Variation in Proteolysis, Sarcomere Length, Collagen Content, and Tenderness Among Major Pork Muscles

T. L. Wheeler¹, S. D. Shackelford, and M. Koohmaraie
USDA-ARS, U.S. Meat Animal Research Center, P.O. Box 166, Clay Center, NE 68933

Introduction

The sources of variation in beef tenderness have been studied extensively (for review see Koohmaraie, 1992, 1996). However, pigs are slaughtered at a younger physiological age, have a smaller carcass mass, a greater, more uniform fat thickness, a faster metabolic rate (Etherington et al., 1987), and lower calpastatin activity (Koohmaraie et al., 1991; Ouali and Talmant, 1990). These differences may alter chilling rate, pH decline, rigor shortening, rate and extent of protein degradation, and connective tissue concentration; all of which could impact tenderness. However, there is limited data on pork tenderness, particularly for muscles other than the longissimus. Thus, the objectives of this experiment were to examine variation in proteolysis, sarcomere length, and collagen content among and within pork muscles, and the association of those factors with tenderness variation among and within muscles at 1 d postmortem.

Materials and Methods

The Roman L. Hruska U.S. Meat Animal Research Center (**USMARC**) Animal Care and Use Committee approved the use and treatment of animals in this experiment. Twenty-three white composite barrows were humanely harvested at the USMARC abattoir, and skinned carcasses were chilled at 0°C for 24 h. The longissimus lumborum, biceps femoris,

¹For more information: phone: 402-762-4229; fax: 402-762-4149; e-mail: wheeler@email.marc.usda.gov.

semimembranosus, semitendinosus, and triceps brachii, long head were dissected from one side of each carcass, vacuum packaged, frozen, and cut into 2.54-cm thick chops. Longissimus chops four, six, nine, and eleven were assigned to sensory evaluation. Longissimus chop three was assigned to biochemical analyses. For each of the other four muscles, four chops were cut. Chops one, two, and four were assigned to sensory evaluation and chop three was assigned to laboratory analyses.

Three raw muscle cubes, representing lateral, central, and medial locations within chop 3 of each muscle, were fixed according to Koolmees et al. (1986), and sarcomere length was determined as described by Cross et al. (1981). Degradation of the myofibrillar protein, desmin, was determined as described by Wheeler and Koohmaraie (1999). Total collagen was estimated from hydroxyproline quantification similar to the method described by Avery et al. (1996). Hydroxyproline was converted to collagen by multiplying by 7.25 (Woessner, 1961; Goll et al., 1963). Chops for trained descriptive attribute panel evaluation were cooked and evaluated as described by Wheeler et al. (1998).

Data were analyzed by mixed model analysis of variance with PROC MIXED of SAS (1997) for a split-plot design. Multiple linear regression and Piecewise regression were used to account for the effects of collagen content, proteolysis, and sarcomere length on tenderness rating.

Results and Discussion

Semitendinosus and triceps brachii received the highest tenderness ratings, followed by longissimus, then semimembranosus, and biceps femoris (Table 1). Longissimus had the most variability in tenderness. Variability in semitendinosus and triceps brachii tenderness ratings was relatively low. Ratings for amount of connective tissue were highest for semitendinosus, triceps brachii, and longissimus (least amount of connective tissue), intermediate for

semimembranosus, and lowest for biceps femoris (most connective tissue). Ratings for juiciness, pork flavor intensity, and off-flavor were slightly higher for semitendinosus and triceps brachii than for longissimus, semimembranosus, and biceps femoris.

The percentage of desmin that was degraded was greatest for longissimus, slightly lower for semimembranosus and biceps femoris, and near zero for semitendinosus and triceps brachii (Table 2). Although muscles were sampled at d 1 postmortem, longissimus, semimembranosus, and biceps femoris had relatively large ranges in the amount of degradation. Sarcomere length was much longer in semitendinosus and triceps brachii than the other three muscles. Longissimus sarcomere length was not different from either semimembranosus or biceps femoris. All semitendinosus and triceps brachii samples had sarcomere lengths of 2.0 µm or longer, whereas all samples from the other three muscles had sarcomere lengths of 2.0 µm or shorter. Collagen concentration was highest in biceps femoris, followed by triceps brachii, then semitendinosus, and lowest in semimembranosus and longissimus. Triceps brachii was most variable in collagen concentration.

Across all muscles, the desmin degradation had significant correlations ranging from weak to moderate with all traits except tenderness and connective tissue ratings (Table 3).

Sarcomere length had the strongest correlation with desmin degradation. Collagen content was weakly, but significantly, correlated with tenderness, connective tissue, flavor intensity, and off-flavor ratings. Sarcomere length was moderately correlated with all traits except collagen concentration. Tenderness rating was strongly correlated with sensory panel connective tissue amount rating, but only weakly correlated with an objective measurement of collagen concentration.

Within individual muscles, fewer significant correlations occurred than with all muscles combined. Tenderness rating was significantly correlated with connective tissue amount rating in all muscles. In triceps brachii, tenderness rating also was significantly correlated with juiciness ratings. In the longissimus, desmin degradation was correlated with sarcomere length and off-flavor rating. Tenderness rating also was significantly correlated with juiciness rating. Collagen concentration was significantly correlated with juiciness rating; and sarcomere length was significantly correlated with tenderness and connective tissue amount ratings. In both semimembranosus and biceps femoris, tenderness rating also was significantly correlated with desmin degradation. In addition, collagen concentration was significantly correlated with juiciness rating in the semimembranosus.

Implications

Sarcomere length, proteolysis, and collagen concentration must be known in order to explain differences among muscles in tenderness and, subsequently, devise strategies to eliminate tenderness deficiencies of specific muscles.

Literature Cited

- Avery, N. C., T. J. Sims, C. Warkup, and A. J. Bailey. 1996. Collagen cross-linking in porcine *M. longissimus lumborum*: Absence of a relationship with variation in texture at pork weight. Meat Sci. 42:355-369.
- Cross, H. R., R. L. West, and T. R. Dutson. 1981. Comparison of methods for measuring sarcomere length in beef semitendinosus muscle. Meat Sci. 5:261-266.
- Etherington, D. J., M.A.J. Taylor, and E. Dransfield. 1987. Conditioning of meat from different species. Relationship between tenderising and the levels of cathepsin B, cathepsin L, calpain I, calpain II and β-glucuronidase. Meat Sci. 20:1-18.
- Goll, D. E., W. G. Hoekstra, and R. W. Bray. 1963. Age-associated changes in muscle composition. The isolation and properties of a collagenous residue from bovine muscle. J. Food Sci. 28:503-509.
- Koohmaraie, M. 1992. The role of Ca²⁺-dependent proteases (calpains) in postmortem proteolysis and meat tenderness. Biochimie 74:239-245.
- Koohmaraie, M. 1996. Biochemical factors regulating the toughening and tenderization processes of meat. Meat Sci. 43:S193-S201.
- Koohmaraie, M., G. Whipple, D. H. Kretchmar, J. D. Crouse, and H. J. Mersmann. 1991.Postmortem proteolysis in longissimus muscle from beef, lamb and pork carcasses. J. Anim.Sci. 69:617-624.
- Koolmees, P. A., F. Korteknie, and F.J.M. Smulders. 1986. Accuracy and utility of sarcomere length assessment by laser diffraction. Food Microstr. 5:71-76.
- Ouali, A., and A. Talmant. 1990. Calpains and calpastatin distribution in bovine, porcine and ovine skeletal muscles. Meat Sci. 28:331-348.

- Wheeler, T. L., and M. Koohmaraie. 1999. The extent of proteolysis is independent of sarcomere length in lamb longissimus and psoas major. J. Anim. Sci. 77:2444-2451.
- Wheeler, T. L., S. D. Shackelford, and M. Koohmaraie. 1998. Cooking and palatability traits of beef longissimus steaks cooked with a belt grill or an open hearth electric broiler. J. Anim. Sci. 76:2805-2810.
- Woessner, J. F. 1961. The determination of hydroxyproline in tissue and protein samples containing small portions of this imino acid. Arch. Biochem. Biophys. 93:440-447.

Table 1. Descriptive statistics for trained sensory

panel traits among muscles

Trait/Muscle	Mean	SD	Minimum	Maximum	CV
Tenderness ^a					
Semitendinosus	7.2^{f}	.45	6.5	7.9	6.3
Triceps brachii	7.1 ^f	.38	6.4	7.7	5.4
Longissimus	6.4^{g}	1.08	3.4	7.5	16.9
Semimembranosus	5.7 ^h	.82	3.1	6.8	14.4
Biceps femoris	4.0^{i}	.62	2.8	5.1	15.5
Amount of connective					
Semitendinosus	7.6 ^f	.21	7.2	7.9	2.8
Triceps brachii	7.5 ^f	.26	7.1	7.9	3.5
Longissimus	7.2^{f}	.58	5.5	7.8	8.1
Semimembranosus	6.7^{g}	.43	5.4	7.3	6.4
Biceps femoris	5.4 ^h	.49	4.1	6.1	9.1
Juiciness ^c					
Semitendinosus	5.9 ^f	.35	5.3	6.6	5.9
Triceps brachii	5.8^{f}	.25	5.4	6.3	4.3
Longissimus	5.3 ^g	.28	4.6	5.8	5.3
Semimembranosus	5.4 ^g	.29	4.6	5.8	5.4
Biceps femoris	5.2^{g}	.25	4.8	5.6	4.8
Pork flavor intensity ^d					
Semitendinosus	4.9^{g}	.40	4.1	5.6	8.2
Triceps brachii	4.8^{g}	.41	4.0	5.4	8.5
Longissimus	5.2 ^f	.34	4.6	5.8	6.5
Semimembranosus	5.1 ^f	.39	4.0	5.7	7.6
Biceps femoris	5.2^{f}	.32	4.3	5.6	6.2
Off-flavor ^e					
Semitendinosus	2.9^{g}	.26	2.2	3.3	9.0
Triceps brachii	2.9^{g}	.24	2.4	3.4	8.3
Longissimus	$3.3^{\rm f}$.24	2.7	3.6	7.3
Semimembranosus	$3.2^{\rm f}$.27	2.5	3.7	8.4
Biceps femoris	3.1 ^f	.25	2.6	3.6	8.1

 $^{^{}a}1$ = extremely tough, 4 = slightly tough, 5 = slightly tender, 8 = extremely

tender.

b1 = abundant, 5 = slight, 6 = traces, 8 = none.

c1 = extremely dry, 5 = slightly juicy, 8 = extremely juicy.

d1 = bland, 4 = slightly bland, 5 = slightly intense, 8 = extremely intense.

2 = moderate 3 = slight, 4 = none.

e1 = intense, 2 = moderate, 3 = slight, 4 = none. fghi}Means in a column within a trait lacking a common superscript differ (P < .05).

Table 2. Descriptive statistics for traits associated with variation in tenderness among muscles.

	Mean	SD	Minimum	Maximum	CV
Desmin, % degraded					
Semitendinosus	.2°	5.2	-11.4	8.2	2600
Triceps brachii	.2°	9.1	-13.8	9.8	4550
Longissimus	39.3 ^a	25.4	-12.6	81.3	64.6
Semimembranosus	21.0^{b}	12.9	.2	43.6	61.4
Biceps femoris	18.5 ^b	15.4	8	54.3	83.2
Sarcomere length, µm					
Semitendinosus	2.45 ^a	.14	2.1	2.8	5.6
Triceps brachii	2.44 ^a	.17	2.07	2.70	7.1
Longissimus	1.78 ^{bc}	.10	1.57	2.01	5.6
Semimembranosus	1.83 ^b	.06	1.68	1.98	3.3
Biceps femoris	1.74 ^c	.10	1.57	1.93	5.9
Collagen, mg/g					
Semitendinosus	5.3°	1.0	3.7	7.4	18.9
Triceps brachii	6.0^{b}	1.5	4.4	9.8	25.0
Longissimus	4.1 ^d	.7	2.8	6.0	17.1
Semimembranosus	4.5 ^d	.7	3.2	5.7	15.6
Biceps femoris	7.1 ^a	1.1	5.2	8.9	15.5

^{abc}Means in a column within a trait comparing muscles lacking a common superscript differ (P < .05).

Table 4. Simple correlation coefficients for various traits within and across all muscles.

14010	Collagen, Sarcomere Connective Flavor						Off-
	mg/g	length, μm	Tenderness ^a	tissue ^b			flavor ^e
			A	ll Muscles			
Desmin, % degraded ^f	23*	61**	16	21	40**	.38**	.48**
Collagen, mg/g	-	.04	34**	38**	09	22*	31**
Sarcomere length, µm	-	-	.64**	.62**	.64**	38**	50**
Tenderness ^a	-	-	-	.94**	.60**	18	22*
		Se	mitendinosus				
Desmin, % degraded ^f	.17	.12	07	08	44*	07	02
Collagen, mg/g	-	14	.10	.00	23	30	35
Sarcomere length, µm	-	-	40	10	48*	.06	.20
Tenderness ^a	-	-	-	.43*	.40	.16	.06
		Tri	ceps brachii				
Desmin, % degraded ^f	03	02	.24	.08	.23	07	22
Collagen, mg/g	-	24	.11	.01	26	23	18
Sarcomere length, µm	-	-	.09	.02	.09	11	17
Tenderness ^a	-	-	-	.67**	.41*	12	05
		La	ongissimus				
Desmin, % degraded ^f	.01	42*	08	33	15	.41	.51*
Collagen, mg/g	-	25	21	04	47*	21	19
Sarcomere length, µm	-	-	.67**	.56**	.35	12	31
Tenderness ^a	-	-	-	.84**	.60**	.27	.16

Semimembranosus										
Desmin, % degraded ^f	.09	.00	.51*	.39	.30	.28	.12			
Collagen, mg/g	-	07	26	05	47*	05	32			
Sarcomere length, µm	-	-	21	06	.06	19	06			
Tenderness ^a	-	-	-	.85**	.23	.09	.05			
Biceps femoris										
Desmin, % degraded ^f	.03	28	.61**	.24	.34	.07	.11			
Collagen, mg/g	-	08	.02	05	04	25	29			
Sarcomere length, µm	-	-	19	02	04	.12	.20			
Tenderness ^a	-	-	-	.73**	.15	01	.20			

al = extremely tough, 8 = extremely tender.
bl = abundant, 8 = none.
cl = extremely dry, 8 = extremely juicy.
dl = extremely bland, 8 = extremely intense.
el = intense, 4 = none.
framework as a first decided and the second and the second are second as a first decided and the second are second as a first decided and the second are second as a first decided and the second are second as a first decided and the second are second as a se

^fPercentage of at-death desmin that was degraded.

^{*}P < .05.

^{**}P < .01.