

# Effect of Transportation and Low Voltage Electrical Stimulation on Meat Quality Characteristics of Omani Sheep

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تأثير النقل والتحفيز الكهربائي ذو الجهد المنخفض على جودة لحوم الأغنام العمانية

عصام توفيق كاظم و عثمان محجوب جعفر ووليد المرزوقي وسميرة قاسم خلف

الخلاصة: يهدف هذا البحث لدراسة تأثير نقل الحيوانات بسيارة مفتوحة خلال الفصل الحار (٣٦ درجة مئوية) واستخدام التحفيز الكهربائي لذبائح الأغنام العمانية على جودة اللحم. تم تقسيم عشرين رأساً من ذكور الأغنام بعمر سنة بالتساوي الى مجموعتين: تم نقل المجموعة الأولى لثلاث ساعات بسيارات نقل مفتوحة لمسافة ٣٠٠ كيلومتر، بينما تم إيواء المجموعة الثانية في حظائر المسلخ المركزي لفترة ثلاثة ايام قبل الذبح مع توفير الماء والأكل. تم جمع عينات الدم من جميع الحيوانات قبل عملية النقل والذبح لقياس تراكيز هرمونات الأجهاد. استخدم التحفيز الكهربائي (٩٠ فولت) على ٥٠٪ من الذبائح المختارة عشوائياً بعد مرور ٢٠ دقيقة من الذبح. جمعت عينات من العضلة العينية لدراسة الصفات النوعية للحوم مثل الحموضة والطرارة والعصارية ونسبة فقد الماء وطول الليفة العضلية ومقياس تكسر اللويحات العضلية و اللون. أظهرت النتائج وجود زيادات ملحوظة في تركيز هرمونات الكورتيزول والادرينالين والغير أدرينالين و الدوبامين في دم الحيوانات المنقولة مقارنة بالمجموعة الغير منقولة. كان للتحفيز الكهربائي تأثير ملحوظ بانخفاض حموضة اللحم وزيادة طول العضلة الليفية والطرارة والعصيرية ومقياس تكسر اللويحات العضلية واللون مقارنة بالغير محفزة كهربائياً. كانت التأثيرات السلبية لعملية النقل على الصفات النوعية للحوم واضحة بتقليل الطرارة وحموضة اللحم والعصارية واللون الداكن مقارنة بالمجموعة الثانية. أكدت هذه الدراسة بأن نقل الأغنام خلال الفصل الحار يؤدي إلى تدهور الصفات النوعية في لحوم الأغنام وان استخدام التحفيز الكهربائي يقلل من التأثيرات السلبية للنقل.

ABSTRACT: The aim of this study was to determine the effects of road transportation during the hot season (36 °C) and low voltage electrical stimulation on meat quality characteristics of Omani sheep. Twenty intact male sheep (1-year old) were divided into two equal groups: 3 hrs transported or non-transported. The transported group was transferred to the slaughterhouse the day of slaughter in an open truck covering a distance of approximately 300 km. The non-transported group was kept in a lairage of a commercial slaughterhouse with *ad libitum* feed and water for 3 days prior to slaughter. Blood samples were collected from the animals before loading and prior to slaughter in order to assess their physiological response to stress in terms of hormonal levels. Fifty percent of the carcasses from each group were randomly assigned to low voltage (90 V) at 20 min postmortem. Muscle ultimate pH, expressed juice, cooking loss percentage, WB-shear force value, sarcomere length, myofibrillar fragmentation index and colour  $L^*$ ,  $a^*$ ,  $b^*$  were measured on samples from *Longissimus dorsi* muscles collected 24 hrs postmortem at 2-4 °C. The transported sheep had significantly ( $P<0.05$ ) higher cortisol, adrenaline, nor-adrenaline, and dopamine levels than the non-transported group. Muscles from electrically-stimulated carcasses had significantly ( $P<0.05$ ) lower pH values, longer sarcomere length, lower shear force value, higher expressed juice, myofibrillar fragmentation index and  $L^*$  values than those from non-stimulated ones. Transportation significantly influenced meat quality characteristics of the *Longissimus dorsi* muscle. Muscle ultimate pH and shear force values were significantly higher, while CIE  $L^*$ ,  $a^*$ ,  $b^*$ , expressed juice and cooking loss were lower in transported than non-transported sheep. This study indicated that pre-slaughter transportation at high ambient temperatures can cause noticeable changes in muscle physiology in sheep. Nevertheless, meat quality of transported sheep can be improved by electrical stimulation post-slaughter.

Keywords: Sheep, transportation, *Longissimus dorsi*, electrical stimulation, shear force.

## Introduction

Transportation of small ruminants in open trucks between farms and slaughterhouse is a routine practice in the Sultanate of Oman. Transport of live animals has been recognized as a stressful event that has significant economic and welfare implications for animals (Schrama *et al.*, 1994; Kadim *et al.*, 2006). Vibrations and movement of the vehicle are novel to the animals, and therefore likely to elicit a stress response (Dantzer and Mormede, 1983). Adverse climatic conditions such as high or low temperatures and high relative humidity

are also additional stressors to animals during transport. Animals are inevitably exposed to handling, loading and transportation. All these stressors may increase catecholamine blood concentrations, which may result in compromising cellular and humoral immune functions, reproduction, digestion, growth and other metabolic processes (Dantzer and Mormede, 1983; Nelson and Drazen, 2000). This may also cause metabolic changes that can in turn adversely affect meat quality (Ashmore *et al.*, 1972; Schrama *et al.*, 1994;

Apple *et al.*, 1995; Kannan *et al.*, 2003; Bond *et al.*, 2004; Kadim *et al.*, 2006). Transportation usually imposes stress on animal and can lead to depletion of muscle glycogen reserves before slaughter that subsequently increase the ultimate pH of meat, and result in low meat quality characteristics (Kadim *et al.*, 2006). Meat tenderness is one of the most important quality characteristics affected by pH and temperature (Marsh *et al.*, 1981). Colour is also an important physical property of meat and consumers usually use it as a quality indicator. Moreover, consumers are becoming more aware of the ethics of meat production and prefer meat that has been produced from animals that have not been mistreated (Warris, 1995).

Electrical stimulation can increase postmortem muscle metabolism and hasten the onset of rigor mortis, which might improve the quality characteristics of stressed animals. A rapid pH decline of electrically stimulated carcasses could potentially result in a brighter coloured meat (King *et al.*, 2004). The objective of the present study was to investigate the impact of road transport and low electrical-stimulation on meat quality characteristics of Omani sheep *Longissimus dorsi*.

## Materials and Methods

### *Animals and Treatments*

Twenty Omani intact male sheep (1-year-old) were randomly selected from a homogenous flock fed Rhodesgrass hay and a commercial ruminant concentrate at the Agricultural Experiment Station, Sultan Qaboos University. The experiment was conducted during the hot season when the average ambient temperature was 36°C. Animals were equally divided into 2 groups and randomly assigned to either non-transported or transported groups. Three days prior to slaughter, the non-transported animals were transferred to a pen under shade in a lairage at Muscat Central Slaughterhouse (40 km). Feed and water were provided *ad libitum*. On the day of slaughter, the transported animals were transported in an open truck for 3 hrs for a distance of approximately 300 km. A blood sample was collected prior to loading (initial), while a second blood sample collection was taken immediately after the transportation and prior to slaughter. The two blood samples were collected from the non-transported animals at the same time as the transported animals.

### *Blood Sampling and Analysis*

Blood samples were collected in vacutainer tubes containing 81  $\mu$ L of 15% EDTA as an anticoagulant. The plasma was separated by centrifugation at 5 °C for 10 minutes at 3000 rpm then placed in 1.5 ml Eppendorf tubes and stored at -80 °C. Chemiluminescence immunoassay was used for the determination of plasma hormone levels using a Beckman Coulter Access 2 immunoassay system and reagents. (Beckman Coulter, Inc.). For the extraction of plasma catecholamines (all reagents Chromsystems GmbH), 75mg of acid washed alumina was placed in a 2.0 ml Eppendorf tube and then 750  $\mu$ L of extraction buffer, 750

$\mu$ L of plasma, and 100  $\mu$ L of dihydroxybenzoic acid (DHBA) standard 12 ng/ml were added. This mixture was vortexed for 20 minutes using an autovortex and then centrifuged at 5500 rpm (ALC International microcentrifuge model # 4214) for 3 minutes and then the supernatant was aspirated. The resulting pellet was washed with 1 ml washing buffer. The mixture was then vortexed as before, centrifuged for 3 minutes, and the wash buffer carefully aspirated. The washing process was repeated three times. To retrieve the catecholamines from the alumina, the pellet was eluted using 240  $\mu$ L elution buffer and vortex for 7 minutes, using the autovortex, centrifuged at 11500 rpm for 5 minutes and the supernatant containing the catecholamines and internal standard was transferred carefully to a clean vial without disturbing the alumina layer. This supernatant was immediately analyzed using an HPLC with an electrochemical detector (Waters 600S, 464 ECD and 717 Autosampler). Results were acquired and processed using Millennium<sup>32</sup> software (Waters).

### *Carcass Electrical Stimulation*

Animals were slaughtered and dressed at the Muscat Municipality Central Slaughterhouse following Halal methods. A random 50% of the carcasses within each group were electrically stimulated for 60s, 20 minutes postmortem using a low-voltage stimulator (Voltage, 90 V; AgResearch, V1.3-R3B. New Zealand). Carcasses were kept at ambient temperature (20-25 °C) until placed in a chiller (2-4 °C) at 100 $\pm$ 5 min postmortem.

### *Meat Quality Evaluation*

The carcasses were chilled for 24 h at 2-3 °C before the *Longissimus dorsi* muscles were removed from the left. The muscles were placed in plastic bags then frozen at -20 °C until processing. The muscles were evaluated for a range of quality characteristics including ultimate muscle pH, expressed juice, percent cooking loss, Warner-Bratzler shear force value, sarcomere length, myofibrillar fragmentation index and colour  $L^*$ ,  $a^*$ ,  $b^*$  were determined. The ultimate pH was assessed in homogenates at 20-22 °C (using a Ultra Turrax T25 homogenizer) of duplicate 1.5-2 g of muscle tissue in 10 ml of neutralized 5-mM sodium iodoacetate and the pH of the slurry measured using a Metrohm pH meter (Model No. 744) with a glass electrode. Chilled muscle samples (13 mmx13 mm cross section) for assessment of shear force by a digital Dillon Warner-Bratzler (WB) shear were prepared from muscle samples cooked in a water bath at 70 °C for 90 min. The cooked samples were carefully dried with tissues to remove excess surface moisture and re-weighed to determine cooking losses. Sarcomere length by laser diffraction was determined using procedure described by Cross *et al.* (1980/1981). Expressed juice was assessed by a filter paper method, as the total wetted area less the meat area (cm<sup>2</sup>) relatively to the weight of the sample (g). Myofibrillar fragmentation index (MFI) was measured using a modification of the

**Table 1.** Effects of road transportation on hormonal concentrations in plasma of Omani sheep.

No. Samples		Treatment		SEM
		Transported	Non-transported	
		10	10	
Cortisol (nmol/l)	Initial	20.9 <sup>a</sup>	20.6	2.91
	Final	38.9 <sup>b</sup>	21.1	2.65
	Sign.	**	NS	
Adrenaline (ng/ml)	Initial	1.61 <sup>a</sup>	1.59	0.15
	Final	2.15 <sup>b</sup>	1.60	0.34
	Sign.	*	NS	
Dopamine (ng/ml)	Initial	0.72 <sup>a</sup>	0.71	0.17
	Final	0.99 <sup>b</sup>	0.71	0.13
	Sign.	*	NS	
Nor-adrenaline (ng/ml)	Initial	0.19 <sup>a</sup>	0.20	0.021
	Final	0.38 <sup>b</sup>	0.21	0.027
	Sign.	*	NS	

SEM: Standard error of mean, Sign. NS not significant, \* P<0.05, \*\* P<0.01.

method of Johnson *et al.* (1990). This measured the proportion of muscle fragments that passed through a 231- $\mu$ m filter after the sample had been subjected to a standard homogenization treatment. A 5 g ( $\pm$ 0.5 g) sample of diced (6 mm<sup>3</sup> pieces) was added to 50 ml of cold physiological saline (85% NaCl) plus five drops of antifoam A emulsion (Sigma Chemical A5758) in a 50 ml graduated cylinder, and homogenized at ¼ speed using an 18 mm diameter shaft on an Ultra-Turrax homogenizer for 30-s periods separated by a 30-s rest period. The homogenate was poured into a weighed filter (231 x 231  $\mu$ m holes). The filter typically ceased dripping after 2-3 h, at which time they were dried at 26-28 °C in an incubator for 40 h before being reweighed. The MFI values presented herein were calculated as 100 minus the percentage of the initial meat sample weight that remained on the filter. Approximately 60 min after exposing the fresh surface, CIE  $L^*$ ,  $a^*$ ,  $b^*$  light reflectance coordinates of the muscle surface were measured at room temperature (25 $\pm$ 2 °C) using Minolta Chroma Meter CR-300 (Minolta Co., Ltd., Japan), with a colour measuring area 1.1 cm diameter. It was calibrated using a Minolta calibration plate ( $L^*$  =97.59,  $a^*$  =-5.00,  $b^*$  = +6.76). The  $L^*$  value relates to Lightness; the  $a^*$  value to Red-Green hue where a positive value relates

to the red intensity; and the  $b^*$  value to the Yellow-Blue where a positive value relates to yellow. The average of two measurements from each sample was recorded as the colour coordinate value of the sample.

#### Statistical Analysis

The effect of transportation and electrical stimulation on blood serum parameters, and meat quality were analyzed using GLM procedures for analysis of variance procedure (SAS, 1993). The first blood sample was regarded as an initial value and the second blood sample as final value. Both samples were taken from the same animal; therefore SAS repeated measurement analysis was used. Differences between means were assessed using the least-significant-difference procedure. Interaction between the transportation and electrical stimulation were excluded from the model when not significant (P>0.05).

#### Results and Discussion

##### Blood Serum Hormonal Levels

Physiological parameters such as cortisol, dopamine, adrenaline and nor-adrenaline have been proposed as sensitive indicators of physiological stress response in

Table 2. Effect of transportation and electrical stimulation on a range of *Longissimus dorsi* muscle quality characteristics (Least square means) of Omani sheep.

	Transported		Non-transported		SEM	Significance		
	Stimu- lated	Non- Stimulated	Stimu- lated	Non- Stimulated		Transport	Stimu- lation	Interaction
No. samples	5	5	5	5				
Ultimate pH	5.72 <sup>b</sup>	5.86 <sup>c</sup>	5.61 <sup>b</sup>	5.71 <sup>b</sup>	0.05	*	*	NS
Expressed juice <sup>2</sup>	26.5 <sup>b</sup>	23.7 <sup>a</sup>	30.0 <sup>c</sup>	26.2 <sup>b</sup>	1.53	*	*	NS
Cooking loss%	22.7 <sup>b</sup>	20.9 <sup>a</sup>	26.7 <sup>c</sup>	22.8 <sup>b</sup>	0.64	*	*	NS
Shear force value	5.17 <sup>b</sup>	7.35 <sup>c</sup>	3.63 <sup>a</sup>	5.21 <sup>b</sup>	0.61	**	**	NS
Sarcomere length (µm)	1.49 <sup>a</sup>	1.67 <sup>b</sup>	1.51 <sup>ab</sup>	1.75 <sup>c</sup>	0.07	*	*	NS
Myofibrillar fragmentation index	78.8 <sup>b</sup>	76.9 <sup>a</sup>	87.7 <sup>d</sup>	82.9 <sup>c</sup>	0.63	*	*	*
L* (lightness)	36.7 <sup>ab</sup>	36.2 <sup>a</sup>	38.4 <sup>b</sup>	37.5 <sup>ab</sup>	0.73	NS	*	NS
a* (redness)	14.4 <sup>ab</sup>	13.8 <sup>a</sup>	14.2 <sup>ab</sup>	15.7 <sup>b</sup>	0.58	NS	NS	*
b* (yellowness)	5.55	5.19	5.22	5.93	0.26	NS	NS	*

<sup>abc</sup>Means within the same row with different superscripts were significantly different (P<0.05). <sup>2</sup> Expressed juice = water area (cm<sup>2</sup>) / sample weight (g). <sup>3</sup> Warner-Bratzler shear force value.

animals that have been exposed to road transportation (Kadim *et al.*, 2006). In the current study, blood parameter values were compared between the transported and non-transported sheep groups to investigate the effects of road transportation on sheep at high ambient temperatures (36 °C) (Table 1). The ambient temperature in the present study was above the suggested upper limit of ruminant heat tolerance (35–40 °C) (Yousef, 1985; Lu, 1989). According to Ali *et al.* (2006), measurement of cortisol is a valuable tool in stressed animals due to a graduated response depending on the severity of the stressor. In the present study, cortisol concentrations were still significantly (P<0.01) higher 3 hrs after the start of transportation. A similar conclusion was reported by Kadim *et al.* (2006). Nwe *et al.* (1996) and Kannan *et al.* (2000) reported that plasma cortisol concentration increased within 30 min after the beginning of transport and reached a peak value at 1 hr in goats. The Omani sheep transported the same day to slaughter had significantly higher (P<0.01) plasma cortisol levels than their non-transported counterpart sheep. This indicates that 3 hrs of transportation in an open truck under Omani conditions (36 °C) appears to produce stress in sheep. Broom *et al.* (1996), Al-Kindi *et al.* (2005) Ali *et al.* (2006) and Kadim *et al.* (2006) examined a range of plasma stress indicators in transported sheep and found that plasma cortisol levels were higher in transported sheep during 2–3 hrs of the transportation in comparison with the non-transported animals. Moreover, Ruiz-De-La-Torre *et al.* (2001) found that the level of cortisol was higher after 4 hrs on a rough journey. The lower hormone values in the non-transported sheep indicate that there was sufficient time for recovery from stress and also the shorter distance and time of transport.

Adrenaline, dopamine and nor-adrenaline concentrations were significantly (P<0.05) higher in the transported

than non-transported sheep (Table 1). Similarly, Kadim *et al.* (2006) found that transportation of Omani sheep for 2 hrs significantly increased concentrations of adrenaline, dopamine and nor-adrenaline compared to non-transported animals. Adrenaline levels increased in sheep after 10 min of transportation with little change in nor-adrenaline level (Parrott *et al.*, 1994). These findings indicated that levels of these hormones stay elevated for at least 3 hrs after transportation. Release of adrenaline enables animals to mobilize body resources quickly for metabolic requirements in response to stress (Dantzer and Mormede, 1983).

### Meat Quality

An elevated ultimate pH is a consequence of low pre-slaughter glycogen as a result of pre-slaughter stress including road transportation (Gregory and Grandin, 1998; Geesink *et al.*, 2001; Kannan *et al.*, 2003; Honikel, 2004; Kadim *et al.*, 2006). Meat from transported sheep had significantly (P<0.05) higher ultimate pH value than meat from sheep that were non-transported (Table 2). Kadim *et al.* (2006) also found that 2 hrs-transportation had a significant effect on muscle-ultimate pH of the Omani sheep. Different types of stressors may elicit varying degrees of responses in animals. Ruiz-De-La-Torre *et al.* (2001) found that transportation of sheep on a rough road for 4 hrs significantly increased ultimate pH compared to those transported on a smooth road. Similarly, muscles from stressed sheep had significantly higher ultimate pH values than non-stressed counterparts (Apple *et al.*, 1995). Transportation not only includes physical stress, but also emotional stress caused by loading and unloading, noise, vibration and social disruptions. The effort needed by the sheep to keep their balance while the vehicle moves is demanding in terms of energy requirements leading to

depletion of glycogen and consequently decreasing muscle pH (Tarrant and Grandin, 1993; Apple *et al.*, 1995).

The ultimate pH of muscle is a major determinant of meat quality (Watanabe *et al.*, 1996) and is related to the rate of glycogen and liberation of lactic acid via glycolysis pre- and post-slaughter. The pH affects meat tenderness and water-holding capacity (Watanabe *et al.*, 1996). Ultimate pH value of meat is the result of a combination of many factors including pre-slaughter handling, postmortem treatment, glycogen store and muscle physiology (Ashmore *et al.*, 1973; Marsh, 1977; Thompson, 2002). There were significant ( $P<0.05$ ) differences in ultimate pH at 24 hrs postmortem between the stimulated and non-stimulated muscles of sheep (Table 2). In the present study, electrical stimulation appears to increase early postmortem glycolysis in muscle samples (Table 2). Similarly, Geesink *et al.* (2001) found that ultimate pH from stimulated sheep carcass was significantly lower than that of non-stimulated carcass. In contrast, Bond *et al.* (2004) and Devine *et al.* (2006) have found that ultimate pH of *Longissimus* muscle was not affected by electrical stimulation.

Water retention of meat is primarily caused by immobilization of tissue water within the myofibrillar system (Hamm, 1981). Applying pressure can cause a shift of water from the intercellular into the extracellular space and then onto the meat surface as a result of structural alterations at the level of the myofilament structure. Expressed juice of sheep muscle was significantly affected by transportation and low voltage electrical stimulation (Table 2). Samples from transported sheep had significantly ( $P<0.05$ ) lower expressed juice and cooking loss than muscles from non-transported ones (Table 2). Decreased cooking loss percentage is a reflection of the increased water-holding capacity (decrease expressed juice) associated with meat of high ultimate pH (Bouton *et al.*, 1971). Similarly, Apple *et al.* (1995) found that muscles from stressed lambs had lower ( $P<0.01$ ) cooking loss than that from non-transported animals. In contrast, Bond *et al.* (2004) found that muscles from exercise-stressed sheep were significantly higher than those from non-exercised animals. They concluded that the mechanism causing greater water loss in the muscle of exercise-stressed lambs is unknown.

Expressed juice was significantly higher ( $P<0.05$ ) for stimulated than for non-stimulated muscle samples. The increase in myofibrillar expressed juice of electrically-stimulated muscles may be partly due to denatured sarcoplasmic proteins in the myofibrillar fraction (Eikelenboom and Smulders, 1986). However, Whiting *et al.* (1981) and Bond *et al.* (2004) found that electrical stimulation had no effect on water-holding capacity of lamb *Longissimus dorsi* muscle. The differences between samples from stimulated and non-stimulated muscle may probably be due to shrinkage of myofibrils as the postmortem pH fall causes denaturation of protein (Offer and Knight, 1988). The decline in pH after slaughter

was much slower in non-stimulated than stimulated *Longissimus dorsi* muscles. Moreover, the thin and thick filaments interaction (sarcomere length) significantly ( $P<0.05$ ) differed between stimulated and non-stimulated muscles (Table 2).

Muscles from non-transported sheep had a significantly lower shear force value compared to transported sheep (Table 2). In agreement with the present study, Kadim *et al.* (2006) found that meat from non-transported sheep was significantly more tender than that from transported ones. Apple *et al.* (1995) also found that muscles from non-transported sheep had lower shear values than stressed ones. In contrast, Chrystall *et al.* (1982) found that the muscle from sheep chased to exhaustion by dogs was tenderer than those from their non-transported counterparts. Decreased glycogen reserves and increased muscle temperature during transport resulted in muscle with a pH above 6.0 (Marsh, 1983), which may activate calpain proteases (Koochmaraie, 1988). In accordance with the shear-value results, muscles from the transported sheep had significantly ( $P<0.05$ ) shorter sarcomere lengths than those from non-stimulated animals (Table 2).

Muscles from non-stimulated carcasses had a significantly ( $P<0.01$ ) higher shear force value compared to stimulated carcasses (Table 2). The most positive advantage for electrical stimulation observed in the present study was similar to reports by Chrystall and Hagyard (1976); Riley *et al.* (1981); Kadim *et al.* (1993); Polidori *et al.* (1999); Geesink *et al.* (2001); and Devine, *et al.* (2006). According to Kadim *et al.* (1993), electrical stimulation of muscles soon after slaughter improved tenderness most probably by hastening the onset of rigor mortis rather than through processes that rapidly reduce muscle pH in sheep and consequently avoid toughening effects of cold shortening and thaw shortening. Shear-force values of muscles chilled at different rates were significantly reduced by electrical stimulation compared with non-stimulated ones (Shorthose *et al.*, 1986). Hwang *et al.* (2003) stated that stimulation may improve tenderness either through effects on physical alteration and/or acceleration of energy turnover during and after the stimulation. Moreover, Ho, *et al.* (1996) and Luo, *et al.* (2008) found a relationship between myofibrillar disruption and improved tenderness of meat. Luo *et al.* (2008) reported that stimulation resulted in ultrastructural changes in beef *Longissimus* muscle, which lowers the resistance to shearing force (Hopkins *et al.*, 2000). In the present study, electrically-stimulated sheep *Longissimus dorsi* muscle had significantly ( $P<0.05$ ) longer sarcomere length than non-stimulated muscles. Similar findings were reported by Whiting *et al.* (1981) and Geesink *et al.* (2001) for lambs.

The *Longissimus dorsi* muscle from transported sheep had significantly lower ( $P<0.05$ ) CIE  $L^*$  value than non-transported counterpart (Table 2). This indicates that muscles from transported sheep were darker than muscle from non-transported sheep. The lower  $L^*$  values of

muscle from transported sheep in the present study are similar to those reported by Apple, *et al.* (1995) who found lower values of  $L^*$  for muscle from lambs subjected to stress. The type and intensity of transportation had an effect on meat colour. Ruiz-De-La-Torre *et al.* (2001) found that redness of sheep muscle which was transported on a rough road for 4 hrs was significantly higher than for those transported smoothly. Bond *et al.* (2004) found that the colour of meat from stressed sheep was darker than those from non-transported animals. When muscle pH increased to above 6.0, CIE  $a^*$  values decreased below 16.0 (Apple *et al.*, 1995). In the present study, mean  $a^*$  values of the *Longissimus dorsi* muscles from transported sheep were below 16.0, indicating that the dark colour was effectively produced by 3 hrs of transportation due to depletion of muscle glycogen reserves before slaughter (Warriss *et al.*, 1990). Muscle darkness is highly related to pH change and occurs at a pH of approximately 6.0 (MacDougall and Jones, 1981). Mitochondrion oxygen uptake is more active at pH values greater than 6.0 (Lawrie, 1958). Postmortem glycolysis acid formation is insufficient to prevent mitochondrial respiration, thus allowing myoglobin to be deoxygenated and causing the muscle to remain dark (Egbert and Cornforth, 1986). Ultimate pH can influence colour independently of meat myoglobin content (Ledward, 1985), which may explain differences between the transported and non-transported muscles in the present study.

The color of meat is influenced by several individual factors and their interactions. There was increased muscle lightness ( $L^*$ ) of the *Longissimus dorsi* muscles from the electrical stimulated carcasses (Table 2). This suggested that early postmortem conditions of these muscles favored protein denaturation (Warriss and Brown, 1987). High muscle temperatures combined with low muscle pH values during early postmortem are associated with increased protein denaturation. However, Bond *et al.* (2004) found that electrical stimulation of sheep carcasses did not improve muscle lightness.

Transportation of sheep for 3 hrs had a significant ( $P<0.05$ ) effect on myofibrillar fragmentation index of the *Longissimus dorsi* muscles (Table 2). The myofibrillar fragmentation index from non-transported muscle was significantly ( $P<0.05$ ) higher than those from transported one by 8.9%. Significantly lower myofibrillar fragmentation index and shorter sarcomere lengths for the transported-non-stimulated sheep are consistent with the tougher meat from that group. The high myofibrillar fragmentation index in stimulated-non-transported sheep may have been caused by myofibrils easily broken into shorter segments.

Myofibril fragmentation index has been used as an indicator of postmortem proteolysis in various types of meat (Lametsch *et al.*, 2007) as it accounts for differences in the rate of postmortem tenderization of meat (Nagaraj *et al.*, 2005). The latter authors concluded that structural changes occurring in meat after slaughter are generally

believed to be caused by interactions of myofibrillar proteins in the muscle. Low voltage electrical stimulation and transportation had a significant ( $P<0.05$ ) effect on myofibrillar fragmentation index of the sheep muscle (Table 2). The myofibrillar fragmentation index from transported muscle was significantly ( $P<0.05$ ) lower than that from non-transported one (Table 2).

## Conclusions

Transportation of sheep for 3 hrs in an open truck at high ambient temperatures (36 °C) led to higher blood concentrations of cortisol, adrenaline, nor-adrenaline and dopamine. Meat quality characteristics were also worsened by transportation during hot ambient temperatures. Low voltage electrical stimulation can be used to reduce the deleterious effects of transportation on meat quality.

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