Correlations between myosin light chain isoforms and glycolytic characteristics in porcine *longissimus dorsi* muscle

Choi Y.M., Choe J.H., Jung K.C. and B.C. Kim

Division of Food Bioscience and Technology, College of Life Sciences and Biotechnology, Korea University, Seoul, South Korea
* Corresponding author (phone: +82-2-3290-3052; fax: +82-2-3290-4984; e-mail: bckim@korea.ac.kr)

Abstract— **The objective of this study was to investigate the content of myosin light chain (MLC) isoforms and their relationships with the glycolytic rate, metabolite contents, and activities of glycolytic enzymes in the porcine *longissimus dorsi* muscle. A total of 57 crossbred pigs were evaluated in this study. There were no correlations between the content of MLC isoforms and muscle pH (45 min and 24 h postmortem). However, MLC ls isoform content was positively correlated with glycogen content at 24 h postmortem (r=0.28, P<0.05), and negatively correlated with lactate content at 45 min postmortem (r=–0.35, P<0.01). The content of MLC 3f isoform had a positive correlation with the activity of pyruvate kinase (r=0.44, P<0.01), while the ratio of fast/slow isoform showed a positive correlation with lactate content at 45 min postmortem (r=0.28, P<0.05). Thus, the composition of MLC isoforms can influence postmortem rigor development in the porcine longissimus dorsi muscle, and consequently meat quality indirectly.**

Keywords— **Myosin light chain isoforms, glycolytic characteristics, pork.**

I. INTRODUCTION

Muscle fibers can be divided into various types depending on the myosin heavy chain (MHC) isoforms when they express [1]. Thus, the composition of MHC isoforms is major factor that influence energy metabolism in live animals, as well as during postmortem conversion of muscle to meat [2, 3]. Moreover, not only MHC, but also other protein isoforms, especially myosin light chain (MLC) isoforms, can influence the contraction speed and energetic properties in live animals [3]. However, the effects of the MLC isoforms on postmortem muscle changes are not yet fully understood. Therefore, the objective of this study was to investigate the content of MLC isoforms and their relationships with the glycolytic rate, metabolite contents, and activities of glycolytic enzymes in the porcine *longissimus dorsi* muscle.

II. MATERIALS AND METHODS

A. Animals and muscle samples

A total of 57 crossbreed pigs (Yorkshire × Landrace × Duroc) were used in this study. The treatment conditions for all pigs were the same both before and after slaughter, and all treatment conditions were approved by the Ministry for Food, Agriculture, Forestry, and Fisheries of South Korea. The pigs (110 ± 5 kg) were transported to the slaughterhouses under the same conditions and handling in two batches (30 and 27 pigs per each slaughter batch). At 45 min postmortem, muscle samples were taken from the *longissimus dorsi* muscles at the 8th thoracic vertebra, promptly measured muscle pH, and frozen in isopentane cooled by liquid nitrogen, and then stored at –80 °C until subsequent analysis. After chilling for 24 h in a 4 °C cold room, the pork loins were removed and evaluated for muscle pH.

B. Myosin light chain isoforms

Myofibrils were prepared using the method reported by Talmadge and Roy [4]. Muscle protein concentration was determined by Bradford's method [5]. The MLC isoforms were analyzed using sodium dodecyl sulphate-polyacrylamide gel electrophoresis, as described by Talmadge and Roy [4]. MLCs separated into 1s, 1f, 2f, and 3f isoforms. The bands were visualized by Commassie Brilliant Blue staining. Each MLC band was examined with an image analysis system for quantitative analysis (Kodak 1D image analysis software, Eastman Kodak Co., USA). The composition of fast vs. slow isoforms was obtained from the ratio of each MLC band density to the

density of total MLC band density within a sample.

C. Glycolytic characteristics

In order to determine the postmortem glycolytic rate, the muscle pH was measured directly on the carcasses at 45 min ($pH_{45\ min}$) and 24 h ($pH_{24\ h}$) postmortem using a spear type electrode (pH27-SS, IQ Scientific Instruments Inc., USA). The metabolite contents evaluated at 45 min postmortem. Glycogen and lactate contents were determined as was previously reported [2]. The muscle metabolite contents were expressed in μmol/g of fresh tissue, assuming 75% moisture content. The activity of pyruvate kinase (PK) was determined via the procedure described by Scopes [6]. The activity of lactate dehydrogenase (LDH) activity was measured via the procedure described by Gil et al. [7]. Enzyme activity was expressed as mmol NADH min g^{-1} of muscle.

D. Statistical analysis

The data for the contents of MLC isoforms were analysed using the SAS PC software [8] to calculate mean values, standard deviations, and overall ranges. Pearson correlation coefficients were evaluated using the partial correlation coefficients in order to determine the relationships between the content of MLC isoforms and glycolytic characteristics.

III. RESULTS AND DISCUSSIONS

The means, standard deviations, and overall ranges for the content of MLC isoforms in the porcine *longissimus dorsi* muscle are shown in Table 1. The content of MLC 1s isoform (4.85%) was smaller than the content of 1f or 3f isoforms (32.87 and 10.60%, respectively), and the essential light chain (ELC) fast/slow ratio was 11.04.

Adult skeletal muscle contains three ELC isoforms (including 1s, 1f, and 3f isoforms), and two regulatory light chain (RLC) consisting of 2s and 2f isoforms [3]. MLC slow isoforms (1s and 2s isoforms) are generally expressed in slow-twitch fiber with MHC slow isoform, whereas MLC fast isoforms (1f, 2f, and 3f isoforms) are expressed in fast-twitch fiber with MHC fast (2A, 2X, and 2B) isoforms [1, 3]. Thus, muscles with a higher content of ELC 1s isoform generally showed a slower contraction speed than muscle with a lower content of ELC 1s and a higher content of 3f isoform [3], and these characteristic is related to the metabolic characteristics in living animals. In this study (Table 2), MLC ls isoform content was correlated with the metabolite contents including glycogen content at 24 h postmortem (r=0.28, P<0.05) and lactate content at 45 min postmortem (r=–0.35, P<0.01), and was negatively correlated with LDH activity (r=–0.29, P<0.05) at 45 min postmortem. However, there were no correlations between the content of MLC isoforms and muscle pH ($pH_{45\ min}$ and $pH_{24\ h}$). The content of MLC 3f isoform had a positive correlation with PK activity (r=0.44, P<0.01), while the ratio of fast/slow isoform showed a positive correlation with lactate content (r=0.28, P<0.05) and PK activity (r=0.50, P<0.001) at 45 min postmortem. These results appear to indicate that the composition of MLC isoform is related to the metabolite contents and activities of glycolytic enzymes during postmortem period, although the composition of MLC isoform was found to have only a limited effect on the glycolytic rate.

Table 1 Content of myosin light chain (MLC) isoforms in the porcine *longissimus dorsi* muscle

	Mean±SD	Minimum	Maximum
MLC 1s isoform (%)	4.85±2.15	1.30	10.34
MLC 1f isoform (%)	32.87±3.60	26.05	39.20
MLC 3f isoform (%)	10.60±2.80	5.47	17.36
1f/3f ratio	3.38±1.17	1.52	6.62
ELC fast/slow ratio	11.04±5.57	3.75	30.00

Abbreviation: ELC, essential light chain.

IV. CONCLSION

The composition of MLC isoforms can influence postmortem rigor development in the porcine *longissimus dorsi* muscle, and consequently meat quality indirectly.

ACKNOWLEDGMENTS

This research was supported by iPET (the Korea Institute of Planning and Evaluation for Technology of Food, Agriculture, Forestry, and Fisheries). The authors thank the Korea University Food Safety Center for the use of their equipment and facilities.

REFERENCES

1. Schiaffino S, Reggiani C (1996) Molecular diversity of myofibrillar proteins: gene regulation and functional significance. Physiol Rev 76:371–423
2. Choi YM, Ryu YC, Kim BC (2007) Influence of myosin heavy- and light chain isoforms on early postmortem glycolytic rate and pork quality. Meat Sci 76:281–288
3. Choi YM, Kim BC (2009) Muscle fiber characteristics, myofibrillar protein isoforms, and meat quality. Livest Sci 122:105–118
4. Talmadge RJ, Roy RR (1993) Electrophoretic separation of rat skeletal muscle myosin heavy-chain isoforms. J Appl Physiol 75:2337–2340
5. Bradford M (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:248–254
6. Scope RK (1977) Multiple enzyme purifications from muscle extracts by using affinity-elution-chromatographic procedures. Biochem J 161:265–277
7. Gil M, Oliver M, Gispert A, Sosnicki AA, Lacoste A, Carrion D (2003) The relationship between pig genetics, myosin heavy chain I, biochemical traits and quality of M. *longissimus*. Meat Sci 65:1063–1070
8. SAS Institute (2004) Version 9.1.2. SAS Institute Inc. Cary, NC.

Table 2 Correlations between the content of myosin light chain isoforms and glycolytic characteristics in the porcine *longissimus dorsi* muscle

	Myosin light chain isoforms				
	1s isoform	1f isoform	3f isoform	1f/3f ratio	ELC Fast/slow ratio
Glycolytic rate					
Muscle $pH_{45\ min}$	0.21	–0.09	0.21	–0.23	–0.19
Muscle $pH_{24\ h}$	–0.13	0.01	0.01	0.01	–0.06
Metabolite contents					
Glycogen at 45 min postmortem	0.16	–0.10	0.14	–0.22	–0.02
Glycogen at 24 h postmortem	0.28^{*}	–0.16	0.09	–0.15	–0.19
Lactate at 45 min postmortem	-0.35^{**}	0.16	0.39^{**}	0.22	0.28^{*}
Lactate at 24 h postmortem	–0.12	0.16	–0.13	0.16	0.21
Activities of glycolytic enzymes at 45 min postmortem					
Pyruvate kinase	–0.20	-0.41^{**}	0.44^{**}	–0.17	0.50^{***}
Lactate dehydrogenase	-0.29^{*}	0.16	0.16	0.18	0.20

Levels of significance: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.
Abbreviation: ELC, essential light chain.