STUDY OF SEVERAL FACTORS THAT INFLUENCE THERMAL ANALYSIS FOR MEAT SAMPLES

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ABSTRACT – Study of several factors that influence thermal analysis for meat samples

Differential scanning calorimetry is thermal analysis technique that detects and monitors thermally induced conformational transitions and phase transitions as a function of temperature. While denaturation of proteins display endotherms, aggregation of proteins manifest themselves as exotherms. In in this paper the influence of following factors was studied: influence of water evaporation, heating rate (°C/min), influence the mechanical process of grinding meat.

Keywords: DSC, myosin, sarcoplasmatic proteins, actin, denaturation

INTRODUCTION

Differential scanning calorimetry (DSC) is a powerfultechnique to characterise the energetics and mechanisms of temperature-induced conformational changes of biological macromolecules (KURGANOV ET. AL., 1997). This technique allows highlighting different temperatures at which the thermal denaturation of the major structural protein species in porcine muscle: myosin, sarcoplasmatic proteins, collagen and actin.

WENDLANDTG (1985) identified some 16 variables which influence the results from DSC experiments. Whilst many are attributable to the design of the equipment or to the inherent properties of the sample there remains a core of variables where the practitioner is able to exert some control (HAINES, 2000).

MATERIAL AND METHOD

Porcine longissimus dorsi were removed from the carcass at 1 day postmortem and stored into a hermetic package at 4^oC until analysis.

A TA differential scanning calorimeter (DSC), model SDT Q600, with computerassisted data acquisition and curve sensitivity analysis function was used for all studies.

Small pieces of meat, free from visible traces of fat and connective tissue, were used into alumina pans. At least 3 samples with 10 to 25 mg meat each weighed accurately to 0.001 mg by an electronic balance were used for each individual sample. The samples were scanned at 10°C/min or 5°C/min at 20 to 90°C under dry nitrogen purge of 30 mL/min.

For probe 5, the meat was finely minced and then dispersed in water in the ratio 1:10, using T 25 digital ULTRA-TURRAX (IKA[®] Werke GmbH & Co. KG) with S 25 N - 18G Dispersing element at 20000 rpm

Samples	Sample pan	Reference pan	Heating rate, °C/min
1	piece of meat	water	10
2	piece of meat	_	10
3	piece of meat	-	5
4	piece of meat + water	_	5
5	homogenized meat with water	_	5

 Table 1. Pans content and heating rate for each samples

Following factors were studied: influence of water evaporation, heating rate (°C/min), influence the mechanical process of grinding meat

RESULTS

Influence of water evaporation

If the water was put into the cup of reference in equal quantity of water in the sample was obtained termograma shown in *Figure 1*. Over 40° C sarcoplasmatics proteins begin to lose their solubility and begin the processes of denaturing which was achieved with energy absorption. (Cross et al., 1986; Sun, 2005). The transition displays at 60.8° C can be attribute to myosin denaturation. In other works, the denaturation temperature for myosin can be found between 54° C and 58° C (Martens and Vold, 1976; Wright, Leach, Wilding,1977). In this case, the slightly higher temperature, can be explained by the fact that cups were used unclosed hermetic.

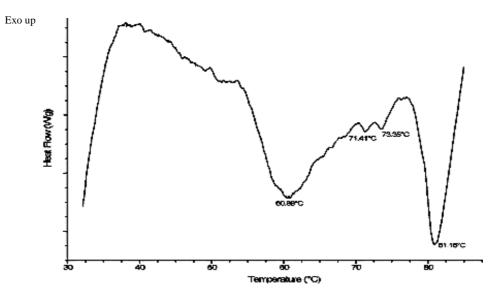


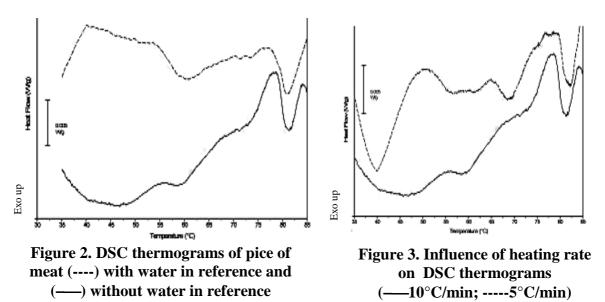
Figure 1. DSC thermograms of pice of meat with water in reference cup

As was expected, the transition which occurs between $65^{\circ}C$ and $67^{\circ}C$ assigned to collagen (Martens and Vold, 1976; Stabursvik and Martens, 1980) could not see clearly because meat was free from visible traces.

 $71,41^{\circ}$ C and $73,35^{\circ}$ C are the temperatures corresponding sarcoplasmatics proteins denaturation. Salvador et al. (2009) found the denaturation temperature corresponding to porcine hemoglobin concentrates Td = 77.8° C. Last transition at 81.16° C corresponds to actin denaturation and it occurred after its thermal aggregation. Levitsky et al. (2008) also analyse in vitro data on the heat-induced aggregation of actin, the process that normally accompanies actin thermal denaturation.

If the water was not added in the cups, the endothermic process was observed in the range from 40^{0} C to 47.5^{0} C. This may correspond to the fat melting process and temperature range is so large due higher heating rate.

Note that unlike the previous analysis, although it was the same meat used, in this case denaturation of myosin is observed at 58.36° C. The two peaks corresponding to temperatures 73.35° C and 71.41° C is not observed, the curve having only a change of the slope. Slope of this region has the lowest value (0,0003548W/g/°C) compared with the previous (0,0006462W/g/°C) and the subsequent regions (0,001416W/g/°C). For actin denaturation, however, similar results were obtained in both cases.



Influence of heating rate

Experimental results of thermal analysis are markedly affected by heating rate. In general, the effect of heating rate can be summarized as follows: a)- the heating rate influences the temperature distribution inside the sample; b)- when a chemical reaction, for example a thermal denaturation is measured by DSC, the initial temperature, peak temperature and final temperature shift to the high temperature side with increasing heating rate; c)- when the heating rate is high, the reaction takes place with greater speed in the higher temperature region. The reaction finishes within a narrow temperature interval, and on this account the derivative curves become sharper. (HATAKEYAMA AND ZHENHAI, 1999).

In this paper influence of heating rate in the analysis of a piece of meat is shown in *Figure 3*. For 5° C/min heating rate one of the most significant benefits from high

heating rate is the resultant increase in sensitivity (GABBOTT P. (2007). As seen, at a lower heating rate is observed two peaks corresponding to temperatures of 56.42°C and 61.19°C, while in the case of higher heating rate is obtained only one peak at 58.69°C, when myosin denaturation occur.

Three endothermic transitions have also been reported in DSC thermograms of rabbit (WRIGHT AND WILDING,1984) and fish myosin (TOGASHI ET AL., 2002). Multiple transitions of myosin imply structural changes in discrete regions of the myosin molecule, namely the hinge, head, and rod regions (WRIGHT AND WILDING,1984). Chicken breast myosin suspended in 0.6 M NaCl at pH 6.5 exhibits four cooperative endothermic transitions (WANG AND SMITH, 1994) and pork and chicken meats were very similar in behaviour (FERNÁNDEZ ET AL., 2000)

Endothermic transition observed at $68,75^{\circ}$ C is obvious when the heating rate of 5° C/min, as in *Figure 1*., and corresponding sarcoplasmatic proteins (KAZEMI ET AL., 2009)

Actin denaturation is observed, that takes place at similar temperatures, the difference being smaller than 1°C.

The mechanical process of grinding meat

The results for finely minced meat and dispersed in water were compared with those obtained for a piece of meat over which water was added so that in both samples to have a similar ratio between meat and water. The two curves obtained are shown in *Figure 4*.

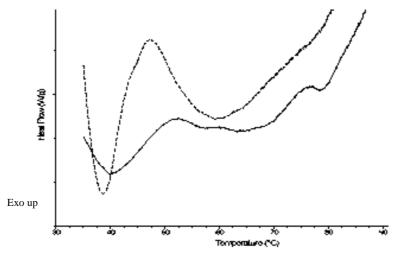


Figure 4. DSC thermograms of (----) finely minced meat and dispersed in water sample and (-----) piece of meat over which water was added sample

For ungrounded meat three transition states were obtained which correspond to 56.7°C, 63.2°C and 78.5°C temperatures, respectively for the distortion of myosin, sarcoplasmatic proteins and actin. These values are closer to those of literature (ZHU ET AL., 2004; KAZEMI, 2009) and on the other hand note that, denaturation enthalpy are lower than in the above cases. After fine mincing of meat and its dispersion, the resulting curve shows a transition with a peak at 59,3°C and a change in slope at 78.8°C.

CONCLUSIONS

It is very important to ensure a good contact between the sample and crucible and between the crucible and the thermocouple or other measuring sensor. Evaporation of water influences the enthalpy of protein denaturing, but the temperatures at which transitions occur can be pointed with computer-assisted data acquisition and analysis sensitivity function curve. Moreover, if necessary, water may be added in the cup of reference, in quantity equal to that of the sample.

The factor that most influences the thermograms, is the speed of heating. According to this peaks of the curve may appear/disappear and consequently the transitions can be more or less visible.

ACKNOWLEDGEMENTS

This work was co-financed from the European Social Fund through Sectoral Operational Programme Human Resources Development 2007-2013, project number POSDRU/89/1.5/S/63258 "Postdoctoral school for zootechnical biodiversity and food biotehnology based on the eco-economy and the bio-economy required by eco-sangenesis"

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