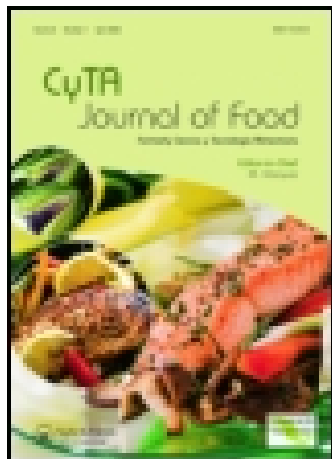


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Effects of breed, muscle type, and frozen storage on physico-chemical characteristics of lamb meat and its relationship with tenderness

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Effects of breed, muscle type, and frozen storage on physico-chemical characteristics of lamb meat and its relationship with tenderness

Efecto de la raza, músculo y tiempo de congelación sobre las características físico-químicas de carne de cordero y su relación con la ternura de la carne

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The effects of breed, muscle types, and frozen storage time on the physico-chemical characteristics of the two most important China breeds were considered. Twenty-four lambs of Bashbay and Xinjiang Merino sheep of 7–9 months old were slaughtered, respectively, and frozen for 1, 7, 15, or 30 days. The meat pH, water holding capacity (WHC), intramuscular fat, cooking loss, myofibrillar fragmentation index (MFI), moisture, shear force (SF), and connective tissue were measured at 24 h postmortem. These physico-chemical characteristics varied with breed and muscle types. Xinjiang Merino had a higher pH ($P < 0.001$) than Bashbay. M. supraspinatus had a higher pH ($P < 0.001$) compared to the M. gluteus and M. longissimus dorsi. The total and insoluble collagen and intramuscular fat were higher in Xinjiang Merino. As the time of frozen storage increased from 1 to 15 days, the intramuscular fat, cooking loss, WHC, and MFI in Xinjiang Merino increased gradually. SF and tenderness were improved.

Keywords: lamb; muscle; freezing; meat quality; tenderness

Los efectos de la raza, los tipos de músculos y el tiempo de almacenamiento congelado sobre las características físico-químicas de las dos razas más importantes de China fueron considerados. Veinte cuatro corderos de Bashbay y Xinjiang Merino fueron utilizados respectivamente. Los animales fueron sacrificados a una edad aproximada de 7 a 9 meses de edad y la carne fue congelada durante 1, 7, 15 o 30 días. El pH de carne, capacidad de retención de agua (WHC), grasa intramuscular, Las pérdidas al cocinar, índice de fragmentación miofibrilar (IFM), humedad, fuerza de corte (SF) y tejido conjuntivo fueron medidos en 24 horas post-mortem. Estas características físico-químicas variaron con raza y tipos musculares. Xinjiang Merino tuvo un pH mayor ($P < 0,001$) que bashbay ovejas. M supraspinatus tenía un pH mayor ($P < 0,001$) en comparación a la M glúteo y M longissimus dorsi. El colágeno total e insoluble y grasa intramuscular fueron superiores en Xinjiang Merino. Como el tiempo de almacenamiento congelado aumentó de 1 día para 15 días, la grasa intramuscular, Las pérdidas al cocinar, la capacidad de retención de agua y la MFI en Xinjiang Merino aumentaron gradualmente. Fuerza de corte y ternura fue mejorado.

Palabras clave: cordero; músculo; la congelación; la calidad de la carne; la ternura

1. Introduction

Tenderness is regarded as one of the most important palatability traits by which consumers judge meat quality, and it depends on many physico-chemical factors (Mette, et al., 2011). Extensive studies have been carried out to understand the factors which regulate the meat quality and it is generally regarded that intrinsic and extrinsic factors of a lamb have an influence on meat tenderness. The intrinsic factors include breed, age, sex, and muscle location. The extrinsic factors include nutrition, chilling, ageing, freezing, and the methods and temperatures for cooking. However, the effect of extrinsic factors on tenderness is somewhat mitigated by the role of intrinsic factors (Chang, Xu, & Zhou, 2009; Mette et al., 2011) such as myofibrillar structure, connective tissue, and intramuscular fat (IMF) content.

Connective tissue plays an important role in determining meat tenderness (Fang, Nishimura & Takahashi, 1999), which is appreciated by consumers after purchase and depends on the distribution, density, and properties of the connective tissue. The contribution of this fibrous protein to tenderness is dependent on collagen content, collagen solubility, and extent of its cross-linking (Kuber et al., 2004). Except that, meat eating

quality traits such as tenderness, flavor, and juiciness are also known to be linked to fat levels (Lambe et al., 2009). Increased intramuscular fat content has long been thought to improve meat tenderness (DeVol et al., 1988). Many studies have analyzed the effect of breed and muscle types on the physico-chemical characteristics of lamb meat (Hoffman, Muller, Cloete, & Schmidt, 2003; Young, Reid, & Scales, 1993), but most have only considered different ageing times rather than frozen storage time (Martínez et al., 2005). In general, increasing the ageing time will increase the tenderness but little is known on how the frozen storage time can vary in terms of breed or muscle types in this lamb meat.

This study evaluated the effects of breed, muscle types, and time of frozen storage on the physical-chemical properties, and assessed the variability in lamb tenderness between two breeds.

2. Materials and methods

2.1. Materials

Xinjiang native sheep from two breeds (Bashbay sheep, $n = 24$ and Xinjiang Merino, $n = 24$, China) were collected

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from the same local farm (Xinjiang Tacheng) at an approximate age of 7–9 months old. The sheep were electrically stunned and slaughtered according to standard procedures. The muscle samples were collected at 24 h postmortem. The *M. supraspinatus*, *M. gluteus*, and *M. longissimus dorsi* from each carcass of the two breeds vacuum packed were frozen stored at -18°C for 1, 7, 15, and 30 days in order to perform comparative analysis between the breeds and test the effect of frozen storage time on certain physico-chemical properties of meat.

2.2. Chemical analysis

2.2.1. pH and chemical composition (intramuscular fat and moisture)

Moisture content was determined by drying 10 g samples in an oven at 105°C until constant weight achievement. The percentage of IMF was determined using the method described by Li, Zhou, and Xu (2008). The pH values were measured using a digital pH meter (model DZ-1, Shanghai, China) in homogenate.

2.2.2. Water holding capacity, cooking losses, and thawing losses

Water holding capacity (WHC) was determined by centrifugation (Han, Wu, Wang, Xu, & Zhou, 2015). Five grams of samples were weighed in duplicate and centrifuged at $1500 \times g$ for 30 min. The samples were cut into 1 cm^3 cubes and were subjected to high speed homogenization in 50 mL ice-cold CaCl_2 (50 mmol/L) and then filtered through nylon nets (100 mesh). The above process was repeated three times. The connective tissue residue was weighed after drying at 105°C .

Thawing was performed in tap water for 15 min and thawing losses were expressed as the average proportion of weight loss by thawing ($\% \text{ThL} = [(\text{fresh weight} - \text{thawed weight})/\text{fresh weight}] \times 100$) (Muela, Monge, Sañudo, Campo, & Beltrán, 2015).

Cooking loss was measured by weighing samples before and after cooking for 1 h at 80°C in water bath. Samples were kept overnight in a fridge before they were subjected to shear testing. The shear force (SF) was measured using a TA-XT2-plus texture analyzer on cooked meat. SF parameters were followed by Martínez et al. (2013).

2.2.3. Collagen content

The total collagen and insoluble collagen of the muscle were determined by using a modified form of that reported by Zheng and Xia (2009) and Chang et al. (2011). Two grams of each meat sample were used to determine the total collagen and insoluble collagen content. One sample was hydrolyzed in 6N HCl for 18 h at 110°C to determine the percentage of the total collagen. Another sample was homogenized for 30 s with 8 mL of one-quarter-strength Ringer's solution and then heated for 60 min at 77°C in a water bath. After cooling to room temperature, the tubes were centrifuged for 15 min at $3300 \times g$. The precipitate was collected and 5 mL of one-quarter-strength Ringer's solution was mixed with the precipitate and centrifuged again for 15 min at $3300 \times g$. Then, the precipitate was hydrolyzed in 6N HCl

for 18 h at 110°C to determine the percentage of insoluble collagen. The percentage of collagen was calculated from the hydroxyproline concentration in the precipitate hydrolysate and expressed as the percentage of the initial sample wet weight, using a conversion factor of 7.25 (Crouse, Calkins, & Seideman, 1986).

2.2.4. Myofibrillar fragmentation index

The myofibrillar fragmentation index (MFI) was determined using the method described by Hopkins, Littlefield, and Thompson (2000) and Delgado, Geesink, Marchello, Goll, and Koochmaraie (2002). Duplicate 2 g samples were homogenized for 30 s with a 10 s break in 20 mL of ice-cold buffer. The buffer used was 0.1 M KCl, 1 mM EDTA (di-sodium), 1 mM sodium azide (NaN_3), and 25 mM potassium phosphate (7 mM KH_2PO_4 and 18 mM K_2HPO_4 giving a pH of 7.0 at 4°C). Myofibril suspensions were filtered (100 mesh strainers) to remove connective tissues. The filtrates were centrifuged at $1000 \times g$ for 15 min at 2°C and the supernatant decanted. The pellets of myofibrils were re-suspended in 10 mL of buffer, shaken thoroughly and centrifuged again. This process was repeated and the pellet finally re-suspended in 5 mL of cold buffer. The protein concentration of the suspensions was determined by using the Biuret method. Aliquots of the suspensions were diluted in buffer to a final protein concentration of $0.5 \pm 0.05 \text{ mg/mL}$. The diluted protein suspensions were poured into a cuvette, mixed, and the absorbance measured immediately at 540 nm using a spectrophotometer. The absorbance was multiplied by 200 to give a MFI value.

2.3. Statistical analysis

All measurements in study were done in triplicate; the results reported here were by means of the three trials. Statistical analyses were carried out using SPSS 16.0 (SPSS Inc., Chicago, IL). Duncan's multiple-range test was carried out to determine difference among the means. A general linear model was used to evaluate the significant differences between breeds, muscles, and frozen storage time. Pearson correlation coefficients were then evaluated to characterize the relationship between meat quality traits and muscle tenderness, using bivariate correlation coefficients.

3. Results and discussion

3.1. Chemical composition and pH of muscles

The moisture and intramuscular fat contents and pH values of fresh meat and frozen meat are summarized in Tables 1–3 for each breed and muscle type.

The moisture content of fresh meat was influenced by breed ($P < 0.05$), but was not affected by the muscle type (Table 1). Hoffman et al. (2003) and Muela, Sañudo, Campo, Medel, and Beltrán (2010) reported that there were no significant differences in the *M. longissimus dorsi* and *M. semimembranosus* between the different lamb breed combinations with regard to moisture and studied the effects of the freezing method and frozen storage duration on instrumental quality of lamb throughout display.

For frozen meat, breed, frozen storage time, and interaction between breed and freezing had significantly different effects on moisture contents (Table 2). The moisture contents of samples of meat from the two breeds following one day of frozen storage were

Table 1. Means ($n = 24$) and standard error values for meat quality properties of fresh meat for different breeds and muscles.Tabla 1. Medias ($n = 24$) y valores de error estándar para la calidad de la carne las propiedades de la carne fresca de diferentes razas y los músculos.

Parameters	Breed type		Muscle type			Significance	
	Bashbay sheep	Xinjiang Merino	M. supraspinatus	M. gluteus	M. longissimus dorsi	Breed	Muscle
Moisture (%)	75.47 ^a (0.19)	74.88 ^a (0.08)	73.07 (3.68)	76.28 (1.83)	75.47 (0.19)	*	NS
IMF (%)	1.45 ^a (0.16)	3.40 ^b (0.80)	2.82 (0.83)	1.17 (0.62)	1.45 (0.16)	NS	NS
pH	5.72 ^a (0.02)	5.98 ^a (0.01)	6.33 ^a (0.02)	5.74 ^b (0.03)	5.72 ^b (0.02)	***	***
CL (%)	26.25 ^a (6.39)	30.21 ^b (2.10)	31.92 (0.28)	34.99 (3.96)	26.25 (6.39)	NS	NS
WHC (%)	86.68 ^a (1.40)	95.58 ^b (0.42)	95.51 ^a (1.02)	92.89 ^a (0.51)	86.68 ^b (1.40)	**	**
MFI	141.04 ^a (3.26)	166.83 ^b (6.97)	98.10 (26.14)	56.43 (21.85)	141.04 (3.26)	*	NS
CTC (%)	10.08 ^a (1.16)	11.69 ^b (0.96)	9.32 (1.02)	11.33 (0.64)	10.08 (1.16)	NS	NS
SF (kg)	7.29 ^a (0.83)	9.52 ^b (0.47)	8.22 ^a (0.16)	11.59 ^b (0.69)	7.29 ^a (0.83)	NS	**
TC (%)	1.70 ^a (0.47)	2.39 ^b (0.26)	0.36 ^a (0.01)	0.49 ^a (0.17)	1.70 ^b (0.47)	NS	*
ISC (%)	1.24 ^a (0.20)	2.12 ^b (0.23)	0.27 ^a (0.04)	0.30 ^a (0.17)	1.24 ^b (0.20)	*	**
CS (%)	22.03 ^a (10.24)	11.29 ^b (2.39)	26.21 (9.22)	50.73 (20.92)	22.03 (10.24)	NS	NS

Notes: ^{a,b}Different superscripts in the same row represent significant differences among muscles ($P < 0.05$).

IMF, intramuscular fat; CL, cooking loss; WHC, water holding capacity; MFI, myofibrillar fragmentation index; CTC, connective tissue content; SF, shear force; TC, total collagen; ISC, insoluble collagen; CS, collagen solubility.

NS = not significant; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.Notas: ^{a,b}Diferentes superíndices en la misma fila representan diferencias significativas entre músculos ($P < 0.05$).

El IMF, la grasa intramuscular; CL, la cocina perdida; WHC, capacidad de retención de agua; IMF, índice de fragmentación miofibrilar; CTC, contenido de tejido conjuntivo; SF, fuerza de corte; TC, el total de colágeno; ISC, insoluble colágeno; CS, colágeno soluble.

NS = no significativas; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

significantly lower ($P < 0.001$) than that obtained at other storage times. There were however significant differences ($P < 0.05$) between the two breeds that had been frozen for 15 and 30 days. The muscle type had no effect on moisture content (Table 1), but frozen storage time and interaction between the muscle type and storage time was significantly affected (Table 3).

Several researchers did not find significant differences in intramuscular fat content between breeds among Moxotó and Canindé goat (Madruga et al., 2008). Also, in the present study, there were no significant effects of breed and the muscle type for IMF of the samples measured at 24 h postmortem (Table 1). However, the IMF was significantly different between Bashbay sheep and Xinjiang Merino on thawing after various times of frozen storage, but there were no differences between the different muscles and frozen storage time. Xinjiang Merino had less moisture and more IMF contents which may contribute positively to the overall eating quality.

Significant differences in pH were found among breeds (Crawford et al., 2010; Warithitham, Lambertz, Langholz, Wicke, & Gauly, 2010) and different muscles (Kadim, Mahgoub, Al-Kindi, Al-Marzooqi, & Al-Saqri, 2006), and were similar to that found in the present study for fresh meat (Table 1). Xinjiang Merino had a higher pH ($P < 0.001$) than Bashbay sheep, and M. supraspinatus had a higher pH ($P < 0.001$) compared to the M. gluteus and M. longissimus dorsi. Hopkins and Fogarty (1998) reported that there were significant differences in pH between genotypes of sheep, while some researchers did not find any differences in pH (Abdullah, Qudsieh, & Nusairat, 2011; Nurinisa et al. 2009). Sañudo et al. (1997) observed that there were no significant variations in ultimate pH among Turkish Merino, Ramlic, Kivircik, Chios, and Imroz lambs.

In our work, the pH was significantly influenced by the time of frozen storage. Also, an increase in pH with frozen storage time was found by Yuan et al. (2011). These variations in muscle pH may result from differences in proportions of red and white fiber types

and the extent or rate of glycolysis, and microbial activity due to the thawing technique (Bulent, et al. 2009; Warithitham et al., 2010).

3.2. WHC of muscles, cooking losses, and thawing losses

Cooking losses were not significant ($P > 0.05$) among the different breeds and different muscle types. Water holding capacities were significantly affected by breed and the muscle type ($P < 0.01$) with Xinjiang Merino having higher WHC than Bashbay sheep and further, the WHC was the highest in M. supraspinatus and M. gluteus than in M. longissimus dorsi (Table 1). Serra et al. (2008) reported that breed had no influence on cooking loss, but did influence the WHC. No differences in cooking losses were seen due to breed as previously described by Madruga et al. (2008) and Bulent, et al. (2009) while others have reported significant effects of breed on cooking loss. Juárez et al. (2009) reported no effects of lamb breeds on WHC.

Freezing caused a loss of meat eating quality as a result of increased thawing and cooking losses, and lower WHC. There were significant differences in thawing loss ($P < 0.05$) and WHC ($P < 0.001$) among the different breeds during frozen storage, while there were no differences in cooking loss (Table 2). Frozen storage and interaction between breed and frozen storage time, influenced cooking loss ($P < 0.05$) and WHC ($P < 0.001$). For meat that had been frozen, Bashbay sheep had the higher thawing loss and the lowest WHC. Differences in thawing losses were large and significant ($P < 0.05$), however there were no differences between different muscles (Table 3). The muscle type had a significant effect ($P < 0.01$) on cooking loss of frozen meat. Water holding capacity was influenced by the muscle type, frozen storage time, and the interaction between muscle and frozen storage time.

In the present study, thawing loss increased with increasing storage time, which is in accordance with the results found by Filguelas, Gatellier, Zambiasi, and Santé-Lhoutellier (2011).

Table 2. Means ($n = 24$) and standard error values for meat quality properties of frozen meat for different breeds (B).
 Tabla 2. Medias ($n = 24$) y valores de error estándar para las propiedades de la calidad de la carne congelada de diferentes razas (B).

parameters	Frozen storage time												Significance		
	1 day			7 days			15 days			30 days					
	Bashbay sheep	Xinjiang Merino	Bashbay sheep	Xinjiang Merino	Bashbay sheep	Xinjiang Merino	Bashbay sheep	Xinjiang Merino	Bashbay sheep	Xinjiang Merino	B	F	B × F		
Moisture (%)	68.10 ^a (0.87)	69.12 ^a (0.11)	76.01 ^b (0.20)	74.57 ^b (1.08)	74.90 ^{bx} (0.23)	73.96 ^{by} (0.22)	76.16 ^{bx} (0.18)	72.86 ^{by} (0.28)	**	***	**	***	**		
IMF (%)	3.35 (0.64)	2.53 (0.72)	1.90 ^x (0.18)	4.52 ^y (0.36)	3.28 (0.63)	4.77 (0.84)	2.28 (0.89)	4.40 (1.40)	*	NS	*	NS	NS		
pH	5.92 ^{ax} (0.01)	6.04 ^{ay} (0.03)	5.59 ^{bx} (0.04)	6.09 ^{by} (0.03)	5.77 ^{ax} (0)	5.87 ^{by} (0.02)	5.87 ^a (0.08)	5.91 ^b (0.01)	***	**	***	***	***		
TL (%)	6.20 (2.92)	4.46 (1.13)	8.14 (0.53)	5.59 (0.28)	9.46 (0.95)	7.56 (0.25)	10.79 (1.06)	3.89 (2.09)	*	NS	*	NS	NS		
CL (%)	17.18 ^{ax} (0.75)	31.52 ^y (1.71)	26.86 ^{ab} (1.61)	31.58 (1.34)	33.44 ^b (1.85)	32.40 (2.03)	34.95 ^b (5.70)	30.05 (0.71)	NS	*	NS	*	*		
WHC (%)	88.28 ^{ax} (0.31)	82.17 ^y (0.31)	72.45 ^{bx} (1.77)	83.81 ^y (0.68)	75.87 ^{bx} (1.62)	83.25 ^y (0.46)	72.78 ^{bx} (0.25)	82.57 ^y (0.97)	***	***	***	***	***		
MFI	164.20 ^x (5.35)	143.39 ^y (5.35)	183.69 ^x (4.05)	157.30 ^y (8.06)	161.11 (14.54)	170.01 (10.74)	160.26 (21.35)	167.30 (6.14)	NS	NS	NS	NS	NS		
CTC (%)	9.61 (1.26)	9.73 (0.09)	8.62 (1.11)	10.83 (1.08)	7.88 ^x (0.73)	11.80 ^y (1.18)	8.23 (1.46)	8.84 (0.61)	*	NS	*	NS	NS		
SF (kg)	7.31 ^{ax} (0.19)	5.13 ^y (0.53)	3.93 ^b (0.75)	5.12 (0.81)	6.95 ^{ax} (0.24)	5.04 ^y (0.34)	5.06 ^b (0.74)	4.56 (0.48)	**	**	**	**	**		
TC (%)	1.19 (0.19)	5.09 ^a (1.51)	1.46 (0.26)	2.50 ^{ab} (0.10)	2.42 (1.08)	1.58 ^b (0.20)	3.03 (1.51)	1.54 ^b (0.24)	NS	NS	NS	NS	*		
ISC (%)	0.95 ^x (0.19)	3.28 ^{xy} (0.74)	1.15 (0.28)	1.20 ^b (0.03)	1.59 (0.33)	1.51 ^b (0.05)	1.90 (0.57)	1.42 ^b (0.17)	NS	NS	NS	NS	**		
CS (%)	18.67 (11.97)	31.82 (7.49)	21.87 (7.25)	38.20 (17.44)	28.58 ^x (7.32)	4.89 ^y (1.24)	39.63 (14.00)	8.85 (4.52)	NS	NS	NS	NS	NS		

Notes: ^{a,b}Different superscripts in the same row represent significant differences among frozen storage times (within breed) ($P < 0.05$).
^{x,y}Different superscripts in the same row represent significant differences among breeds (within frozen storage time) ($P < 0.05$).
 IMF, intramuscular fat; TL, thaw loss; CL, cooking loss; WHC, water holding capacity; MFI, myofibrillar fragmentation index; CTC, connective tissue content; SF, Shear force; TC, total collagen; ISC, insoluble collagen; CS, collagen solubility.
 NS = not significant; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.
 Notas: ^{a,b}Diferentes superíndices en la misma fila representan diferencias significativas entre almacenamiento congelado veces (de raza) ($P < 0.05$).
^{x,y}Diferentes superíndices en la misma fila representan importantes diferencias entre razas (en almacenamiento congelado el tiempo) ($P < 0.05$).
 El IMF, la grasa intramuscular; TL, thaw pérdida; CL, la cocina pérdida; WHC, capacidad de retención de agua; MFI, índice de fragmentación miofibrilar; CTC, contenido de tejido conjuntivo; SF, fuerza de corte; TC, el total de colágeno; ISC, insoluble colágeno; CS, colágeno soluble.
 NS = no significativas; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Table 3. Means ($n = 24$) and standard error values for meat quality properties of frozen meat for different muscles (M).Tabla 3. Medias ($n = 24$) y valores de error estándar para las propiedades de la calidad de la carne congelada para diferentes músculos (M).

Parameters	Frozen storage time												significance					
	1 day				7 days				15 days							30 days		
	MS	MG	MLD	MS	MG	MLD	MS	MG	MLD	MS	MG	MLD	MS	MG	MLD	M	F	M × F
Moisture (%)	75.50 ^{ax} (0.88)	75.44 ^{ax} (0.25)	68.10 ^{ay} (0.87)	76.03 ^x (0.17)	74.15 ^{aby} (0.65)	76.01 ^{bx} (0.20)	73.87 (2.47)	74.46 ^{ab} (0.13)	74.90 ^b (0.23)	75.79 ^x (0.49)	73.36 ^{by} (0.36)	76.16 ^{bx} (0.18)	75.79 ^x (0.49)	73.36 ^{by} (0.36)	76.16 ^{bx} (0.18)	NS	*	***
IMF (%)	1.85 ^h (0.33)	2.97 ^h (0.19)	3.35 (0.64)	5.52 ^{bx} (0.61)	1.71 ^{by} (0.26)	1.90 ^y (0.18)	4.02 ^{ab} (1.50)	2.43 ^{ab} (0.49)	3.28 (0.63)	2.23 ^{bx} (0.14)	6.47 ^{zy} (0.34)	2.28 ^x (0.89)	2.23 ^{bx} (0.14)	6.47 ^{zy} (0.34)	2.28 ^x (0.89)	NS	NS	***
pH	5.93 ^{ax} (0.05)	5.77 ^{aby} (0.05)	5.92 ^{ax} (0.01)	5.89 ^{ax} (0.03)	5.71 ^{ay} (0.01)	5.59 ^{bz} (0.04)	5.96 ^{ax} (0.01)	5.79 ^{aby} (0.03)	5.77 ^{ay} (0)	6.26 ^{bx} (0.03)	5.86 ^{by} (0.03)	5.87 ^{ay} (0.08)	6.26 ^{bx} (0.03)	5.86 ^{by} (0.03)	5.87 ^{ay} (0.08)	***	***	**
TL (%)	2.02 (0.39)	6.74 (0.92)	6.20 (2.92)	3.34 (0.84)	3.94 (2.24)	8.14 (0.53)	6.17 (1.20)	9.98 (3.43)	9.46 (0.95)	9.50 (3.98)	7.75 (0.28)	10.79 (1.06)	9.50 (3.98)	7.75 (0.28)	10.79 (1.06)	NS	*	NS
CL (%)	43.19 ^x (1.52)	40.73 ^x (1.00)	17.18 ^{ay} (0.65)	37.17 (12.68)	41.05 (2.64)	26.86 ^{ab} (1.61)	37.69 (3.29)	39.57 (0.05)	33.44 ^b (1.85)	36.27 (6.96)	42.55 (1.27)	34.95 ^b (5.70)	36.27 (6.96)	42.55 (1.27)	34.95 ^b (5.70)	**	NS	NS
WHC (%)	89.84 ^d (1.61)	88.82 ^d (0.46)	88.28 ^d (0.63)	79.20 ^{oxy} (0.41)	80.70 ^{xx} (2.95)	72.45 ^{by} (1.77)	83.68 ^{xx} (1.50)	76.76 ^{by} (0.55)	75.87 ^{by} (1.62)	83.50 ^{xx} (1.39)	85.97 ^{xx} (0.75)	72.78 ^{by} (0.25)	83.50 ^{xx} (1.39)	85.97 ^{xx} (0.75)	72.78 ^{by} (0.25)	***	***	**
MFI	135.01 ^{xy} (19.43)	113.87 ^{ax} (9.51)	164.20 ^y (4.25)	140.74 (24.17)	169.77 ^b (7.01)	183.69 (4.05)	152.16 (10.95)	169.04 ^b (2.63)	161.11 (14.54)	161.24 (8.67)	116.38 ^a (13.29)	160.26 (21.35)	161.24 (8.67)	116.38 ^a (13.29)	160.26 (21.35)	*	NS	NS
CTC (%)	12.27 ^{ax} (0.37)	7.16 ^{ay} (1.44)	9.61 ^{xy} (1.26)	14.38 ^{bx} (0.46)	7.20 ^{ay} (0.68)	8.62 ^y (1.11)	12.64 ^{ax} (0.26)	11.61 ^{bx} (0.55)	7.88 ^y (0.73)	10.43 ^c (0.54)	8.68 ^a (0.28)	8.23 (1.46)	10.43 ^c (0.54)	8.68 ^a (0.28)	8.23 (1.46)	***	NS	**
SF (kg)	9.95 ^d (0.98)	9.40 ^d (1.27)	7.31 ^a (0.38)	7.21 ^{abx} (0.85)	6.85 ^{bx} (0.35)	3.93 ^{by} (0.75)	6.21 ^{ab} (1.96)	5.82 ^{bx} (0.20)	6.95 ^a (0.24)	5.61 ^{bx} (0.54)	7.87 ^{aby} (0.34)	5.06 ^{bx} (0.74)	5.61 ^{bx} (0.54)	7.87 ^{aby} (0.34)	5.06 ^{bx} (0.74)	**	*	NS
TC (%)	3.30 (0.56)	2.98 ^d (1.10)	1.19 (0.19)	5.03 ^x (1.00)	2.65 ^{by} (0.50)	1.46 ^y (0.26)	3.28 (0.95)	2.57 ^{ab} (0.33)	2.42 (1.08)	3.60 (0.60)	4.08 (0.52)	3.03 (0.51)	3.60 (0.60)	4.08 (0.52)	3.03 (0.51)	**	NS	NS
ISC (%)	2.14 ^x (0.21)	1.72 ^x (0.14)	0.95 ^y (0.19)	3.33 ^x (0.99)	2.53 ^{xy} (0.10)	1.15 ^y (0.28)	1.72 (0.32)	2.36 (0.34)	1.59 (0.55)	2.84 (0.57)	2.08 (0.23)	1.90 (0.57)	2.84 (0.57)	2.08 (0.23)	1.90 (0.57)	**	NS	NS
CS (%)	33.47 (4.38)	30.39 ^{ab} (16.98)	18.67 (11.97)	33.96 (18.66)	4.10 ^a (1.56)	21.87 (7.25)	43.05 ^x (10.21)	8.73 ^{xy} (3.68)	28.58 ^{xy} (7.32)	21.54 (4.94)	48.72 ^b (3.80)	39.63 (14.00)	21.54 (4.94)	48.72 ^b (3.80)	39.63 (14.00)	NS	NS	NS

Notes: ^{a,b,c}Different superscripts in the same row represent significant differences among frozen storage times (within individual muscle) ($P < 0.05$).^{x,y,z}Different superscripts in the same row represent significant differences among individual muscles (within frozen storage time) ($P < 0.05$).

MS, M. supraspinatus; MG, M. gluteus; MLD, M. longissimus dorsi; IMF, intramuscular fat; C.L., cooking loss; WHC, water holding capacity; MFI, myofibrillar fragmentation index; CTC, connective tissue content; SF, Shear force; TC, total collagen; ISC, insoluble collagen; CS, collagen solubility.

NS = not significant; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.Notes: ^{a,b,c}Diferentes superíndices en la misma fila representan diferencias significativas entre almacenamiento congelado veces (dentro de cada músculo) ($P < 0.05$).^{x,y,z}Diferentes superíndices en la misma fila representan diferencias significativas entre los músculos individuales (en almacenamiento congelado el tiempo) ($P < 0.05$).

El MS, M. supraspinatus; MG, M. glúteos; MLD, M. longissimus dorsi; IMF, grasa intramuscular; CL, la cocina perdida; WHC, capacidad

de retención de agua; ISC, índice de fragmentación miofibrilar; CTC, contenido de tejido conjuntivo; SF, fuerza de corte; TC, el total de colágeno; ISC, insoluble colágeno; CS, colágeno soluble.

NS = no significativas; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

Crouse and Koohmaraie (1990) found that cooking loss increased with increasing time of storage, contrarily Filgueras et al. (2011) reported there was a drop in cooking loss after freezing. Sebranek, Sang, Topel, and Rust (1979) did not find any changes in WHC with frozen storage time.

The variations in thawing loss and cooking loss among breeds and muscles may be due to water holding capacity which was associated with ultimate meat pH and rates of pH decline (Bulent et al., 2009), initial water content of the muscle (Abdullah et al., 2011), and differences in postmortem degradation of muscle proteins (Melody et al., 2004). Differences in cooking loss also related to the fat level of the muscles and the time and temperature of cooking. The differences in thawing loss, cooking loss, and WHC of frozen meat at different storage time have been mainly attributed to denaturation of muscle proteins and extensive fragmentation and fracturing of myofibril (Berry et al., 1971).

3.3. MFI of muscles

In this work, the MFI was significantly affected by breed ($P < 0.05$), but not influenced by the muscle location when measured at 24 h postmortem (Table 1). During frozen storage, the muscle type had a significant effect on MFI, but no differences existed among breeds, storage times, and interactions between muscle×freezing and breed×freezing (Tables 2 and 3). Whipple, Koohmaraie, Dikeman, and Crouse (1990) found no differences in MFI among breeds. Differences in MFI have also been shown to result from variations in time and speed of homogenization (Hopkins et al., 2000). MFI was related to tenderness. The figures in Xinjiang Merino were increased from 1 day to 15 days, and then decreased to 30 days. According to the variation in MFI, Xinjiang Merino was tender than Bashbay sheep at the end of frozen storage time.

3.4. Connective tissues

The connective tissue content, total, and insoluble collagen content and collagen solubility of fresh meat and frozen meat are shown in Tables 1–3.

Breed had no effect on the connective tissue content of fresh meat (Table 1), but had significant ($P < 0.05$) impact on that from frozen meat (Table 2). Xinjiang Merino had a higher connective tissue content than Bashbay sheep; the difference was significant ($P < 0.05$) at 15 days of frozen storage. The effect of freezing on connective tissue content between breeds was not found ($P > 0.05$).

The muscle type had a significant influence on connective tissue content ($P < 0.001$) during frozen storage, and the effect of the muscle type was greater than breed (Table 3). Significant interactions ($P < 0.01$) were found between the muscle type and frozen storage. Prost, Pelczynska, and Kotula (1975) reported there were significant differences among seven muscles, and the connective tissue content of the M. psoas major was the lowest, and infraspinatus was the highest. In this experiment, the connective tissue content of M. supraspinatus was higher than those of M. gluteus and M. longissimus dorsi.

The total collagen content of fresh meat was influenced by the muscle type ($P < 0.05$), but was not affected by breed ($P > 0.05$). Breed and the muscle type had a significant influence on the insoluble collagen content, but had no effect on collagen solubility. In the frozen meat, there were no significant differences in the total collagen,

insoluble collagen or collagen solubility among breeds and storage time (Tables 2 and 3). The interaction between both factors had a significant impact on the total collagen content ($P < 0.05$) and insoluble collagen content ($P < 0.01$). Significant differences among individual muscles existed for the total collagen content and insoluble collagen content during frozen storage.

Many studies have indicated that collagen content varied among breeds (Sañudo et al., 2004; Serra et al., 2008). However, other reports have noted that breed had no effect on collagen content (Warithitham et al., 2010). Nurinisa et al. (2009) observed that breed and muscle had significant effect on collagen. Differences in collagen solubility among breeds and muscles was found by Mette et al. (2011), this difference was attributed to the nutritional level and age of animal.

3.5. SF of muscles

Wheeler and Koohmaraie (1999) reported that SF was influenced by individual muscles type. We also observed differences with WB value being greater ($P < 0.01$) in M. gluteus than in the M. supraspinatus and M. longissimus dorsi of fresh samples, but there was no breed effect on shear force (Table 1). Also Madruga et al. (2008) did not find any differences in SF among breeds. However, there are many reports that find significant effects of breed on shear force values and appears to be dependent upon breeds being compared (Ann-Charlotte et al., 1997; Sañudo et al., 2004). The variations in shear force among breeds and muscles were probably due to differences in connective tissue content, collagen characteristics, and sarcomere length of the myofiber, fiber size, and fiber-type composition (Abdullah et al., 2011).

SF of frozen meat differed ($P < 0.01$) among breeds, individual muscles, freezing and there was an interaction between breed and freezing. There were significant differences in SF values of meat between Bashbay sheep and Xinjiang merino that had been frozen storage, and the SF value decreased with increasing time of storage except 7 days in Bashbay sheep. This was similar in previously reported data for lamb (Duckett et al., 1998) and pork (Van Laack, Stevens, & Stalder, 2001). The differences in shear force among the two breeds were significant at day 1 and 15 of frozen storage. After frozen storage, the SF gradually declined. M. supraspinatus had the highest shear force value, while M. longissimus dorsi had the lowest shear force value.

3.6. Relationship between physico-chemical properties and tenderness

In the present study intramuscular fat, cooking loss, water holding capacity, and MFI were related to shear force value (Table 4). The shear force value and intramuscular fat content exhibited high negative correlation ($P < 0.01$), and intramuscular fat content was correlated with cooking loss, water holding capacity, and MFI.

There are many reports on the influence of intramuscular fat on meat tenderness (DeVol et al., 1988; Ann-Charlotte et al., 1997) and breed had influence on intramuscular fat (Van Laack et al., 2001). The water holding capacity has a large influence on tenderness, and these results confirm the earlier studies (Jeremiah et al., 1999; Melody et al., 2004). Muscles that were most tender exhibited the lowest shear force values and the highest MFI. The intramuscular fat, cooking loss, and shear

Table 4. Correlation coefficients between meat quality properties and frozen storage time and tenderness.

Tabla 4. Los coeficientes de correlación entre las propiedades la calidad de la carne congelada y tiempo de almacenamiento y la ternura.

	Moisture	IMF	pH	TL	CL	WHC	MFI	CTC	SF	TC	ISC	CS
Moisture	1											
IMF	0.253	1										
pH	-0.215	-0.198	1									
TL	0.415	0.825	0.257	1								
CL	0.049	0.973**	-0.160	0.967*	1							
WHC	-0.241	-0.932*	0.518	-0.492	-0.894*	1						
MFI	0.129	0.881*	-0.601	0.417	0.871	-0.986**	1					
CTC	0.299	-0.266	-0.642	0.121	-0.271	0.090	-0.009	1				
SF	-0.284	-0.972**	0.384	-0.597	-0.924*	0.987**	-0.948*	0.198	1			
TC	-0.434	0.631	-0.117	-0.585	0.689	-0.672	0.690	-0.613	-0.669	1		
ISC	-0.319	0.722	-0.226	-0.542	0.763	-0.779	0.794	-0.526	-0.771	0.987**	1	
CS	-0.299	-0.294	0.847	-0.259	-0.294	0.484	-0.561	-0.821	0.376	0.115	-0.015	1
Frozen storage time	0.186	0.972**	0.024	0.894	0.966**	-0.824	0.760	-0.379	-0.892*	0.597	0.666	-0.141

Notes: IMF, intramuscular fat; CL, cooking loss; WHC, water holding capacity; MFI, myofibrillar fragmentation index; CTC, connective tissue content; SF, Shear force; TC, total collagen; ISC, insoluble collagen; CS, collagen solubility.

* $P < 0.05$; ** $P < 0.01$.

Notas: El IMF, la grasa intramuscular; CL, la cocina perdida; WHC, capacidad de retención de agua; IMF, índice de fragmentación miofibrilar; CTC, contenido de tejido conjuntivo; SF, fuerza de corte; TC, el total de colágeno; ISC, insoluble colágeno; CS, colágeno soluble.

* $P < 0.05$; ** $P < 0.01$.

force were significantly affected by frozen storage time. As the intramuscular fat was increased, and the cooking loss and shear force values decreased with the increasing storage time, the tenderness tended to improve; possibly as a result of calpain and catheptic activities (Whipple & Koohmaraie, 1992; Whipple et al., 1990).

4. Conclusion

In the present article, we reported on some chemical and physical properties of two breeds and three muscles during frozen storage time. The results indicate that tenderness improved with increasing time of frozen storage. For frozen meat, the intramuscular fat content, cooking loss, water holding capacity, and MFI were most closely related to tenderness of lamb. Generally, breed and the muscle type had significant effects on meat tenderness during storage. As the time of frozen storage increased from 1 day to 15 days, the intramuscular fat, cooking loss, water holding capacity, and MFI in Xinjiang merino were increased gradually. Shear force and tenderness were improved. Compared with Bashbay sheep, Xinjiang merino was tenderer and thus preferred by consumers.

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Disclosure statement

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