



## Estimation of Pork Quality Traits Using Exsanguination Blood and Postmortem Muscle Metabolites

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**ABSTRACT:** The current study was designed to estimate the pork quality traits using metabolites from exsanguination blood and postmortem muscle simultaneously under the Korean standard pre- and post-slaughter conditions. A total of 111 Yorkshire (pure breed and castrated male) pigs were evaluated under the Korean standard conditions. Measurements were taken of the levels of blood glucose and lactate at exsanguination, and muscle glycogen and lactate content at 45 min and 24 h postmortem. Certain pork quality traits were also evaluated. Correlation analysis and multiple regression analysis including stepwise regression were performed. Exsanguination blood glucose and lactate levels were positively correlated with each other, negatively related to postmortem muscle glycogen content and positively associated with postmortem muscle lactate content. A rapid and extended postmortem glycolysis was associated with high levels of blood glucose and lactate, with high muscle lactate content, and with low muscle glycogen content during postmortem. In addition, these were also correlated with paler meat color and reduced water holding capacity. The results of multiple regression analyses also showed that metabolites in exsanguination blood and postmortem muscle explained variations in pork quality traits. Especially, levels of blood glucose and lactate and content of muscle glycogen at early postmortem were significantly associated with an elevated early glycolytic rate. Furthermore, muscle lactate content at 24 h postmortem alone accounted for a considerable portion of the variation in pork quality traits. Based on these results, the current study confirmed that the main factor influencing pork quality traits is the ultimate lactate content in muscle via postmortem glycolysis, and that levels of blood glucose and lactate at exsanguination and contents of muscle glycogen and lactate at postmortem can explain a large portion of the variation in pork quality even under the standard slaughter conditions. (**Key Words:** Exsanguination, Glucose, Glycogen, Lactate, Postmortem Glycolysis, Pork Quality)

### INTRODUCTION

Technological meat quality includes meat color, water

holding capacity (WHC), and texture properties (Warner et al., 2010; Lee et al., 2012). These properties are developed by the rate and extent of postmortem anaerobic glycolysis. A paler lean meat color, decreased WHC, and soft texture, such as found in pale, soft, and exudative (PSE) meat, is the deteriorative condition of pork. An accelerated rate of postmortem glycolysis is considered to be a primary cause of PSE meat (Bowker et al., 2000; Scheffler and Gerrard, 2007). During postmortem, muscles produce adenosine triphosphate via anaerobic glycolysis (i.e. catabolize muscle glycogen into lactate). This lactate accumulates in the muscle, resulting in a muscle pH decline which causes the denaturation of muscle proteins. A rapid decline in muscle pH at early postmortem while muscle temperature is still high causes severe denaturation of the myofibrillar and sarcoplasmic proteins (Ryu and Kim, 2006; Scheffler and

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Gerrard, 2007). So, measurement of postmortem muscle glycogen and lactate content could help to explain variations in postmortem glycolysis and subsequently pork quality (Kocwin-Podsiadla et al., 2006). Ryu et al. (2005) and Choe et al. (2008) also showed that higher contents of muscle glycogen and lactate at early postmortem was related to paler color and higher drip loss of pork.

There are numerous factors influencing postmortem metabolism, one of which is pre-slaughter stress. In general, stress immediately before slaughter accelerates postmortem glycolysis, resulting in PSE meat in pigs. Hormones and metabolite levels in blood are frequently used to measure livestock stress level. Blood cortisol and lactate levels, particularly, have been used as reliable indicators in many studies (Warriss, 1990; Hambrecht et al., 2004; 2005a, b; Edwards et al., 2010a, b; Choe and Kim, 2014). In addition, Choe and Kim (2014) and Choe et al. (2009) showed that the blood glucose level at exsanguination could be an indicator of stress and that it is strongly associated with pork quality traits. Furthermore, previous studies (Edwards et al., 2010a, b; Choe and Kim, 2014) showed that there are considerable variation in blood glucose and lactate levels although the experimental pigs were under the same standard slaughter conditions. However, most studies about pre-slaughter stress and metabolism imposed artificial stress on livestock. Moreover, there is limited research about the relationship between metabolites from blood and muscle and meat quality simultaneously. There are also few reports investigating how metabolites influence ultimate pork quality traits. Therefore, the objective was to estimate the pork quality traits using metabolites from exsanguination blood and postmortem muscle simultaneously under the Korean standard pre- and post-slaughter conditions.

## MATERIALS AND METHODS

### Animals, pre-slaughter handling, and slaughter procedure

All experimental conditions were in accordance with the Korean Animal Protection Act, including detailed provision for transport and slaughter. Treatments, handling conditions, and experimental procedures were controlled and/or approved by the Ministry of Agriculture, Food, and Rural Affairs of South Korea. Slaughter procedure was under the supervision of the Korea Institute for Animal Products Quality Evaluation.

A total of 111 Yorkshire (pure breed castrated male) pigs were evaluated. All pigs were non-carriers of the halothane gene and clinically healthy. The treatment conditions, both before and after slaughter, were same for all pigs. The pigs were divided randomly into different pens in a commercial farm, with a stocking density of 1.0 m<sup>2</sup> per pig. The temperature was maintained at 20±2°C, and artificial light was provided from 0900 to 2100. Water and food were

available *ad libitum* at nipple drinkers and food dispensers. In addition, all pigs were fed the same commercial diet in accordance with the National Research Council (1998). When the pigs reached a live weight of 115±5 kg, they were transported to a commercial slaughter facility. Approximately 4 h before transport, feed was withdrawn. The loading and unloading took place as parallel to the ground as possible. Electric prods were not used during loading or unloading. The truck had two straight decks, and a hydraulic lift raised the floor of the deck to create the second deck. The truck was equipped with natural ventilation, and the stocking density was approximately 0.47 m<sup>2</sup> per pig. The pigs were transported for 1 h to the slaughter facility. The pigs were not mixed with unfamiliar pigs during transport or lairage. To reduce stress during lairage, the pigs were showered with water and provided with drinking water *ad libitum*. After being rested for approximately 12 h at the slaughter facility, the pigs were slaughtered. All the pigs were slaughtered during the winter period in one of 4 batches (30, 30, 30, and 21 pigs per batch). The pigs were immobilized through electrical stunning (2 to 4 s with over 1.25 A), shackled by the right leg, and exsanguinated while hanging. At exsanguination, blood samples were collected from each pig. A dehairing process was performed in a scalding-singeing combination by scalding for 3 min at 60°C, and then singeing via passing through gas burner for 10 s at 1,200°C, and any remaining hair was removed using a knife and flame. The carcasses were eviscerated, split into the left and right sides, and then cooled in the chilling room (4°C).

At 45 min postmortem, muscle samples for the analysis of glycogen and lactate contents were taken from the *longissimus lumborum* muscle at the 9th and 10th thoracic vertebrae. After 24 h of chilling, muscle samples for the measurement of muscle glycogen and lactate contents and pork quality traits were taken from the *longissimus lumborum* muscle at the 10th and 15th thoracic vertebrae. The collection of all muscle samples and pH measurements was made on the right side of each carcass.

### Blood samples and blood metabolites measurements

Blood samples were collected from each pig at exsanguination after electrical stunning. The blood samples were collected using tubes treated with potassium oxalate/sodium fluoride (BD Vacutainer fluoride tube; Becton Dickinson, Franklin Lakes, New Jersey, USA) to inhibit further glycolysis. Blood glucose and lactate levels were then measured using hand-held devices (blood glucose: OneTouch Ultra, LifeScan, Inc., Milpitas, California, USA; blood lactate: Lactate Scout, EKF Diagnostics, Barleben, Magdeburg, Germany). All measurements were completed within 10 min of exsanguination at the slaughter facility. Blood glucose level

was expressed in milligrams per deciliter (mg/dL), and blood lactate level was expressed in millimoles per liter (mmol/L).

#### Measurement of muscle glycogen and lactate contents

Glycogen content of muscle was measured via the method described by Dreiling et al. (1987). Approximately 1.5 g of tissue was minced, suspended in 10 mL of 9% cold perchloric acid (PCA), and thoroughly homogenized for 30 to 45 s with a mechanical tissue disrupter. After centrifugation ( $15,000\times g$  at  $4^{\circ}\text{C}$ ), the supernatant was decanted and saved for glycogen determination. An iodine color reagent was prepared by combining 100 mL of saturated  $\text{CaCl}_2$  with 1.3 mL of the solution (0.26 g of iodine, 2.6 g of potassium iodide in 10 mL of distilled water) (prepared freshly every day). The 2.6 mL of the iodine color reagent was added to 0.4 mL of the glycogen standards or tissue extracts. In this way, glycogen standard curves were developed for each set of samples so that linear regression equations could be used to determine the glycogen concentrations of the test samples.

Lactate content was determined spectrophotometrically at 340 nm using a commercial kit (Boehringer-Mannheim, Darmstadt, Germany). Approximately 500 mg of muscle was homogenized for 30 s in 2 mL of 1 M PCA. Potassium hydroxide (2 M) was added to neutralize the solution, and distilled water was added to make a final volume of 10 mL. After 20 min of refrigeration and centrifugation, lactic acid concentration was measured.

#### Pork quality evaluation

Muscle pH at 45 min postmortem ( $\text{pH}_{45 \text{ min}}$ ) and 24 h postmortem ( $\text{pH}_{24 \text{ h}}$ ) was measured in the chilling room directly at the 7th and 8th thoracic vertebrae of each carcass (right side) using a portable pH meter for meat (HI 99163, Hanna instruments, Woonsocket, Rhode Island, USA).

For analysis of meat color, WHC, and Warner-Bratzler shear force (WBS), samples were obtained from the pork loin at the 10th to 15th thoracic vertebrae from the right side of each cold carcass. Meat color was measured with a Minolta Chroma Meter (CR-400, Minolta Camera Co., Tokyo, Japan), following a slightly modified version of the method of Honikel (1998). Samples were cut from the pork loin at 24 h postmortem and placed on a table for 30 min, exposing their surfaces to air without any packaging (for bloom) before color measurement. Triplicate measurements were averaged, and the results were expressed as Commission Internationale de l'Eclairage lightness ( $L^*$ ), redness ( $a^*$ ), and yellowness ( $b^*$ ).

To evaluate the WHC, drip loss, cooking loss (Honikel, 1998), and filter-paper fluid uptake (FFU) (Kauffman et al., 1986) were measured. To determine drip loss, meat samples

were cut from the pork loin at 24 h postmortem and weighed immediately to obtain their initial weight of drip loss. The samples were then placed in netting and suspended in an inflated bag, ensuring that the sample was not in contact with the bag. After a 48 h storage period at  $4^{\circ}\text{C}$ , the samples were taken from the bag, gently blotted dry, and reweighed. Drip loss was expressed as a percentage of the sample's initial weight. To measure cooking loss of water, different samples were freshly cut from the pork loin at 24 h postmortem and weighed to obtain their initial weight. The samples were then put in thin-walled polyethylene bags and placed in a continuously boiling water bath. The samples were cooked to  $75^{\circ}\text{C}$  internal temperature and, once this end-point temperature had been reached, the bags were removed from the water bath. Thereafter, the samples were cooled in an ice slurry and kept under chilled conditions ( $1^{\circ}\text{C}$  to  $5^{\circ}\text{C}$ ) until equilibration. Then, the samples were taken from the bag, blotted dry, and reweighed. Cooking loss was expressed as a percentage of the sample's initial weight. For FFU, further samples were cut from the pork loin at 24 h postmortem. Filter paper (Whatman #2, 42.5 mm in diameter, GE Healthcare, Little Chalfont, Buckinghamshire, UK) was pre-weighed, placed on the surface of the sample for just under 2 s to absorb fluids, and weighed. The FFU was expressed as milligrams of exudate absorbed into the filter paper.

To measure WBS, the pork loin was cut into 20-mm-thick chops. Pork chops from each sample were cooked to a final core temperature of  $75^{\circ}\text{C}$  in a continuously boiling water bath. After cooking, six cores (1.27 cm in diameter), parallel to the longitudinal orientation of the muscle fibers, were taken from each pork chop for WBS measurement. WBS was determined using an Instron Universal Testing Machine (Model Series IX, Instron Co., Norwood, MA, USA) with a Warner-Bratzler shearing device. The samples were sheared perpendicular to the long axis of the core, and the WBS value was calculated taken from the peak of the force curve (Honikel, 1998).

#### Statistical analysis

All statistical analysis was performed using the Statistical Analysis System (2009). Pearson correlation coefficients between blood and muscle metabolites or between blood and muscle metabolites and pork quality traits were determined using the CORR procedure. To establish multiple regression models for pork quality traits, blood or muscle metabolites were used as independent variables in the REG procedure. Furthermore, the stepwise procedure was used to examine the percentage of variation in certain pork quality traits explained by blood and muscle metabolites. The qualitative variable (batch) was coded as

**Table 1.** Correlation coefficients between blood glucose and lactate levels and muscle glycogen and lactate contents

	Blood lactate	Muscle glycogen <sub>45 min</sub>	Muscle lactate <sub>45 min</sub>	Muscle glycogen <sub>24 h</sub>	Muscle lactate <sub>24 h</sub> <sup>3</sup>
Blood glucose <sup>1</sup>	0.46***	-0.25**	0.51***	-0.35***	0.47***
Blood lactate <sup>1</sup>		-0.28**	0.39***	-0.10	0.24*
Muscle glycogen <sub>45 min</sub> <sup>2</sup>			-0.47***	0.05	-0.28**
Muscle lactate <sub>45 min</sub> <sup>2</sup>				-0.27**	0.44***
Muscle glycogen <sub>24 h</sub> <sup>3</sup>					-0.25**

<sup>1</sup> Blood glucose (mg/dL) and lactate (mmol/L) level measured at exsanguination.

<sup>2</sup> Muscle glycogen and lactate content (mg/g) measured at 45 min postmortem.

<sup>3</sup> Muscle glycogen and lactate content (mg/g) measured at 24 h postmortem.

\* p<0.05; \*\* p<0.01; \*\*\* p<0.001.

dummy variables to eliminate their effect on the regression model.

## RESULTS

### Correlation between levels of blood glucose and lactate and contents of muscle glycogen and lactate

Table 1 shows the correlation coefficients between the following variables: exsanguination blood glucose and lactate levels and postmortem muscle glycogen and lactate contents. Levels of blood glucose and lactate showed a positive relationship. Blood glucose level was negatively related to muscle glycogen content at postmortem (i.e. 45 min and 24 h postmortem) but positively correlated with muscle lactate content at postmortem. Blood lactate level was also negatively related to muscle glycogen content at 45 min postmortem and positively correlated with muscle lactate content at postmortem. These relationships mean that high levels of blood glucose and lactate are associated with not only rapid glycolytic rate at early postmortem (glycogen and lactate contents measured at 45 min postmortem) but also extended postmortem glycolysis (glycogen and lactate contents measured at 24 h postmortem). There are negative correlations between postmortem muscle glycogen and lactate contents.

### Correlation between blood and muscle metabolites and pork quality traits

There are significant relationships between metabolites

in exsanguination blood and postmortem muscle and pH, lightness, and WHC (Table 2). Blood glucose level is negatively related to muscle pH<sub>45 min</sub> and pH<sub>24 h</sub> and positively correlated to lightness, FFU, and drip loss. Blood lactate level is also negatively related to postmortem muscle pH and positively correlated with FFU and drip loss. Higher levels of exsanguination blood metabolites were associated with paler color and decreased WHC.

Muscle glycogen content at 45 min postmortem had a positive relationship with muscle pH<sub>45 min</sub> and negative relationship with lightness, FFU, and drip loss. Muscle glycogen content at 24 h postmortem exhibited similar relationships, except that it was significantly correlated with muscle pH<sub>24 h</sub>, rather than muscle pH<sub>45 min</sub>. On the other hand, muscle lactate content at both 45 min and 24 h postmortem were negatively related to postmortem muscle pH and positively correlated with lightness, FFU, and drip loss. Lower glycogen content and higher lactate content in postmortem muscle were correlated with deteriorative pork quality.

### Influence of blood and muscle metabolites with pork quality traits

Multiple regression analysis including stepwise regression was performed to investigate the influence of exsanguination blood metabolites (Table 3), postmortem muscle metabolites (Table 4), or both the blood and muscle metabolites together (Table 5) on pork quality traits. Levels of blood glucose and lactate at exsanguination explained

**Table 2.** Correlation coefficients between blood and muscle metabolites and certain pork quality traits

	pH <sub>45 min</sub>	pH <sub>24 h</sub>	Lightness	FFU	Drip loss	Cooking loss	WBS
Blood glucose <sup>1</sup>	-0.41***	-0.51***	0.25**	0.35***	0.32***	0.01	-0.16
Blood lactate <sup>1</sup>	-0.40***	-0.20*	0.15	0.41***	0.35***	0.10	-0.26**
Muscle glycogen <sub>45 min</sub> <sup>2</sup>	0.67***	-0.01	-0.23*	-0.20*	-0.34***	-0.20*	0.14
Muscle lactate <sub>45 min</sub> <sup>2</sup>	-0.53***	-0.30**	0.20*	0.33***	0.32***	0.02	-0.18
Muscle glycogen <sub>24 h</sub> <sup>3</sup>	0.16	0.40***	-0.28**	-0.25**	-0.22*	-0.04	0.04
Muscle lactate <sub>24 h</sub> <sup>3</sup>	-0.30**	-0.55***	0.53***	0.51***	0.61***	0.14	-0.09

FFU, filter-paper fluid uptake; WBS, Warner-Bratzler shear force.

<sup>1</sup> Blood glucose (mg/dL) and lactate (mmol/L) level measured at exsanguination.

<sup>2</sup> Muscle glycogen and lactate content (mg/g) measured at 45 min postmortem.

<sup>3</sup> Muscle glycogen and lactate content (mg/g) measured at 24 h postmortem.

\* p<0.05; \*\* p<0.01; \*\*\* p<0.001.

**Table 3.** Multiple regression models for certain pork quality traits using blood glucose and lactate levels measured at exsanguination

Pork quality traits	Intercept	Blood metabolites level		$R^2$	Significance
		Glucose <sup>1</sup>	Lactate <sup>1</sup>		
pH <sub>45 min</sub>	6.534	-0.001	-0.014	0.218	***
pH <sub>24 h</sub>	5.794	-0.001	-0.001	0.240	***
Lightness	43.607	0.015	0.022	0.086	*
FFU	4.062	0.061	1.086	0.210	***
Drip loss	1.196	0.007	0.105	0.162	***
WBS	64.089	-0.002	-0.680	0.074	*

FFU, filter-paper fluid uptake; WBS, Warner-Bratzler shear force.

<sup>1</sup> Blood glucose (mg/dL) and lactate (mmol/L) level measured at exsanguination.

\*  $p < 0.05$ ; \*\*\*  $p < 0.001$ .

21.8% and 24% of the variation in muscle pH<sub>45 min</sub> and pH<sub>24h</sub>, respectively. They also accounted for 21% and 16.2% of the variation in FFU and drip loss, respectively. On the other hand, compared to blood metabolite levels, contents of muscle glycogen and lactate at postmortem explained a higher proportion of the variation in pork quality traits. Over 40% of the variability in postmortem muscle pH was explained by muscle metabolite content. Metabolites contents in postmortem muscle accounted for over 40% of the variation in lightness and drip loss. The variability of FFU was also explained up to 31.2% by

muscle metabolite content.

When a stepwise regression analysis using metabolites from both exsanguination blood and postmortem muscle was carried out, the variability of pork quality traits was explained slightly further. Blood metabolites and muscle metabolites at 45 min postmortem accounted for 54.4% of variation in muscle pH<sub>45 min</sub>. In the case of muscle pH<sub>24 h</sub>, 46.2% of the variation was explained by blood and muscle metabolites. In fact, muscle lactate content at 24 h postmortem alone accounted for 30.1% of the variation. Variability of lightness, FFU, and drip loss showed similar

**Table 4.** Multiple regression models for certain pork quality traits using muscle glycogen and lactate contents measured at 45 min and 24 h postmortem

Pork quality traits	Intercept	Muscle metabolites content				$R^2$	Significance
		Glycogen <sub>45 min</sub> <sup>1</sup>	Lactate <sub>45 min</sub> <sup>1</sup>	Glycogen <sub>24 h</sub> <sup>2</sup>	Lactate <sub>24 h</sub> <sup>2</sup>		
pH <sub>45 min</sub>	6.030	0.557	0.261	-0.048	-0.026	0.522	***
pH <sub>24 h</sub>	6.503	-0.130	0.627	-0.018	-0.106	0.414	***
Lightness	32.495	-1.936	-9.814	-0.247	2.244	0.406	***
FFU	-47.565	-4.315	-35.495	1.063	10.267	0.312	***
Drip loss	-6.489	-1.348	-3.011	0.012	1.455	0.435	***
Cooking loss	25.764	-2.818	0.161	-0.025	0.800	0.101	*

FFU, filter-paper fluid uptake.

<sup>1</sup> Muscle glycogen and lactate content (mg/g) measured at 45 min postmortem.

<sup>2</sup> Muscle glycogen and lactate content (mg/g) measured at 24 h postmortem.

\*  $p < 0.05$ ; \*\*\*  $p < 0.001$ .

**Table 5.** Proportion of variability in certain pork quality traits explained by blood and muscle metabolites using stepwise regression

	pH <sub>45 min</sub>	pH <sub>24 h</sub>	Lightness	FFU	Drip loss	Cooking loss	WBS
Blood metabolites measured at exsanguination							
Glucose	7.9 <sup>1</sup> ***	9.0***					
Lactate	1.7**			9.2***	4.8**		7.3**
Muscle metabolites measured at 45 min postmortem							
Glycogen	44.8***	3.9*	1.7		2.2*	7.3**	
Lactate							
Muscle metabolites measured at 24 h postmortem							
Glycogen		3.2*	2.5*	1.6			
Lactate		30.1***	35.5***	27.7***	39.1***	2.8	
Cumulative contribution	54.4	46.2	39.7	38.5	46.2	10.1	7.3

FFU, filter-paper fluid uptake; WBS, Warner-Bratzler shear force.

<sup>1</sup> Percentage of partial  $R^2$

\*  $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ .

tendencies, with muscle lactate content at 24 h postmortem alone explaining a considerable portion of the variation once again.

## DISCUSSION

One of the interesting subjects in meat science is pre-slaughter stress because pre-slaughter stress causes variations in postmortem muscle metabolism and subsequently influences meat quality (Warriss, 1990; D'Souza et al., 1998a, b; Faucitano, 1998; Hambrecht et al., 2004; 2005a, b; Edwards et al., 2010a, b; Koknaroglu and Akunal, 2013; Choe and Kim, 2014). Specifically, stress just before slaughter accelerates the glycolytic rate, resulting in rapid decline in muscle pH and consequently deteriorative meat quality (D'Souza et al., 1998a, b; van der Wal et al., 1999; Hambrecht et al., 2004; 2005a; Edwards et al., 2010a). Blood glucose and lactate levels at exsanguination are usually used to measure livestock stress levels. Blood lactate level is one of the most reliable indicators of stress level and is partially associated with meat quality (Warriss et al., 1994; Hambrecht et al., 2004; 2005a, b; Edwards et al., 2010a, b; Warriss, 2010; Choe and Kim, 2014). On the other hand, blood glucose level is an indirect indicator of stress because there are numerous factors which influence blood glucose level (Shaw and Tume, 1992; Mota-Rojas et al., 2012; Choe and Kim, 2014). Nevertheless, Choe and Kim (2014) and Choe et al. (2009) showed that blood glucose and lactate levels at exsanguination were significantly related to pork quality traits.

In the current study, exsanguination blood glucose and lactate levels were positively related with each other. Similarly, Choe and Kim (2014) showed that blood glucose levels are positively correlated with blood lactate and cortisol levels. A physiological response to stress is the activation of two main neuroendocrine systems, the hypothalamic-pituitary-adrenocortical (HPA) axis and the sympathetic nervous system. Stimulating the HPA axis releases cortisol, which elevates blood glucose level via gluconeogenesis (Warriss, 2010). Stimulating the sympatho-adrenal system leads to an increase in blood glucose level due to a rapid breakdown of liver glycogen. In addition, livestock consume energy via anaerobic glycolysis under stressful conditions, resulting in increased blood lactate level (Warriss, 2010; Foury et al., 2011; Choe and Kim, 2014).

Kocwin-Podsiadla et al. (2006) have reported that the change of metabolite levels in postmortem muscle indicates normal and deteriorative quality meat, such as PSE and acid meat. Especially, glycogen and lactate contents in the muscle were the significant indicator of muscle pH change and ultimate quality conditions. Choe et al. (2008) also showed that different levels of glycogen and lactate content

in postmortem muscle can explain the rate and extent of postmortem glycolysis. In that study, muscle lactate content at early postmortem was associated with the early postmortem glycolytic rate, while muscle glycogen content at early postmortem was related to ultimate muscle pH. Similarly, the current study showed that blood glucose and lactate levels were negatively related to postmortem muscle glycogen content and positively correlated with muscle lactate content. Exsanguination blood metabolite levels were also associated with a rapid and extended postmortem glycolysis. Furthermore, muscle lactate content at early postmortem was negatively correlated with muscle pH<sub>45 min</sub>. However, there was no significant relationship between the muscle glycogen content measured at 45 min postmortem and muscle pH<sub>45 min</sub>. Similar results were observed in the study of Edwards et al. (2010a), showing that exsanguination blood lactate level was negatively related to muscle pH measured at 60 min postmortem. D'Souza et al. (1998a) reported that muscles with lower glycogen and higher lactate content caused by handling stress showed lower muscle pH postmortem. Hambrecht et al. (2004; 2005a) also observed similar results.

The rate and extent of postmortem pH decline have a strong impact on the development of pork quality (D'Souza et al., 1998a, b; Kocwin-Podsiadla et al., 2006; Ryu and Kim, 2006; Scheffler and Gerrard, 2007). A rapid and extended postmortem glycolysis causes severe denaturation of myofibrillar and sarcoplasmic proteins (Joo et al., 1999). It is obvious that severely denatured muscle proteins have a negative effect on pork quality, resulting in both a lighter color and reduced WHC. D'Souza et al. (1998a) showed that low glycogen and high lactate content during postmortem due to poor handling just prior to slaughter caused a rapid glycolytic rate, high amounts of exudate, and a high incidence of PSE meat. Hambrecht et al. (2005a, b) similarly reported that low glycogen and high lactate at postmortem decrease WHC. Ryu et al. (2005) showed that high muscle lactate content at 24 h postmortem was closely correlated with decreased muscle pH, reduced protein solubility, paler color, and high fluid loss. Kocwin-Podsiadla et al. (2006) have reported that the change in postmortem muscle pH can be used to diagnose normal and deteriorative meat. In addition, Choe et al. (2009) and Choe and Kim (2014) showed that high levels of blood glucose and lactate levels were also significantly correlated with deteriorative pork quality. As expected, the results of the current study were consistent with those of previous ones. Paler colors and high amounts of fluid loss were significantly associated with high levels of blood glucose and lactate at exsanguination and low contents of glycogen and high lactate in postmortem muscle. Particularly, correlation coefficients between muscle lactate content at 24 h postmortem and muscle pH<sub>24 h</sub>, lightness, FFU, and drip loss, respectively, had higher values than others. This

confirmed again that pork quality traits are significantly affected by the accumulation of lactate from postmortem glycolysis.

Hambrecht et al. (2004) reported that blood lactate level and glycolytic potential explained a considerable portion of the variation in lightness and drip loss. Blood lactate level alone explained about 30% of the variation in lightness and drip loss but blood lactate level and glycolytic potential together could explain about 50% (Hambrecht et al., 2004). In the current study, exsanguination blood glucose and lactate levels explained approximately 20% of the variation in muscle pH and FFU, and about 15% of the variation in drip loss. On the other hand, muscle glycogen and lactate content measured at 45 min and 24 h postmortem accounted for 31% to 52% of variation in muscle pH, lightness, FFU, and drip loss. The study of Hambrecht et al. (2004) imposed pre-slaughter stress on experimental pigs, but in the current study, all experimental pigs were under the Korean standard pre-slaughter condition. This difference may explain the lower  $R^2$  values of the result of multiple regression analysis in the present study compared to those of Hambrecht et al. (2004).

Stepwise regression analysis is a type of multiple regression analysis for selecting independent variables that have an impact on the dependent ones or can explain a higher proportion of their variation (Lee et al., 2012). The current study showed that levels of blood glucose and lactate at exsanguination and muscle glycogen content at 45 min postmortem explained 54.4% of the variation in muscle  $\text{pH}_{45 \text{ min}}$ . On the other hand, muscle lactate content at 24 h postmortem accounted for a considerable portion of the variability of muscle  $\text{pH}_{24 \text{ h}}$ , lightness, FFU, and drip loss. Other metabolite concentrations in blood and muscle accounted for a small portion of variation in pork quality traits.

## CONCLUSION

The current study showed that high levels of exsanguination blood glucose and lactate were associated low content of glycogen and high content of lactate in postmortem muscle. In addition, these metabolite levels were associated with accelerated and extended postmortem anaerobic glycolysis, resulting in paler color and reduced WHC. Stepwise regression also showed metabolite levels from exsanguination blood and postmortem muscle accounted for up to 54.5% of variation in pork quality. Especially, ultimate lactate content in postmortem muscle explained a considerable variation (27.7% to 39.1%) in pork quality traits. These results indicated that a major factor which effects pork quality is the ultimate lactate content in muscle through postmortem glycolysis and that exsanguination blood glucose and lactate levels and postmortem muscle glycogen and lactate contents can

explain a large portion of the variation in pork quality even under the standard slaughter conditions.

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