THERMAL PROPERTIES OF MUSCLE PROTEIN AND MEAT QUALITY

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INTRODUCTION

Differential scanning calorimetry (DSC) can detect the heat denaturation of a protein in complex protein systems as an endothermic peak in its thermogram. Moreover, DSC technique has the advantage that it can be used to observe thermal changes and denaturation of muscle proteins in meat. Changes in exothermic and endothermic peaks on DSC thermogram of muscle during storage could be conducted to determine whether such changes could be uniquely associated with quality of meat such as freshness, functionality and adulteration. The purpose of this study is to determine freshness and quality of pork by DSC and compared with ATP-related compounds, K-value which are determined with NMR and HPLC.

Materials and Methods

M. longissimus dorsi between 5th and 11th rib of pork was obtained immediately after slaughtering from the local meat market and chilled/stored at -2° and 25°C (ambient temperature) for 0, 12, 24, 36, 48 and 96 hours for analyses.
The pH value was measured with an HI 8424 Microcomputer pH-meter (HANNA Instrument, Italy). Bacterial number was measured by the method of FDA (1975). Volatile basic nitrogen content was determined by the modified method of AOAC (1984).
NMR-spectra were measured by using the method of Vogel and Lundberg (1985) NMR-spectrometer 7.05 Tesla, Varian Instrument Ltd., USA. DSC was performed on a ULVac DSC-7000 (Sinku-Riko, Japan) equipped with a thermal analyzer. Samples (15-20mg) were weighed in aluminum pans (No. 201-53090) and then sealed. The scanning temperature as 25° - 99°C at a heating rate of 10°C/min. Triplicate samples were analyzed. A reference containing 12-13mg distilled water was used. The instrument was temperature calibrated using Indium. Each DSC analysis was repeated three times. After DSC analysis, the sample pans were punctured and the dry weight of the samples determined after drying at 105°C overnight. The enthalpy of denaturation of muscle proteins was also collected. Procedure of high peroformance liquid chromatography (HPLC) analysis of ATP-related compounds were modified according to the methods of Boyle et al. (1991) and Ryder (1985). The HPLC analysis was performed on Model L-6200. (Hitachi Co., Japan). K-value (%) was calculated by ratio of inosine (HxR) + hypoxanthine (Hx) to the amount of ATP-related compounds using the data obtained from the result of HPLC analysis (Saito et al., 1959). And SDS-PAGE electrophoretic behavior of muscle proteins was carried out by the method of Laemmli et al. (1970).

Results and Discussion

pH value of pork stored at -2°C dropped slowlier than pork stored at 25°C and then remained stable (see Fig. 1). Bacterial counts of pork stored at -2°C increased up to the first 24 hours of storage then decreased, but the pork stored at 25°C increased up to a maximum level after 24 hours of storage, then kept constantly (see Fig. 2).
VBN value of the sample stored at 25°C increased after 24 hr storage with the storage time increased, but the sample stored at -2°C remained stable (see Fig. 3).
The changes of ATP-related compounds for the pork stored at -2°C and 25°C were shown in Fig. 4 and 5, respectively. ATP concentration of pork stored at 25°C depleted faster than the sample stored at -2°C, and reached a minimum level after 12 hours of storage, but the sample stored at -2°C reached a minimum level after 24 hour of storage. Both treatments had the same trends in the changes of ATP concentration. AMP

level eleavated up to a maximum level after 12 hour storage, then dropped. However, AMP concentration of the sample stored at 25°C was lower than stored at -2°C.

IMP concentration reached the peak after 12 hours of storage for the pork stored at 25°C and then dropped, but the pork stored at -2°C reached the peak after 24 hours of storage then kept stable.

Inosine concentration of the pork stored at 25°C increased sharply up to 12 hours of storage then decreased. But the pork stored at -2°C reached the peak after 36 hours of storage and then decreased gradually. Hypoxanthine concentration of the sample stored at 25°C to a maximum level after 60 hours of storage, but the sample stored at -2°C kept lowest level stably.

Fig. 6 showed the K-values which were calculated by the data obtained from the ATP-related compounds concentration of the samples. The result showed K-value in the sample stored at 25°C increased with the storage time increased, but the sample stored at -2°C remained stable. This suggested that meat stored at lower temperature could obtain longer shelf-life. However, meat stored at higher temperature would spoil faster.

DSC thermal property analysis was shown in Fig. 7 and 8. It was found the exothermic peak and two endothermic peaks appeared on the thermogram of the pork stored at -2°C 1 hour postmortem (Fig.7). However exothermic peak disappeared from the thermogram of the pork after 12 hours of storage, and three edothermic peaks for myosin ($Tmax_1$), sarcoplasmic proteins ($Tmax_2$) and actin ($Tmax_3$) appeared on the thermogram. It was noted that Tmax of trannsition in myosin, sarcoplasmic proteins and actin had a gradually shift to the lower temperature with increasing postmortem time. However, the trend in Tmax of transition for the sample stored at 25°C was same as the sample stored at -2°C for 48 hour storage, exceptly, it only appeared two peaks after 96 hours storage which were very difficulty to be identified as a given protein (fig. 8). K-value for the samples stored at 25°C after 24 hours of storage was above 44% which is near spoilage but the sample stored at -2°C still kept at very lower level (0.70 0.7. These changes can be used to indicate the index of freshness in fish (Kimura et al., 1987; Negishi et al., 1992), but DSC thermogram changes are very difficult to express the freshness of meat.

Fig, 9 showed NMR-spectra for ATP-related compounds in the pork stored at -2°C and 25°C during storage. ATP, CrP, sugar-phosphate and inorganic phosphate of normal pork can be detected on NMR-spectra. Their chemical shifts were -5.1, -10.2, and -19.2 ppm for r, a and beta ATP, -2.3 ppm for CrP, 2.5ppm for inorganic phosphate and 3.5-4.5ppm for sugar-phosphate, respectively. Sugar-phosphate and Pi two peaks presented on NMR-spectra for the pork sample stored at -2°C during storage for 96 hours, but sugar-phosphate disappeared from the NMR-spectra of the sample stored at 25°C after 48 hours of storage. Whether this change can be used as an indicate of freshness of meat, it is worth our consideration.

Fig. 10. showed the changes of electrophoretogram of myofibrillar proteins in the pork stored at -2°C and 25°C. The bands of myofibrillar proteins in the sample stored at -2°C remained stable during storage, but 30K dalton component appeared on the electrophoretogram for all the pork samples both stored at -2°C and 25°C during storage. The concentration of 30K dalton component increased with increasing postmortem time. Band of actin became weak after 96 hr postmortem. This change may cause the Tmax of transition of actin disappeared from the DSC thermogram after 96 hrs postmortem. (see Fig. 8)

All the attributes as indicators for freshness of meat can not indicate the actual degree of freshness, it may indicate a part of meat quality or freshness. Several studies indicated DSC can detect the changes of physico-chemical properties of muscle postmortem. Wright et al. (1977) reported that the presence of a exothermal peak in DSC thermogram of pre-rigor rabbit at 54°C which disappeared with the onset of rigor mortis. We also found the same result occuring in the porcine muscle within 4hr postmortem In our preuier work. (Chen et al., 1992). Watanabe et al. (1992) reported changes of K-value was associated with the storage temperature and time. The results of DSC thermogram was in agreement with the previer studies of Wright et al. (1977) and Stabursvik and Martens (1980). Park and Lanier (1988) also found a large exothermic peak near 50°C, observed soon after sacrificing was the most striking difference between thermogram of pre-rigor muscle compared to that of post-rigor muscle. As a result, DSC thermogram can be employed for determination of meat quality and some degree of freshness of meat within a short period, but it can not be used as an indicator of spoilage of meat. Whenever, the properties mentioned above are used to express quality of meat --- freshness, it has to compare to sensory detection.

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