P-10-13

Influence Of pH On Soluble Myosin Heavy Chain In Early Postmortem Porcine Skeletal Muscle (#489)

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Introduction

This research aims to evaluate the contribution of protein profile in early postmortem (PM) muscle to fresh pork quality. Previous studies [1] have found myofibrillar proteins, such as myosin [2] were re-localized in the soluble fraction. A difference in water-soluble muscle protein profile at 45 min postmortem (Fig. 1) has been observed. A primary difference was the presence of myosin heavy chain (MHC) at approximately 200 kDa, demonstrating that conditions [3] of skeletal muscle early postmortem could impact the fate of MHC. Therefore, the objective of this study was to understand the relationship between the abundance of soluble MHC in early PM muscle and pH decline as well as fresh pork quality and value.

1. C. Li, G. Zhou, X. Xu, K. Lundström, A. Karlsson, R. Lametsch (2015) Phosphoproteome analysis of sarcoplasmic and myofibrillar proteins in bovine longissimus muscle in response to electrical stimulation. *Food Chemistry*, 175: 197-202
2. S. K. Matarneh, M. Beline, S. L. Silva, H. Shi, D. E. Gerrard (2018) Mitochondrial F_1-ATPase extends glycolysis and pH decline in an *in vitro* model. *Meat Science,* 137: 85-91
3. Y. H. Brad Kim, R. D. Warner and K. Rosenvold (2013)Influence of high pre-rigor temperature and fast pH fall on muscle proteins and meat quality: a review. *Animal Production Science,* 54: 375-395

Methods

Pigs (n=47) were fed a commercial corn and soybean diet, and were harvested at approximately 125 kg using standard industry procedures. Longissimus muscle (LM) was sampled and pH was measured at 45 min PM. Drip loss was determined on chops between 1 and 3 d of storage. At the completion of aging (14 d), fresh chops were used to determine color score, pH, cook loss, and star probe (kg) values. Proteins from 45 min and 1 d samples were solubilized in a low ionic strength buffer (50 mM Tris–HCl, 1 mM EDTA, pH 8.5), fractionated on non-reducing 8% SDS PAGE gels, and abundance of soluble MHC was determined using western blots. Primary antibody concentrations of 1:1,000 α-myosin (2F7; Developmental Studies Hybridoma Bank) and secondary antibody concentration of 1:10,000 goat anti-mouse (A2554; Sigma) were used [4]. Densitometry was used to quantify soluble MHC (200 kDa) in LM samples from 45 min and 1 d PM. A pooled whole muscle reference sample, aged 14 d, was loaded on each gel and used to normalize abundance of soluble MHC. Loins were categorized based on abundance of soluble MHC at 45 min PM: low MHC (LMHC, n=23) and high MHC (HMHC, n=24) groups. Statistical analysis was done using GLM procedure of SAS 9.4 with fixed effect of MHC classification group.

4. K. B. Carlson, K. J. Prusa, C. A. Fedler, E. M. Steadham, E. Huff-Lonergan, S. M. Lonergan (2017) Proteomic features linked to tenderness of aged pork loins. *Journal of Animal Science*, 95: 2533-2546

Results

Quality data from HMHC and LMHC classification groups are summarized in Table 1. A representative western blot is shown in Fig. 2. The pH of LM in HMHC was significantly greater than the LMHC group at 45 min PM. At 1 d PM, soluble MHC was not detected. Classification based on MHC solubility did not affect 14 d pH, cook loss, visual color score, drip loss, or star probe values. These results demonstrate that a greater abundance of MHC can be associated with a higher pH (pH>6.4) at 45 min PM. Amount of soluble MHC at 45 min had no affect on fresh pork quality characteristics.

Conclusion

These results demonstrate that the abundance of soluble MHC is dependent on rate of pH decline and a greater abundance may indicate slower metabolism. However, in this study there were no indications that classification based on soluble MHC in the early PM period could be utilized to predict fresh pork quality. While soluble MHC was not detected after the completion of rigor, early onset of rigor in PM metabolism could potentially be identified through the sarcoplasmic protein profile of skeletal muscle. Collectively, these observations show the connection between the rate of early PM pH decline on soluble MHC. The results suggest that muscle pH and time PM should both be considered when conducting trials aimed at defining early PM proteomic characteristics in pork.

Acknowledgement:
Funded in part by the Iowa Pork Producers Association project 18-199.

Notes

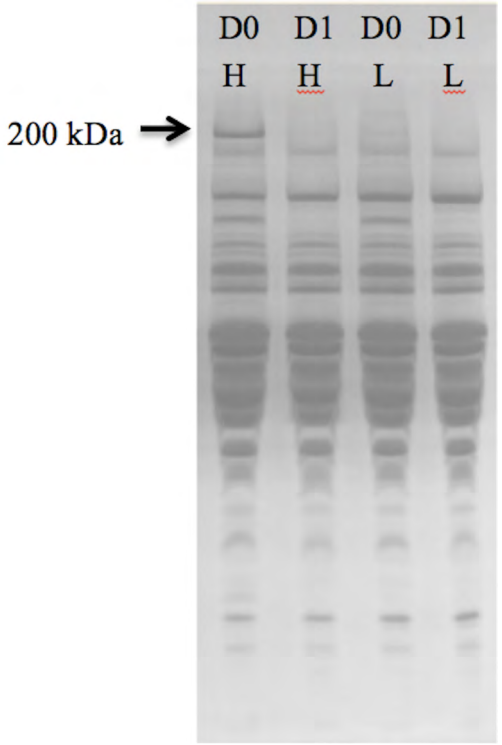

Figure 1.
Representative 8% SDS PAGE with Coomassie stain of MHC from HMHC (H) and LMHC (L) at 45 min (D0) or 1 d (D1) PM.

Item	HMHC (n=24)	LMHC (n=23)	SEM	*P*-value
45 min pH	6.45[a]	6.21[b]	0.04	<0.01
14 d pH	5.50	5.50	0.01	0.56
Cook Loss (%)	21.3	21.0	0.65	0.72
Color Score	3.19	3.17	0.11	0.93
Drip loss (%)	3.32	2.96	0.30	0.41
Star Probe (kg)	6.04	5.90	0.67	0.56
MHC Ratio[1]	0.52	0.05	0.17	

Table 1.
Summary of pork loin quality characteristics compared to MHC abundance, separated on MHC group.
[a, b] Means with different superscripts within rows are significantly different within classification ($P<0.01$).
[1] Ratio indicates abundance of MHC 45 min PM compared to MHC from a 14 d reference sample present on each gel. Because the groups were classified based on this trait, a statistical comparison was not made.

Notes

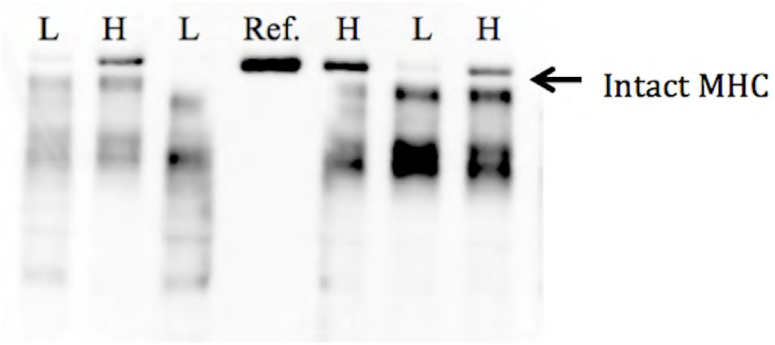

Figure 2.
Representative western blot of MHC from HMHC (H) and LMHC (L).

Notes