EFFECTS OF GLYCOGEN AND LACTATE CONTENT ON PROTEIN DENATURATION AND THEIR INFLUENCE ON PORK QUALITY

J. H. Choe[1], Y. M. Choi[1], S. H. Lee[1], Y. K. Kang[1], K. C. Hong[2], and B. C. Kim[1*]

[1] Division of Food Science & Technology, Korea University, 5-1 Anam-Dong, Seongbuk-Gu, Seoul 136-701, South Korea

[2] Division of Biotechnology, Korea University, 5-1 Anam-Dong, Seongbuk-Gu, Seoul 136-701, South Korea

[*]Corresponding author (phone: +82-2-3290-3052; fax: +82-2-925-1970; e-mail: bckim@korea.ac.kr)

***Abstract*—The purpose of this study was to investigate the influence of glycogen and lactate content on total protein solubility during the postmortem period and to examine the effect of total protein solubility on pork quality. Two groups were categorized by total protein solubility. Although there were no significant differences in postmortem glycogen contents between the groups, the low total protein solubility group contained significantly more lactate at 45 min postmortem than the high total protein solubility group. The low group exhibited a rapid metabolic rate during the early postmortem period compared to the high group. In addition, the low group was paler in color, and had a lower water holding capacity than the high group. Based on these results, the lactate content at 45 min postmortem had an impact on the extent of protein denaturation, which affected pork quality.**

***Index Terms*—Gylcogen, Lactate, Pork quality, Total protein solubility**

I. INTRODUCTION

Muscles attempt to maintain homeostasis by preserving cellular adenosine triphosphate (ATP) concentrations. Muscles produce ATP using glycogen and glucose by means of oxidative and glycolytic pathways, and lactate formed by glycolysis is converted back to pyruvate to be used oxidatively via the tricarboxylate acid cycle, or is transported out of the muscle fiber if there is a lack of oxygen and/or mitochondria (Poso & Puolanne, 2005). However, because postmortem muscles lack the oxygen required for oxidative metabolism due to exsanguination, they produce ATP by glycolysis. Moreover, the lactate generated through glycolysis accumulated in postmortem muscle because of the failure of blood circulation (Bowker, Grant, Forrest, & Gerrard, 2000; Scheffler & Gerrard, 2007). The accumulation of lactate induces a decline in muscle pH, and can affect muscle protein characteristics. Rapid postmortem glycolysis can induce muscle to have a low pH at relatively high temperatures or high extent glycolysis can induce muscle having abnormally low ultimate pH. Both conditions can cause severe denaturation of muscle protein, which may contribute to decreased pork quality (Joo, Kauffman, Kim, & Park, 1999; Ryu, Choi, & Kim, 2005; Scheffler & Gerrard, 2007). Thus, determining glycogen and lactate contents will lead to a better understanding of denaturation of muscle protein and pork quality.

Pork quality can be defined as the combination of diverse properties of fresh meat. One aspect of these properties is technological quality, such as meat color, water holding capacity, and texture; all of which are affected by postmortem metabolism during the coversion of muscle to meat (van der Wal, Engel, & Hulsegge, 1997, Ryu et al., 2005). The main metabolic pathway in the conversion of muscle to meat is glycolysis and continuous postmortem glycolysis lower muscle pH (Bowker et al., 2000). The high rate of pH decline or abnormally low ultimate pH can cause severe denaturation of muscle proteins (Scheffler & Gerrard, 2007). Moreover, denaturation of sarcoplasmic and myofibrillar proteins is closley related to pork quality parameters including color and water holding capacity (Joo et al., 1999) and the solubility of sarcoplasmic protein can be used as an indicator for meat quality (Sayre & Brisky, 1963). Thus, muscle protein characteristics are a critical factor affecting pork quality development (Scheffler & Gerrard, 2007).

Therefore, the purpose of this study was to investigate the influence of glycogen and lactate content on total protein solubility during the postmortem period and to examine the effect of total protein solubility on pork quality traits.

II. MATERIALS AND METHODS

1. Animals and muscle samples

A total of 51 crossbred (Landrace × Yorkshire × Duroc) pigs were evaluated. The reaing conditions for animals were the same before slaughter. The pigs were transported to a commercial abattoir under the same conditions and handling, and were slaughtered at a similar live weight (110 ± 5 kg). The slaughter plant used electrical stunning and a traditional

scalding-singeing process. At 45 min postmortem, muscle samples were taken for glycogen and lactate analysis from the *longissimus dorsi* muscles at the 9th thoracic vertebra. After 24 h of chilling, samples from the pork loins (the 10th–13th thoracic vertebra) were taken to measure glycogen, lactate, meat quality traits and protein solubility.

2. Protein solubility

Samples were taken from carcasses at 24 h postmortem in a 4 °C cold room, promptly frozen by liquid nitrogen and stored at –80 °C until analyzed for protein solubility. To determine protein solubility, muscle samples were made into muscle powder using a Waring blender (7010/51BL30, Waring Commercial, USA) adding liquid nitrogen. Sarcoplasmic protein was extracted from 1 g of the muscle powder using 10 ml of ice-cold 0.025 M potassium phosphate buffer (pH 7.2) (Joo et al., 1999), and total (sarcoplasmic + myofibrillar) protein was extracted from 1 g of the muscle powder using 20 ml of ice-cold 1.1 M potassium iodide in a 0.1 M phosphate buffer (pH 7.2) (Joo et al., 1999). The samples were homogenized on ice with a polytron at the lowest setting, and then left on a shaker at 4 °C overnight. Next, the samples were centrifuged at 1500 × g for 20 min, and the protein concentrations of the supernatants were determined via the Biuret method (Gornall, Bardawill, & David, 1949). Myofibrillar protein solubility was determined from the difference between the total and sarcoplasmic protein solubilities (Joo et al., 1999).

3. Glycogen and lactate content

The muscle glycogen content was measured via the method described by Dreiling, Brown, Casale, and Kelly (1987). Glycogen standard curves were developed for each set of samples. Linear regression equations were used to determine the glycogen concentrations in the corresponding samples. Glycogen change values were calculated by the difference between glycogen content at 45 min and 24 h postmortem.

Lactate content was determined spectrophotometrically (340 nm) using a commercial kit (Boeringer-Mannheim, Germany). Lactate change values were obtained by the difference between lactate content at 24 h and 45 min postmortem.

4. Meat quality traits

In a cold room, muscle pH at 45 min postmortem ($pH_{45\ min}$) and 24 h postmortem ($pH_{24\ h}$) were measured directly on the carcasses at the 7th/8th thoracic vertebra using a portable pH meter (HM-17MX, TOADKK, Japan). To determine ATP depletion during the early postmortem period, the *R*-value (hypoxanthine/adenosine ratio) was determined by the procedure of Calkins, Dutson, Smith, and Carpenter (1982).

Meat color was measured with a Minolta chromameter (CR-300, Minolta Camera Co., Japan). Samples were cut from carcasses at 24 h postmortem in a 4 °C cold room and placed on the table for 30 min without any packaging (for bloom) prior to measuring meat color. The average of triplicate measurements was recorded and the results were expressed as Commission Internationale de l'Eclairage (C.I.E, 1978) lightness.

In order to evaluate water holding capacity (WHC), measurements for drip loss (Honikel, 1998) and filter-paper fluid uptake (FFU) (Kauffman, Eikelenboom, van der Wal, Merkus, & Zaar, 1986) were performed. Drip loss was expressed as percentages of the initial sample weight (Honikel, 1998). FFU was expressed as milligrams of exudate absorbed into the filter-paper (Kauffman et al. 1986).

5. Statistical analysis

Total protein solubility was classified using cluster analysis with the FASTCLUS program from the SAS Institute (2004). The data were classified into two clusters according to total protein solubility (high, n = 22; low, n = 29). A General Linear Model was used to evaluate the significant differences among the high and low total protein solubility groups. The results for the groups were presented as the least square means with the standard errors of the least square means.

III. RESULTS AND DISCUSSION

The differences of glycogen and lactate contents during the postmortem period between the groups are presented in Table 1. No significant differences were observed in glycogen contents during the postmortem period. On the other hand, the lactate contents at 45 min postmortem significantly different between the groups ($P < 0.05$), but not at 24 h postmortem. The low group contained more lactate at 45 min postmortem, which declined less compared to the high group.

Previous studies have shown that muscles with a higher lactate content at 45 min postmortem exhibited lower muscle protein solubilities (Choe et al., 2008), and lactate content at 45 min postmortem negatively correlated with myofibrillar and total protein solubility (Choe et al., 2007). This study also showed similar results compared to previous studies. Thus, lactate content at 45 min postmortem had an impact on muscle protein denaturation.

To examine the effect of total protein solubility on pork quality traits, two groups were categorized by total protein solubility (Table 2). There were significant differences in muscle pH at 45 min postmortem and the *R*-value between the groups ($P < 0.05$). The low total protein solubility group exhibited a lower $pH_{45\ min}$ and a higher *R*-value than the high

total protein solubility group. However, there was no significant difference in muscle pH at 24 h postmortem between the groups. The lightness ($P < 0.05$) is different between the groups. In addition, drip loss ($P < 0.1$) and FFU ($P < 0.001$) are different between the groups. The low group also exhibited lower sarcoplasmic and myofibrillar protein solubility ($P < 0.001$) than the high group.

Ryu et al. (2005) showed that the fast metabolic rate group exhibited lower $pH_{45\ min}$ and muscle protein solubility. Moreover, Joo et al. (1999) reported that sarcoplasmic, myofibrillar, and total protein solubility were negatively related to lightness, drip loss, and FFU. The results of this study are consistent with previous studies. Thus, total protein solubility is influenced by $pH_{45\ min}$, and affects pork quality traits.

Table 1. Glycogen and lactate contents of *longissimus dorsi* muscle in the groups categorized by total protein solubility

	Total protein solubility		**Level of significance**
	Low (n=29)	**High (n=22)**	
Glycogen content (mg/g)			
45 min postmortem	0.92 (0.13)[1]	1.13 (0.15)	NS
24 h postmortem	0.30 (0.02)	0.26 (0.02)	NS
Glycogen change value[2]	0.63 (0.12)	0.86 (0.14)	NS
Lactate content (mg/g)			
45 min postmortem	5.83[a] (0.30)	4.79[b] (0.35)	*
24 h postmortem	8.10 (0.18)	7.90 (0.21)	NS
Lactate change value[3]	2.27[b] (0.26)	3.11[a] (0.31)	*

Level of significance: NS, not significant; * $P < 0.05$.
[a-b] Least square means with different superscripts in the same row differ significantly ($P < 0.05$).
[1] Standard error of least-square means.
[2] Glycogen change value = glycogen measured at 45 min – glycogen measured at 24 h postmortem.
[3] Lactate change value = lactate measured at 24 h – lactate measured at 45 min postmortem.

Table 2. Pork quality traits of *longissimus dorsi* muscle in the groups categorized by total protein solubility

	Total protein solubility		**Level of significance**
	Low	**High**	
Muscle pH & R-value			
$pH_{45\ min}$	5.92[b] (0.04)[1]	6.09[a] (0.05)	*
$pH_{24\ h}$	5.57 (0.02)	5.56 (0.02)	NS
R-value	1.12[a] (0.03)	1.00[b] (0.04)	*
Pork quality traits			
Lightness	48.24[a] (0.40)	46.79[b] (0.46)	*
Drip loss (%)	5.47[a] (0.38)	4.43[b] (0.43)	†
FFU[2] (mg)	57.17[a] (3.85)	33.59[b] (4.43)	***
Protein solubility (mg/g)			
Sarcoplasmic protein	66.38[b] (0.93)	75.03[a] (1.06)	***
Myofibrillar protein	115.1[b] (1.55)	132.4[a] (1.78)	***
Total protein	181.5[b] (1.42)	207.5[a] (1.63)	***

Level of significance: NS, not significant; † $P < 0.1$; * $P < 0.05$; *** $P < 0.001$.
[a-b] Least square means with different superscripts in the same row differ significantly ($P < 0.05$).
[1] Standard error of least-square means.
[2] Filter-paper fluid uptake

IV. CONCLUSION

The lactate contents at 45 min postmortem have an impact on the extent of protein denaturation, and denatured proteins affect pork quality traits.

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