

## TENDERIZATION OF MUSCLE VIA PRE-RIGOR INJECTION OF IONS

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**Key words:** tenderization, muscle, pre-rigor, hot-boning, injection, ions, sarcomere length, shortening**Background**

Consumer studies have shown that tenderness is the most important palatability factor in the acceptance of beef (Bratzler, 1971). It is known, and generally accepted, that meat pre-rigor is inherently more tender than its post-rigor counterpart but if pre-rigor muscle is chilled rapidly, cold shortening will occur. If certain substances could be used to modify the myosin-actin filaments of the myofibrils in pre-rigor musculature, then it is possible that cold shortening could be prevented. Muscle fibers could theoretically remain in a "relaxed state" to prevent toughening (Marsh and Carse, 1974) and improve water-holding capacity (Offer and Trinick, 1983).

There is need to more rapidly and efficiently process meat, one approach is to hot bone to conserve energy. However, one of the major problems associated with hot-boning is cold-shortening (Marsh et al., 1968, 1972). Cold-shortening occurs when the muscle is removed from the skeletal attachments and chilled rapidly (Locker and Hagyard, 1963). Shortening has a profound effect on meat tenderness: a shortening of 30-40% toughens muscle considerably. If hot-boning is to ever become applicable for the transformation of livestock into tender, fresh (including frozen), boneless, defatted, portion-controlled cuts for retail and institutional distribution, then it will be necessary to incorporate special procedures which could serve to insure desirable flavor, appearance (color and minimal purge, i.e. high water-holding capacity) and tenderness.

Several researchers have studied the effects of various compounds on shortening and tenderness of hot-boned pork (Kamstra and Saffle, 1959; Weiner and Pearson, 1969; Hoes et al., 1980) and beef (Carpenter et al., 1961; Wheeler et al, 1991) but the degree of tenderization obtained and the presence of detrimental effects (flavor undesirability, dark color etc) as well as the mechanisms of action remained unclear. There is some evidence that the mechanistic explanation could involve actomyosin dissociation (Cooke and Pate, 1985), calcium chelation (Weiner and Pearson, 1969), pH and/or ionic strength (Deatherage, 1963; Wu and Smith, 1987; Offer and Knight, 1988).

**Objectives**

The first objective of these studies was to investigate compounds selected in preliminary trials, particularly sodium pyrophosphate ( $\text{Na}_2\text{P}_2\text{O}_7$ ), sodium chloride ( $\text{NaCl}$ ) and calcium chloride ( $\text{CaCl}_2$ ) as to their effectiveness to prevent cold-shortening and subsequently maintain or improve tenderness. The second objective was to confirm preliminary results in a larger group of animals and investigate possible mechanisms of action.

**Methods**

For hot-boned treatments, ovine semimembranosus muscles were removed within 30 minutes of death. Injections were made using a syringe with a 12-gauge needle, injecting a volume equal to 10% of the muscle weight at 2 cm intervals in a square lattice pattern across the muscle. The hot-boned muscles were placed in individual plastic bags and placed on metal trays in a cooler at 2-4°C for 48 hr. Cold-boned muscles were left on the carcass in the same cooler as the hot-boned muscles and dissected at 48 hr postmortem (PM). The pH of each muscle was measured using an Orion pH meter with a spear-tip glass electrode inserted to 2.5 cm. Temperature was measured in the center of the muscle and muscle lengths were measured. Exudate was measured as the amount of liquid remaining in the plastic bag. Each muscle was cut into one or two 2.5 cm thick steaks, placed in a low-density polyethylene (LDPE) bag and cooked either by an 80°C waterbath for 60 min, cooled in ice water for 30 min or, broiled to an internal temp of 73°C then cut into eight 1.0 cm cubes and sheared with a Warner-Bratzler shear (WBS) device. Sarcomere lengths were measured on samples taken 48 hr PM which were homogenized in rigor buffer using a Polytron homogenizer. Sarcomere lengths were determined in the homogenized samples by measuring 10 sarcomeres in 50 myofibrils in each muscle sample with a Nikon microscope that had a phase contrast setting (Nikon Inc., Instrument Group, Garden City, N.Y.) at a magnification of 400X by use of an eyepiece micrometer standardized with a stage micrometer using an image analysis program.

**Results and discussion**

Initially it was found (Table 1) that injection of both 0.15 M sodium chloride ( $\text{NaCl}$ ) and 0.15 M  $\text{NaCl}$  + 0.225 M sodium pyrophosphate ( $\text{Na}_2\text{P}_2\text{O}_7$ ) produced more tender meat than the non-injected control. The result for the 0.15 M  $\text{NaCl}$  injection was unexpected since this was intended as a physiological injection control. Injection of 0.3 M calcium chloride ( $\text{CaCl}_2$ ) treatment produced the most tender muscles, but caused significant shortening and greater exudate. Since the aim of this work was to keep muscle in a pre-rigor state and avoid shortening, use of  $\text{CaCl}_2$  was discontinued. Since sodium chloride was considered to be a more desirable additive than sodium pyrophosphate from a consumer perspective, use of sodium pyrophosphate was discontinued. Also, since a non-hot-boned control was not used initially, it was included in the next trial as shown in Table 2. However, in this trial it was found that the cold-boned muscles were less tender than the hot-boned muscles, and, injection of  $\text{NaCl}$  did not improve tenderness of the hot-boned muscles. It did not appear that the hot-boned muscles cold-shortened, even though they were below 10°C by 3 hours postmortem. These muscles were stored in a different cooler than those of the previous trial, and they were exposed to a more constant temperature and greater air circulation. It is not known why these muscles did not undergo cold-shortening or cold-induced toughening. The differences in tenderness between the cold-boned and hot-boned muscles are similar to those found by Marsh et al. (1987) corresponding to the 3 hr pH value associated with a rapid early PM glycolytic rate.

Table 1: Comparison of non-injected and injected (0.15 M NaCl, 0.15 M NaCl + 0.15 M NaPPi and 0.3 M CaCl<sub>2</sub>) mean Warner-Bratzler shear (kgF) values, pH, exudate (ml) after 24 and 48 h, and, muscle length (cm) and sarcomere length ( $\mu$ m) after 48 h of chilling for hot-boned sheep Semimembranosus muscles (n=3 muscles per treatment).

TREATMENT:	24 HOURS POSTMORTEM				48 HOURS POSTMORTEM				s.e.
	Non-injected	0.15 M NaCl	0.15 M NaCl + 0.225 M Na <sub>2</sub> PPi	0.3 M CaCl <sub>2</sub>	Non-injected	0.15 M NaCl	0.15 M NaCl + 0.225 M Na <sub>2</sub> PPi	0.3 M CaCl <sub>2</sub>	
Shear values	7.34 <sup>de</sup>	7.23 <sup>de</sup>	6.67 <sup>bcd</sup>	5.91 <sup>b</sup>	7.96 <sup>c</sup>	7.21 <sup>de</sup>	5.75 <sup>ab</sup>	5.41 <sup>ab</sup>	0.27
pH	5.68 <sup>bc</sup>	5.67 <sup>bc</sup>	5.57 <sup>ab</sup>	5.68 <sup>bc</sup>	5.63 <sup>b</sup>	5.66 <sup>bc</sup>	5.52 <sup>a</sup>	5.70 <sup>ab</sup>	0.03
Exudate	0.86 <sup>a</sup>	2.51 <sup>b</sup>	2.08 <sup>b</sup>	5.34 <sup>c</sup>	1.09 <sup>a</sup>	2.66 <sup>b</sup>	2.71 <sup>b</sup>	5.97 <sup>c</sup>	0.80
Muscle length					10.6 <sup>b</sup>	13.5 <sup>c</sup>	13.5 <sup>c</sup>	7.4 <sup>a</sup>	0.8
Sarcomere length					1.81 <sup>a</sup>	1.81 <sup>a</sup>	1.92 <sup>a</sup>	1.35 <sup>b</sup>	0.10

<sup>a,b,c,d,e</sup>Means in an entire row lacking a common superscript letter differ (P < .05).

Table 2: Comparison of cold-boned (CB), hot-boned (HB) and hot-boned + injected with 0.15 M NaCl (HB+I) ovine semimembranosus muscle mean temperature and pH after 3 and 48 hr of chilling, and mean muscle shortening, sarcomere length ( $\mu$ m) and Warner-Bratzler shear (kgF) values after 48 h of chilling (n=6 sides per treatment, split plot model).

TREATMENT:	3 HOURS POSTMORTEM				48 HOURS POSTMORTEM			
	Cold-Boned	Hot-Boned	Hot-boned + injected	s.e.	Cold-boned	Hot-boned	Hot-boned + injected	s.e.
Temperature	17.3 <sup>a</sup>	7.1 <sup>b</sup>	6.8 <sup>b</sup>	0.6	2.8	2.9	2.8	0.1
pH	5.88 <sup>a</sup>	6.24 <sup>b</sup>	6.28 <sup>b</sup>	0.05	5.54	5.58	5.60	0.03
Muscle shortening					0 <sup>a</sup>	19.8 <sup>b</sup>	22.1 <sup>b</sup>	2.4
Sarcomere length					2.02	1.96	2.03	0.08
Shear values					9.09 <sup>a</sup>	7.64 <sup>b</sup>	6.74 <sup>b</sup>	0.50

<sup>a,b</sup>Means in a row within each time postmortem lacking a common superscript letter differ (P < .05).

## Conclusions

There is some evidence that injection of ions causes a tenderizing effect in pre-rigor excised muscle. However, this may be related to the degree of shortening of the muscle. Simply excising muscles and quickly chilling them such that the internal temperature declines rapidly does not ensure the cold-shortening or cold-induced toughening phenomenon.

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