EXPLORATORY EXPERIMENTS ON VERY FAST CHILLING OF BEEF MUSCLE: A SURVEY OF RESULTS WITH CONCLUSIONS

D. Demeyer, D. Steen & E. Claeys

Department of Animal Production, Laboratory of Meat Science and Meat Technology, University of Gent, 9090 Melle, BELGIUM

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BACKGROUND

Over the past year, three small scale experiments were performed at our laboratory to explore the possible benefits of "very fast chilling" (VFC) on beef quality. Indeed, initially doubt was expressed as to the feasibility of VFC of beef carcasses, in view of

* the size of carcasses and the thermal conductivity of beef limiting rate of chilling and
* the risks of cold shortening and toughening

Beneficial effects of VFC have however been demonstrated, mainly using lamb carcasses. The authors have elegantly rationalized the tenderizing effects observed in terms of an accelerated release of calcium ions within the muscle cell, stimulating calpain activity (Beltrán et al, 1986) (Jaime et al, 1992) (Jaime et al, 1993). Similar effects have been demonstrated for beef (Bowling et al, 1987). Our experiments made use of excised muscles, chilled to obtain VFC conditions: -2 - 0°C within 4-5 hrs. p.m. (Bowling et al, 1987). In earlier reports, a detailed description and discussion of individual experiments was presented, with emphasis on between animal and between muscle differences and on anomalies in texture development (Steen et al, 1995a, 1995b).

OBJECTIVES

An attempt is made to present overall results pertaining to final meat quality characteristics and to mechanisms of action of VFC.

METHODS

Animals and treatments: Experiments involved a total of five bulls (4 Belgian Red White, 1 Belgian Black White; 278 - 525 kg live weight, carcass yield ca. 50 %) slaughtered at our slaughterhouse after captive bolt stunning and pithing. In expt.1, a *Longissimus thoracis* sample (7-8th rib) was excised 1.5 h after slaughtering from one carcass side of one animal each, chilled in ice for 5 hrs. and, after further chilling at 10°C, compared with its analog on the other sides chilled conventionally (2°C) on carcass and excised 24 hrs. p.m. Both steaks were then vacuum packed and chilled at 2°C for up to 7 d p.m. Expt.2 involved excision of the *Semitendinosus* muscle from both sides of two animals at 30 min. p.m. and restraining all muscles individually on small wooden boards by nailing. One muscle of each animal was then wrapped in a plastic bag and immersed in ice, whereas the other muscles were conventionally cooled (room temperature 15-20°C). After 24 hrs., muscles were removed from the boards, vacuum packed and further chilled conventionally (2°C). Expt.3 was a replicate of expt.2, involving one animal only, but *Semitendinosus* muscles were divided longitudinally into three parts, subjected in pairs (left and right hand side) to ice immersion, refrigerator cooling and conventional chilling. After 24 hrs., samples were vacuum packed and held for another 24 hrs. at 2°C.

Analyses: Each experiment involved continuous recording of temperature and measurement at regular intervals of pH. Samples taken at regular intervals were vacuum packed, stored at - 80°C and, after thawing overnight at 15°C, used for determination of sarcomere length, shear force, cooking losses and drip. Separate samples kept at - 80°C were put at - 18°C overnight and then used for determination calpain/calpastatin activities. Other samples were ground, vacuum packed, kept at - 80°C and used without thawing for myofibrillar fragmentation by semi-quantitative SDS-PAGE as described earlier (Claeys et al, 1995). Waterextractable calcium was also determined on these samples using atomic absorption after extraction as described by Nakamura et al (1973).

RESULTS AND DISCUSSION

Temperature and pH drop

In all experiments, VFC conditions were practically reached: at 5 hrs. p.m. temperatures were 0; 5 and 0°C (expts.1, 2 and 3 respectively) for ice chilled samples compared to conventional chilling (CC) temperatures of 15-25, 23 and 15-18 °C. No clear differences in rates of pH drop could be observed: 6.1-6.3, 6.4-6.8 and 6.4 for ice chilled samples at 5 hrs. p.m. for expts.1, 2 and 3 respectively, to be compared with 6.2, 6.0-6.4 and 6.6 for conventional chilling. A significantly lower rate of pH drop was observed only in expt.2 (Steen et al, 1995b).

Meat quality

Table 1 shows that in all experiments VFC resulted in higher shear force measurements, when compared to CC. Also, this finding was associated with higher cooking and drip losses. It should be mentioned however that some surprising observations were made: one animal showed very tough meat in expt.1 and for one animal in expt.2, the muscle studied (*Semitendinosus*) did not tenderize. Meat was however very tender in this experiment.

Mechanisms

De data in table 2 provide some evidence suggestive of increased myofibrillar fragmentation due to VFC: in expt.2, VFC results in more Troponin T degradation and 30 kD production, both at 2 and 7 days post mortem, whereas lower amounts of titin and nebulin are recovered at 7 days post mortem. This suggests a slower degradation of the latter proteins, compared to troponin T. This finding is

corroborated by lower recoveries of m-calpain after VFC, indicative of its higher activity, in expts.1 & 3. An increase in free Ca^{2+}, claimed to be the causative factor for increased calpain activity was only observed in expt.3. Any extra tenderizing effect of such changes however is probably overcome by cold shortening induced by VFC and apparent from shorter sarcomeres, observed in the VFC samples. It should be mentioned that much shorter sarcomeres indicative of cold shortening due to VFC were observed 1 h p.m. in expt.1 (Steen et al. 1995a).

Table 1: Summary of effects of very fast chilling (VFC) on some beef quality characteristics compared to conventional chilling (CC)[a] Two successive values separated by ";" represent measurements from 2 different animals

expt.	days p.m.	Shear force (N)		Cooking loss (%)		Drip (%)	
		CC	VFC	CC	VFC	CC	VFC
1	8	36.8 ; 97.3	69.0 ; 105.5	24.6 ; 33.5	25.7 ; 35.4		
2	2	22.7 ; 34.5	33.5 ; 48.6	30.5 ;32.5	33.7 ; 32.4		
	7	23.4 ; 29.3	27.8 ; 39.6	31.7 ; 32.5	32.7 ; 32.8	1.2 ; 1.9	5.0 ; 5.2
3	2	56.2	71.8*	30.4	34.0*		

* significant difference ($p < .05$) [a] Experimental details in Methods.

Table 2: Effects of very fast chilling (VFC) on sarcomere length (SL), myofibrillar protein fragmentation, m-calpain activity and free Ca (Ca) concentration in beef muscle at different times post mortem, as compared to conventional chilling (CC)[1]

	Experiment 1				Expt. 2		Expt. 3	
	CC		VFC		CC	VFC	CC	VFC
days p.m.	1	8	1	8	2	2	1	1
SL (µm)	1.86 ; 1.50	2.02 ; 1.48	1.16 ; 1.09	1.09 ; 1.05	1.99 ; 2.01	2.02 ; 2.06	2.15	1.89
Titin[a]	54 ; 55	52 ; 58	54 ; 59	47 ; 41				
Nebulin[a]	22 ; 24	16 ; 18	26 ; 26	16 ; 13				
Trop.T[a]	5 ; 16	0.4 ; 7	6 ; 11	0.3 ; 1				
30 kD[a]	4 ; 0.5	18 ; 8	5 ; 2	22 ; 10				
Ca (mg/kg)	11 ; 4		13 ; 9		18 ; 17	15 ; 15	9	9
µ-calpain[b]	2.3 ; 7.3		0.5 ; 2.8		28[c] ; 31[c]	27[c] ; 33[c]	11	5

[1] details in Methods [a] in mg BSA equivalents/mg myofibrillar protein [b] units/g meat [c] values 1h p.m. At 2 d p.m. m-calpain could not be detected

CONCLUSIONS

The preliminar experiments presented here suggest that VFC of isolated beef muscles toughens beef, probably due to cold shortening and/or cold toughening. In some experiments, some evidence was obtained for a increased rate of protein fragmentation, associated with increased release of Calcium and increased m-calpain activity. Any tenderizing effect however of such change, seems to be overcome by cold shortening and/or toughening. Associated defects of moisture binding capacity were also observed.

PERTINENT LITERATURE

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