EFFECT OF SALT TREATMENT ON BEEF MEAT PROTEIN THERMAL BEHAVIOUR

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Introduction

Differential scanning calorimetry (DSC) has become a useful technique to study the thermal behaviour of proteins (Stabursvik and Martens, 1980). Analysis performed in different muscles established the presence of three distinctive peaks, 54-58°C, 67°C and 71-83°C (Parson and Paterson, 1986, Mietsch *et al.*, 1994), mainly related to the thermal denaturation of myosin heads; myosin tails, sarcoplasmic proteins and collagen; and actin respectively. Studies of thermal transitions have involved protein conformation changes induced by environmental factors, such as pH and ionic strength (Findlay and Barbut, 1990) or by the addition of different additives (Farkas and Mohácsi-Farkas, 1996). These changes are responsible for the variation of meat protein stability (Ensor *et al.*, 1991). Previous research of the group was focused on the increase of water holding capacity (WHC) of beef products containing added sodium chloride (SC) and sodium tripolyphosphate (STPP) and cooked by the *sous vide* system (Vaudagna *et al.*, 2005). The purpose of the present study was to investigate the thermal behaviour of muscle proteins, including isolated myofibrillar proteins, when SC and/or STPP were injected in beef *Semitendinosus* muscle.

Materials and Methods

Ten *Semitendinosus* muscles were obtained 72 h *postmortem* from a commercial processor. Salt solutions were injected into the muscles (10% w/w) with an automatic multineedle injector to give the final concentrations (g/100 g injected muscle) indicated in Table 1.

Table 1.

Salt Treatment	SC	STPP
1	0.7	0.25
2	1.2	0.25
3	0.7	-
4	-	0.25
5	non injected	

After the injection, muscles were first vacuum packaged, then continually tumbled at 5 rpm for 8h (1.5±0.5°C) in a Lance Industries tumbler, and stored at -80°C until analyses were performed. All salt treatments were assay in duplicate. Myofibrillar proteins (MP) isolation was conducted according to Culler *et al.*, (1978). Final protein content was determined by estimating the concentration of total N by Kjeldahl method. MP patterns were monitored using discontinuous 3-12% SDS-PAGE according to Laemmli (1970).

A Perkin-Elmer Pyris-1 DSC was used to study the thermal denaturation of the whole muscle proteins and of the isolated MP. Samples (5-15 mg) were weighed into the aluminium pan, then hermetically sealed, allow to equilibrate at 25°C for 2 min., and finally heated from 25 to 100°C at a rate of 10°C/min. An empty pan was used as reference. Heat capacity profiles (endotherm: C_p vs T), calorimetric enthalpy (ΔH) and endothermic peak (T_m) were obtained using the software Pyris-7 (Perkin-Elmer). Finally, samples were rescanned to corroborate the irreversibility of the denaturation process.

All experiments were replicated twice with duplicate measurements of each one. Analysis of variance (ANOVA), means, and standard errors were determined using SAS (1994). Least significant differences ($p<0.05$) were used to establish differences among salt treatments.

Results and Discussion

Electrophoretic pattern of MP showed non significant differences among treatments, suggesting the absence of structural changes related to protein mobility, induced by the presence of salts.

Thermal analysis of non injected whole muscle is shown in Figure 1 as an example. Data show three endothermic peaks at 56.4±0.5°C (I); 68.4±0.5°C (II) and 79.4±0.4°C (III), corresponding to myosin heavy chain (MHC), sarcoplasmic proteins/collagen and actin, respectively. ΔH parameter of these peaks were 1.80±0.13 J/g protein; 0.56± 0.10 J/g protein and 1.71± 0.90 J/g protein, respectively.

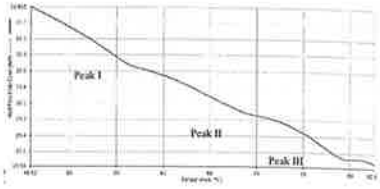

Figure 1: Thermal analysis of non injected Semitendinosus muscles.

When 0.7%SC was injected, actin T_m significantly decreased, denoting the protein thermal sensitivity increment. This effect is probably due to the perturbation of protein hydration induced by SC. In contrast, myosin and sarcoplasmic proteins/collagen T_m showed non significant reduction. Similar results were published by Benjakul *et al.*, (2000), who reported that actin T_m decreased when a threadfin bream muscle was injected with 2.5%SC. On the other hand, injection of 0.25%STPP did not lead to significant changes in T_m or ΔH, with the exception of sarcoplasmic proteins/collagen ΔH, which was non significantly reduced. When both salts were injected in combination, 0.7%SC and 0.25%STPP, a significant decrease in actin T_m and sarcoplasmic proteins/collagen ΔH was observed. This thermal behaviour, increase in actin instability, is similar to the one caused by 0.7%SC injection. Muscles injected with higher SC concentration (1.20%SC + 0.25%STPP) showed a significant decrease in actin T_m, and a small decrease in the T_m of the other proteins. These results are in accordance with Shand *et al.*, (1994), who had reported that in *Semimembranosus* muscles injected with 0.35%STPP and 1%SC, the first transition peak (myosin T_m) was not affected, and actin transition became smaller and its T_m shifted to a lower temperature.

MP thermal analysis of non-injected muscles showed the presence of only two of major thermal transitions, peak A at 59.4°C (4.29 J/g protein) and peak B at 72.9°C (2.28 J/g protein) corresponding to myosin and actin respectively. It is interesting to denote that in the isolated myofibrils there is a shift to lower transition temperatures for actin. This modification could probably be due to the effect of pH and ionic strength variation induced by the isolation conditions (Ma and Harwalkar, 1991; Murphy *et al.*, 1998). When 0.7%SC was injected, both myosin and actin ΔH decreased. The reason for the difference in myosin instability increment obtained between the whole muscle and the isolated proteins is the higher exposure of the extracted proteins to the environmental conditions. Similarly to the whole muscle, non significant differences were found when 0.25% STPP was injected. When salts were injected together (0.7%SC + 0.25%STPP), both myosin and actin ΔH decreased. Even more, when SC was increased (1.20%SC + 0.25%STPP), both thermal parameters (T_m and ΔH) of myosin and actin decreased. The present results corroborate previous ones, suggesting the important effect of SC on protein thermal stability. Again, in the case of isolated proteins (MP) greater protein instability was demonstrated by the reduction of both thermal parameters, involving probably the mentioned effect of the isolation conditions. The thermal behaviour of the isolated MP corroborates and supports the protein thermal profile of whole muscle.

Conclusions

Present results demonstrate that MP are effectively involved in the thermal behaviour depicted by the whole muscle. SC has an important destabilising effect on muscular myofibrillar proteins in a salt concentration dependent manner. The effect of SSTP was not clear and further studies are needed.

References

Benjakul, S, Visessanguan, W. and Kookkeaw, P. (2000). *Songklanakarin Journal of Science Technology*, 22(3), 329-338.

Culler, R.D., Parrish, F.C.(Jr.), Smith, G.C. and Cross, H.R. (1978). *Journal of Food Science,* 43, 1177.

Ensor, S.A., Sofos, J.N. and Schmidt, G.R. (1991). *Journal of Food Science*, 56, 175-179.

Farkas, J. and Mohácsi-Farkas, C. (1996). *Journal of Thermal Analysis and Calorimetry*, 47(6), 1787-1803.

Findlay, C.J. and Barbut, S. (1990). In: VR Harwalkar, CY Ma, (Eds), Thermal analysis of Foods. Elsevier. New York.

Laemmli, U.K. (1970). *Nature*, 227, 680-685.

Ma, C.-Y. and Harwalkar, V. R. (1991). In Advances in food and nutrition research, Academic Press, Inc., New York.

Mietsch, F., Halász, A. and Farkas, J. (1994) Die Nahrung, 38 (1), 47-52.

Murphy, R. Y., Marks, B. P. and Marcy, J. A. (1998). *Journal of Food Science*, 63(1), 88-91.

Parsons, S.E. and Patterson, R.L.S. (1986). *J. Food Technol.*, 21(2):123-131.

SAS (1994) Cary, N.C.: SAS Institute.

Shand, P.J., Sofos, J.N. and Schmidt, G.R. (1994). *Journal of Food Science*, 59(4), 711-715.

Stabursvik, E. and Martens, H. (1980). *Journal of Science and Food Agriculture*, 31, 1034-1042.

Vaudagna, S. R., Lasta, J. A. and Sánchez, G. (2005). In Y. H. Hui, I. Guerrero Legarreta & M. R. Rosmini (Eds.), Ciencia y tecnologías de carnes. México D.F.: Limusa S.A.