Effectiveness of Combined-Tenderization Processes in Tough Muscles.

Gonzalez, Claudia B.
Instituto Tecnología de Alimentos, Centro Agroindustria, CNIA, Instituto Nacional de Tecnología Agropecuaria (INTA), Casilla de Correo 77, (1708), Morón, Buenos Aires, Argentina. claugon@arnet.com.ar

The *Cutaneus trunci* (matambre) and *Biceps femoris* (carnaza cuadrada) muscles are used extensively in Argentina to prepare different types of food. However these muscles have the disadvantage of being traditionally tough or their tenderness level varies from animal to animal, hence if higher or at least a consistent level of tenderness could be guaranteed these muscles could be marketed at higher prices.

It was well-demonstrated that tenderness improvement can be managed by the use of the refrigerated *postmortem* storage, however in Argentina, the drawback of using the aging process for muscles of a low-commercial value is based on economics. Consequently, a study was conducted to apply a calcium chloride treatment in combination with the aging process in order to produce tenderization and increase market prices.

The mechanism of action of calcium-induced tenderization could be either described by the theory which postulates that calcium-dependent calpain proteolytic system is responsible for the postmortem proteolysis that results in meat tenderization (Koohmaraie, 1994) or by "the calcium theory", which suggests that a calcium-ion concentration of 0.1 mM induces the weakening of myofibrils and rigor linkages formed between protein filaments (Takahashi, 1999). Even though the aim of this research was not focused on establishing the mode of action of the calcium-ion, it was our intent to demonstrate that the effect exerted by the calcium-ion on the muscle ultra structure is related to meat tenderization.

For this research, two sets of samples were prepared to compare the tenderness level achieved: treated-samples (calcium chloride treatment) and non-treated samples (no calcium), both set of samples were then vacuum packaged and aged at 1-2°C during 0, 1, 2, 3, 4, 5 and 7 days.

For the *Biceps femoris* muscle the traditional injection procedure was used to incorporate 10% (w/w) calcium chloride solution; but in the case of *Cutaneus trunci* muscle, which is a thin and fibrous tissue, the best incorporation was obtained with a marination procedure. In order to avoid introducing undesirable flavors a trained panel evaluation was conducted to determine the maximum calcium chloride concentration to be used. Panelists found that 0.25M calcium chloride solution was a suitable concentration to apply in both cases (Gonzalez *et al.* 2000).

Calcium-ion concentration and distribution was performed by taking aliquots from different areas of each sample, then digested and calcium-ion was determined by Atomic Absorption Spectrophotometry. To evaluate the integrity of muscular protein network by SDS-PAGE or by the light (LM) and transmission electron (TEM) microscopy the same set of samples were used, plus a 30 min.-slaughtered, and a 20 hours-slaughtered water-injected or marinated muscles in order to separate the effects of water from the calcium effects.

Tenderness was evaluated by the Myofibrillar Fragmentation Index (MFI) method, and by a consumer panel.

Calcium salt was detected only in calcium-treated samples and was evenly distributed throughout the muscular tissue. The average calcium ion concentration in the tissue was 4.25 and 3.00 mg/g protein for *B. femoris* and *C. trunci* muscles.

LM results showed muscular structural modifications, and also morphological changes were observed in muscular fibers, in the space in between myofibrils, swelling of the adjacent regions of the sarcomere, and edema between tissue elements caused by aging process but exacerbated by calcium addition. TEM confirmed the previous results, and the magnification helped in showing the modifications of the surface ultra structure of muscle fibers, which indicated disruption of perimysium with an increase in the fascicular space, rupture of the myofibrils -in the region of the Z-line area- and of the covering membrane leading to fragmentation of the fibers.

Protein degradation of calcium-treated and aged samples was also corroborated by means of the SDS-PAGE methodology, which showed increase in low molecular weight protein bands and disappearance of desmin-like band.

MFI scores of *Cutaneus trunci* muscle showed that marinated samples after three days of aging achieved a 58.5 % increase in tenderness compare to day 0 and almost the same value obtained after aging the non-treated samples during 7 days. Consumer panel evaluation included only calcium treated samples aged 3days and non-treated samples aged 7 days, it was found that there were not a significant difference between treated samples and non-treated samples, and when panelists were asked to choose between them, marinated samples were preferred because of tenderness.

In the case of *Biceps femoris* muscle, the MFI values made evident once more that the calcium chloride injection produced higher tenderness level in a substantially shorter aging period, since the percentage of tenderness increment achieved by treated samples + 2 days of aging (45.1% increase compared to day 0) was similar to those obtained with longer aging periods and even higher than the one obtained in non-treated samples aged for 7 days (38.2%). Two consumer panel evaluations were carried out for this muscle, the first comparison included a control sample (C, without any treatment) and the sample injected with calcium chloride-aged 2 days (Ca2). In a second comparison also evaluated was Ca2 *vs.* the sample aged 7 days (A7). A significant difference ($p<0.05$) was noted between both samples, C and Ca2, with 71% of the members selecting the treated sample as the tenderer sample. There was not a significant difference between Ca2 and A7.

This technological approach indicated that even for tough muscles such as *Cutaneus trunci* and *Biceps femoris*, the calcium chloride treatment was effective in reducing the postmortem aging time necessary to achieve an acceptable level of tenderness. Also it was demonstrated that the calcium-ion participation in this process modified the tissue integrity, which was translated into an improvement of meat tenderness.

The effectiveness of the combined procedure to improve tenderness would allow the marketing of low-commercial-value meat products at better prices leading to a benefit for both, consumers and meat industry.

References

Gonzalez, C. B; Salitto, V; Carduza, F; Pazos, A. and Lasta, J. (2000). *Meat Science* accepted for publication, June 2000.
Koohmaraie, M. (1994). *Meat Science,* 36, 93-104.
Takahashi, K. (1999). In *Proceeding* 45th International Conference of Meat Science & Technology , Japan.