			0.898**
	Numbers of ST Type IIB	0.877**		
Morkaraman	Diameters of ST Type I			0.858**

LD = longissimus dorsi muscle. ST = semitendinosus muscle. WHC = water holding capacity.
P < 0.01**.

composition and growth performance when different breeds compared within a species. In the present study Morkaraman lambs had higher number of type IIB muscle fibers in LD muscle compared to lambs born to other breeds. Also, number of type IIA muscle fibers in ST muscle was higher at Awassi lambs. These results indicate that the muscle development of Morkaraman and Awassi breeds may be more affected by environmental factors. These differences between lambs of six Turkish sheep breeds may also be due to maternal nutrition level during gestation, because the number of skeletal muscle fibers especially type II muscle fibers are affected from low maternal nutrition level during gestation (Dwyer et al., 1994; Fahey et al., 2005; Sen et al., 2016). Moreover, the low level of maternal nutrition causes to a decrease in the number of secondary muscle fibers (Wigmore and Stickland, 1983). Unfortunately observations regarding to maternal nutrition level during gestation were not recorded in the present study which may help to interpret whether these had any effect on the muscle fiber characteristics of lambs of six Turkish sheep breeds.

4.2. Muscle fibers diameter

Environmental factors such as lamb's maintenance, nutrition and lamb mobility during postnatal period do not affect the number and the type of muscle fibers. However, these factors, especially

nutrition, affect the diameter of muscle fibers (Fahey et al., 2005). The diameter of the fibers in LD muscle did not show variations between six Turkish sheep breeds. However, Kıvrıcık lambs had higher diameter of type I fibers in ST muscle than the lambs of other breeds. This result may be explained by the fact that lambs of Kıvrıcık sheep had lower number of type I fibers in ST muscle. The fewer total number of muscle fibers may have led to higher increase in muscle fibers diameter (Dwyer et al., 1994). Similarly Gondret et al. (2006) showed that low total muscle fiber number in the LD muscle increased the diameter of muscle fiber after fattening period in pigs.

4.3. Correlations between muscle fiber characteristics and meat quality traits

Joo et al. (2013) reported that traits of muscle fibers such as total number of fibers, muscle fiber diameter and composition of muscle fiber types are related with meat quality. Muscle fiber characteristics may influence pH, meat colour, WHC and tenderness in meat (Joo et al., 2013). Renner, (1990) reported that an increase in the type I muscle fiber numbers in skeletal muscle tissue decreases colour stability of the meat. In the present study, we observed positive correlations between meat lightness, redness and type I muscle fiber number in LD and ST muscles, respectively. These results indicate the importance of muscle fiber types in skeletal muscle tissue on meat colour characteristics. Numbers and size of type IIA and IIB fibers increase lightness and decrease WHC of pork meat (Larzul et al., 1997; Kim et al., 2013). Similarly, in the present study numbers and size of type IIA and IIB fibers increased meat lightness in some Turkish native sheep breed (Karayaka and Kıvrıcık). Contrast to previous studies, there were positive relationship between WHC and number of type IIB muscle fibers in Akkaraman and Kıvrıcık male lambs. However, the pooled data for all breeds showed a negative relationship between diameter of type I muscle fibers and WHC in LD muscle. These differences may be explained by species and breed differences because the meat quality parameters can vary depending on the species and breed (Joo et al., 2013).

Maltin et al. (2003) showed that the composition of muscle fiber types in different muscles affect meat tenderness. Kovanen et al. (1984) also reported that type I (slow-twitch) muscle fibers have more collagen, which leads to a decrease in tenderness of

meat. Muscles with larger fiber size, especially type II fibers, exhibit tougher meat characteristic than muscles with smaller fiber size in cattle (Renand et al., 2001) and in pig (Karlsson et al., 1993). These findings support observations in the present study that diameter of type I and type IIA muscle fibers in LD and ST muscle, respectively, showed a positive correlation with tenderness of meat in all breeds. Ryu et al. (2008) showed that there is a negative correlation between muscle pH value and numbers of type IIB muscle fibers. In contrast, Choi et al. (2007) reported an increase in proportion of type I fibers in muscle extents pH drop. In the present study, there were no significant correlations between muscle fibers number and meat pH in all breeds, but positive correlations were observed between diameters of type I muscle fibers in LD muscle. Additionally, higher diameter of type IIB muscle fibers was related to the development of higher pH in LD muscle of Akkaraman male lambs. The understanding of the impact of muscle fiber characteristics on meat quality can provide information for the optimization of muscle growth and meat quality in meat producing species (Joo et al., 2013). Consequently, the results observed in the present study suggest that muscle fiber characteristics of Turkish native sheep breeds may be used as an important indicator of the meat quality of Turkish native sheep breeds.

5. Conclusion

In conclusion, the results of the present study suggest that breed is an important factor affecting skeletal muscle fiber characteristics in sheep. Differences in muscle fiber characteristics may influence meat production and quality. Hence, these results may have implications for selecting the fattening material to increase quality of meat production from lambs and suggest that muscle fiber characteristics should be taken into account.

Conflict of interests

The authors declare that there is no conflict of interests.

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