

## EFFECT OF TEMPERATURE CONDITIONING ON TOUGHNESS IN HOT BONED PORK LOINS WITH HIGH OR LOW INITIAL pH

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### INTRODUCTION

Prevention of cold shortening and the accompanying toughness is the primary objective of using high temperature conditioning (HTC) in the hot boning system. Despite the faster post mortem glycolysis and earlier onset of rigor mortis in porcine muscles as compared to beef and lamb, cold shortening and cold toughening has been observed in pork (Marsh et al. 1972; Fischer et al. 1980; Dransfield and Lockyer 1985; Møller and Vestergaard 1987). Tenderness improvements have been demonstrated in excised loins by using delayed hot boning (Frye et al. 1985; Møller and Vestergaard 1987) or by holding hot excised cuts at elevated temperature before further chilling (Reagan and Honikel 1985). While HTC treatment may be effective for preventing cold induced toughness, such a system, especially in pork, may create a condition very similar to PSE characteristics or rigor contracture by causing increased drip loss and pale appearance of the meat (Honikel et al. 1986). Dransfield and Lockyer (1985) observed that excised pork loins held at 20°C tended to be tougher than by holding at 5°C. In ovine muscles, Bouton et al. (1984) showed that muscles accelerated in postmortem glycolysis by electrical stimulation could go into rigor at high temperatures and, in consequence, undergo shortening with resulting higher shearforce values and toughness. In unrestrained bovine muscles, Locker and Daines (1975) reported progressively greater shortening but a decline in shear force as the temperature of rigor approaches 37°C. The present experiments were designed mainly to study the effect on tenderness by allowing hot boned pork loins, representing lower high pH values, to go into rigor at 0, 15 and 30°C or by holding the hot excised, excised muscles at various combinations of 30/15°C before chilling at 0°C.

### MATERIALS AND METHODS

*M. longissimus dorsi* (LD) from both sides of conventionally slaughtered Danish Landrace or Yorkshire pigs (90 kg) were used in two experiments, each consisting of 16 carcasses. The pH value was measured in the LD muscle at the 15th rib 45 min p.m. and used to allocate carcasses in each experiment in two pH groups according to low (5.7 pH 6.1) or high (6.1 pH 6.5) pH1 values. Left sides served as controls and a section of the LD muscle, 12-15th rib, was removed at 24 h p.m. Right sides were used for treatments and an LD section, 9-15th rib, was excised/subsampled within 90 min p.m. and assigned by random to different temperature

treatments. Subsamples (approx. 250 g) were wrapped in polyethylene bags and totally immersed in ice water at 0°C or in water baths controlled at the selected temperatures until 24 h p.m. The conditioning temperatures and times were in Exp. 1: 0°C/24 h; 15°C/j h then 0°C/19 h; 30°C/3 h then 0°C/21 h. In Exp. 2 the following four treatments were used: 15°C/5 h then 0°C/19 h; holding at 30°C for 30, 60 or 90 min then at 15°C for respectively 4.5, 4.0 and 3.5 h before 0°C until 24 h p.m. Cooking was performed on blocks, 20x20x50 mm, taken from subsamples and individually heated in 0.09% NaCl at 80°C for 25 min.

**Measurements:** pH values of the muscles were determined using a Knick digital model 653 and a direct insertion probe electrode, Ingold lot 406-M3. An He-Ne laser with a wavelength of 632.8 nm was used for measurements of sarcomere lengths (Møller and Vestergaard 1987).

**Myofibril fragmentation:** Myofibrils were isolated as earlier described (Møller et al. 1973) and the average length of fragments determined by counting the number of sarcomeres per myofibril from a total of 100 myofibrils per subsample. Measurement of Troponin T and the proteins in the 28-32K dalton region after SDS gel electrophoresis of the myofibrils were carried out mainly as outlined by Penny and Ferguson-Pryce (1979). Two gels from each sample within a treatment were scanned

Table 1.- Mean values for temperature and pH in pork loins at different time post mortem obtained from controls (experiments 1 and 2)

	pH <sup>1</sup> group	n	time, hr p.m.				
			1	3	5	8	24
pH	L	16	5.90(.12)	-	5.81(.19)	5.66(.16)	5.60(.08)
pH	H	16	6.31(.09)	-	6.20(.12)	6.00(.13)	5.66(.07)
temp. <sup>o</sup> C	L/H	32		11.1(3.0)	5.2 (1.4)	5.0 (.7)	3.1 (.2)

Parenthetical values are standard deviations. <sup>1</sup>L = 5.7 ≤ pH<sub>1</sub> < 6.1; H = 6.1 ≤ pH<sub>1</sub> ≤ 6.5.

Table 2.- Warner-Bratzler shear force (WB), sarcomere length (SL), myofibril fragmentation, Troponin T, 28-32K dalton and color score in pork loins subjected to different post mortem treatments.

	pH group <sup>2</sup>	n	treatments <sup>1</sup>			
			control	1	2	3
WB, kg/cm <sup>2</sup>	L	8	4.87 <sup>a</sup>	6.80 <sup>b</sup>	5.65 <sup>ab</sup>	5.42 <sup>a</sup>
WB, kg/cm <sup>2</sup>	H	8	4.99 <sup>a</sup>	9.04 <sup>c</sup>	6.71 <sup>b</sup>	5.67 <sup>ab</sup>
SL, μ	L	8	1.65 <sup>bc</sup>	1.59 <sup>b</sup>	1.63 <sup>bc</sup>	1.64 <sup>bc</sup>
SL, μ	H	8	1.65 <sup>bc</sup>	1.44 <sup>a</sup>	1.61 <sup>b</sup>	1.67 <sup>c</sup>
fragmentation <sup>3</sup>	L/H	16	9.29 <sup>a</sup>	11.93 <sup>b</sup>	9.65 <sup>a</sup>	11.87 <sup>b</sup>
troponin T, % <sup>4</sup>	L/H	16	2.60	2.55	2.38	2.62
28-32K dalton, % <sup>4</sup>	L/H	16	1.13	1.13	1.22	1.12
color score <sup>5</sup>	L/H	16	2.93 <sup>c</sup>	2.82 <sup>c</sup>	2.31 <sup>b</sup>	1.62 <sup>a</sup>

<sup>1</sup> 1 = 0°C/24 h; 2 = 15°C/5 h - 0°C/19 h; 3 = 30°C/3 h - 0°C/21 h.

<sup>2</sup> see table 1(1). <sup>3</sup> number of sarcomeres/myofibril. <sup>4</sup> % of total myofibrillar proteins. <sup>5</sup> lean color, 1 = pale and 5 = dark. <sup>abc</sup> means with different superscripts differ significantly (P < .05).

on a LKB 2202 laser densitometer and protein concentrations (expressed as BSA protein equivalent) in the peak areas of Troponin T and the 28-32K dalton bands expressed as a percentage of the total myofibrillar proteins.

**Colour evaluation:** Following the 24 h chilling period, a section from each subsample was allowed to bloom for 30 min after cutting. A five member trained panel evaluated lean colour using a 5 point scale (1 = pale and 5 = dark).

**Shear force measurements:** Rectangular strips, 1 cm<sup>2</sup> in cross sectional area, were cut from the cooked meat and sheared at right angles to the fibre axis, using a Warner-Bratzler shear device for measuring peak shear force values (kg/cm<sup>2</sup>). Statistical analysis: Analysis of variance was used to estimate the significance of the main effects of conditioning treatments and pH group. Least significant differences were obtained using the error term from the analysis of variance.

## RESULTS AND DISCUSSION

**Experiment 1:** Means and standard deviations for pH and temperature from controls in experiments 1 and 2 are presented in Table 1. Differences in pH level between the two pH groups remain almost unchanged throughout the initial chilling period. Honikel et al. (1984) reported that the development of rigor in porcine muscles occur at pH 5.9 which implies that muscles can be rapidly chilled at this point without excessive muscle shortening. In the present experiment, the potential of cold shortening between the two pH groups is therefore expected to differ substantially if excised samples are exposed to rapid chilling at 1 h p.m. After conditioning at 15°C/5 h pH declined to 5.36 ± .09 in the low pH group and 5.60 ± .13 in the high pH group. Samples conditioned at 30°C/3 h showed 5.41 ± .04 and 5.51 ± .16 respectively in the low and high pH group. Thus, the period of conditioning at 15 or 30°C appear long enough to ensure completion of rigor.

The analysis of variance on the additional parameters is showing no main effect of pH group while conditioning treatment effects all measurements ( $P < .001$ ) except Troponin T and the 28-32K dalton component. However, pH group and treatment interact on WB values ( $P < .05$ ) and sarcomere length ( $P < .001$ ). Means of measurements are given in Table 2. As shown, rapid chilling at 0°C immediately after removal of muscles results in cold toughening and decreased sarcomere length in agreement with earlier reports (Dransfield and Lockyer, 1985; Møller and Vestergaard 1987). The extent of cold toughening is most pronounced in the high pH group where WB values increased 80% above the controls as compared to a 40% increase in the low pH group. Compared to controls, conditioning at 15°C/5 h results in higher WB values ( $P < .05$ ) in the high pH group only, which was unexpected although sarcomere length tended to be shorter in this group. Conditioning at 30°C/3 h causes a minor but nonsignificant increase in WB values as compared to controls.

In addition to prevent excessive cold shortening, HTC treatment may also accelerate the aging process (Locker and Daines 1975; Bouton et al. 1984). Tenderisation by aging has previously been associated with increased degradation of Troponin T and concomitantly higher levels of proteins in the 28-32K dalton region (Olsen et al. 1977; Penny and Ferguson-Pryce 1979). As shown in Table 2, no significant change is observed in either Troponin T or the 28-32K dalton proteins, which may be due to the conditioning at 30°C for only 3 h in the present study. Also myofibril fragmentation has been shown to increase as a result of aging (Olsen et al. 1977). From Table 2 is seen that myofibrils obtain higher resistance to fragmentation when conditioned at 30°C than at 15°C as revealed by longer fragments at 30°C. The latter result could reflect a higher degree of protein denaturation due to high temperature/low pH combinations (Wisner-Pedersen 1960). Decreased fragmentation also appeared from conditioning at 0°C. Low initial pH values or rapid pH decline at high temperature is well known to affect detrimentally the colour of pork muscles (Borchert and Briskey 1964). This is also supported by the present results (Table 2) and most pronounced for samples exposed to 30°C. However, both pH groups responded similarly to increased temperature during the conditioning period by progressive reduction in scores for meat colour acceptability.

Table 3.- Mean pH values prior to chilling at 0°C for hot boned pork loins subjected to different post mortem treatments.

	pH group	n	treatments <sup>1</sup>			
			1	2	3	4
pH	L	8	5.54(.09)	5.56(.15)	5.48(.09)	5.39(.04)
pH	H	8	5.83(.27)	5.94(.23)	5.88(.26)	5.78(.30)

Parenthetical values are standard deviations.

<sup>1</sup> 1 = 15°C/5 h - 0°C/19 h; 2, 3 and 4: holding at 30°C for 30, 60 or 90 min., then at 15°C for 4.5, 4.0 or 3.5 h, before 0°C until 24 h p.m.

<sup>2</sup> see table 1<sup>(1)</sup>.

Table 4.- Warner-Bratzler shear force (WB), sarcomere length (SL), myofibril fragmentation and color score in pork loins subjected to different post mortem treatments.

	pH group <sup>2</sup>	n	treatments <sup>1</sup>				
			control	1	2	3	4
WB, kg/cm <sup>2</sup>	L	8	4.47 <sup>a</sup>	5.29 <sup>abc</sup>	5.65 <sup>bc</sup>	5.59 <sup>abc</sup>	4.99 <sup>ab</sup>
WB, kg/cm <sup>2</sup>	H	8	5.30 <sup>abc</sup>	6.76 <sup>d</sup>	6.28 <sup>cd</sup>	5.34 <sup>abc</sup>	5.50 <sup>abc</sup>
SL, μ	L	8	1.72 <sup>bc</sup>	1.66 <sup>ab</sup>	1.69 <sup>ab</sup>	1.67 <sup>ab</sup>	1.68 <sup>ab</sup>
SL, μ	H	8	1.77 <sup>c</sup>	1.66 <sup>ab</sup>	1.64 <sup>a</sup>	1.68 <sup>ab</sup>	1.67 <sup>ab</sup>
fragmentation <sup>3</sup>	L/H	16	8.21 <sup>a</sup>	10.71 <sup>b</sup>	10.01 <sup>b</sup>	9.32 <sup>ab</sup>	10.05 <sup>b</sup>
color score <sup>4</sup>	L/H	16	3.05 <sup>b</sup>	2.63 <sup>ab</sup>	2.56 <sup>a</sup>	2.75 <sup>ab</sup>	2.50 <sup>a</sup>

<sup>1</sup> see table 3. <sup>2</sup>, <sup>3</sup>, <sup>4</sup> see table 2. abc means with different superscripts differ significantly ( $P < .05$ ).

**Experiment 2:** Mean pH values within treatments before chilling at 0°C are shown in Table 3. If pH 5.9 is considered as a useful guideline from which rapid chilling can begin in pork (Honikel et al. 1984), the length of conditioning periods used seems reasonable to prevent severe shortening. None of the additional parameters are significantly affected by pH group or interactions between pH group and treatment. Means are presented in Table 4. As also observed in exp. 1, samples exposed to 15°C/5 h caused significant higher WB values only in the high pH group as compared to controls ( $P < 0.05$ ). In the high pH group, conditioning at 30°C for 60 or 90 min followed by 15°C in 3.5-4 h results in similar shear force values as obtained from controls. Muscles from the low pH group did not respond much on WB values to the various treatments. The latter results appear to be in contrast with those obtained by Bouton et al. (1984) indicating heat toughening in electrical stimulated, hot boned ovine muscles held at 35 or 39°C from 0 to 120 min. Sarcomere length did not differ between groups of treatment but all treatments in the high pH group obtained shorter sarcomere length than controls ( $P < .05$ ). Data on myofibril fragmentation show no consistent response to increased time at 30°C but all, except 30°C/60 min, show longer fragments as compared to controls. Colour scores in conditioned samples at 30°C, except 30°C/60 min, are significantly lower than controls.

#### CONCLUSION

Rapid chilling of hot boned pork induce cold shortening and cold toughening which is highly related to the extent of post mortem glycolysis before boning. High temperature conditioning before rapid chilling improves tenderness without causing heat toughening but temperature should not exceed 15°C if detrimental effects on other muscle properties such as colour and water holding capacity are to be avoided.

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