The Influence of Freezing and Further Defrosting on the Process of Heat Denaturation of Myofibrillar Proteins of PSE Pork

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SUMMARY: In the present work structural change of myofibrillar proteins and their water holding ability in PSE (pH_1=5.4) and normal pork (pH_1=6.15) in process of heat denaturation and on the background of meat ageing from 2-3 to 96 hours post mortem, were studies, using methods of native protein fluorescence. Investigations were conducted on chilled ahd defrosted meat.

It was shown that contrary to traditional pork with normal pH-value freezing and further defrosting positively influence PSE meat , stabilizing structure of myofibrillar proteins and increasing water-binding ability during ageing time, exceeding 24 hours.

INTRODUCTION: It is known that PSE meat is characterized by partial protein denaturation already during the first hour post mortem (Honikel K.O. et al.,1986; Schwagele et al., 1989; Loper-Bofe C. et al.,1989; Barton-Gade P.A.,1980). This is particularly related to enzymes of sarcoplasma, however, partial changes also seem to take place in myofibrillar proteins. This is confirmed by our research in the field of fluorescent study of heat denaturation process of myofibrillar proteins of PSE pork where we showed that development of coagulation of its proteins takes place at lower temperatures (63-65°C), compared to normal meat(Oreshkin E.F. et al.,1989). Considering this fact, it could be supposed that behaviour of myofibrillar proteins of PSE meat after freezing and further defrosting would be significantly different from the assumed point of view. It also seemed interesting to follow ability of PSE pork proteins to retain water under conditions of freezing and defrosting. For the abovementioned reasons, the aim of the present study was to investigate behaviour of myofibrillar proteins of PSE meat during heat denaturation after freezing and defrosting, and also to characterize water-binding capacity(WBC) of PSE pork under these contions.

MATERIALS AND METHODS: Research was done on M.longissimus dorsi during its ageing from 2-3 to 96 hours post mortem. Freezing was performed at t=-18°C every day post mortem. Defrostation took place in the same sequence during 24 hours.

The work was done at 3-5 fold repeatability. Deviations constituted $< 0.5\%$. pH-value pf PSE pork was pH_1=5.4 and of normal - pH_1=6.15. Myofibrillar proteins were studied by method of native protein fleuorescence (Oreshkin E.F. et al.,1985); WBC in process of heat treatment was defined by thermogravimetric method (Permyakov E.A.,1986).

RESULTS AND DISCUSSION: Analysis maximum position of fluorescence spectrum of PSE pork (chilled and defrosted at room temperature) and its shift during heating(Fig.1), the state of myofibrillar proteins can be characterized as follows:1. At room temperature their structure was more dense as in traditional meat with normal pH-value, maximum position of fluorescence spectrum at 330-391nm confirmed this fact; at the same time normal meat maximum is situated at 332 nm (Oreshkin et al.,1985).

2. During heat treatment of PSE meat, graph of temperature dependence of maximum position of fluorescence spectrum shows certain "two-humped" appearance, i.e. between two peaks of a long-wave shift, coagulational short-wave shift is observed, its maximum being apparent at 70°C. This phenomenon is characteristic (not always) of meat with normal pH-value, usually after curing. The change of quantum outlet of fluorescence evidence that the main peak of denaturational loosening of protein structure in the first, "low-temperature" one (60-70°C); its position changes depending on quality and conditions of meat treatment. At the same time, position of the second high-temperature denaturation peak is usually constant by temperature (80°C), and degree of its expressibility in a great extent depends on ageing time. The nature of emergence of the second high-temperature denaturation peak during heat denaturation of myofibrillar meat proteins demands further investigation. However, it is necessary to note, that on curves of temperature dependence of maximum position of fluorescence plotted for pure actomyosin in solution , the second high-temperature denaturation peak is seen, however, solution of myosin doesn't show it(Permyakov E.A. et al.,1986). 3. After freezing and defrosting of PSE pork regularities of change of maximum fruorescence spectrum during heating are preserved (Fig.1),i.e. position of both denaturation peaks is the same at the same temperature as for chilled meat. The only exceotion is meat, defrosted 24 hours post mortem, in this case both denaturation peaks merge, forming only one peak with average value by temperature. However, the main difference of heat denaturation of defrosted PSE pork is lower, than in chilled meat, degree

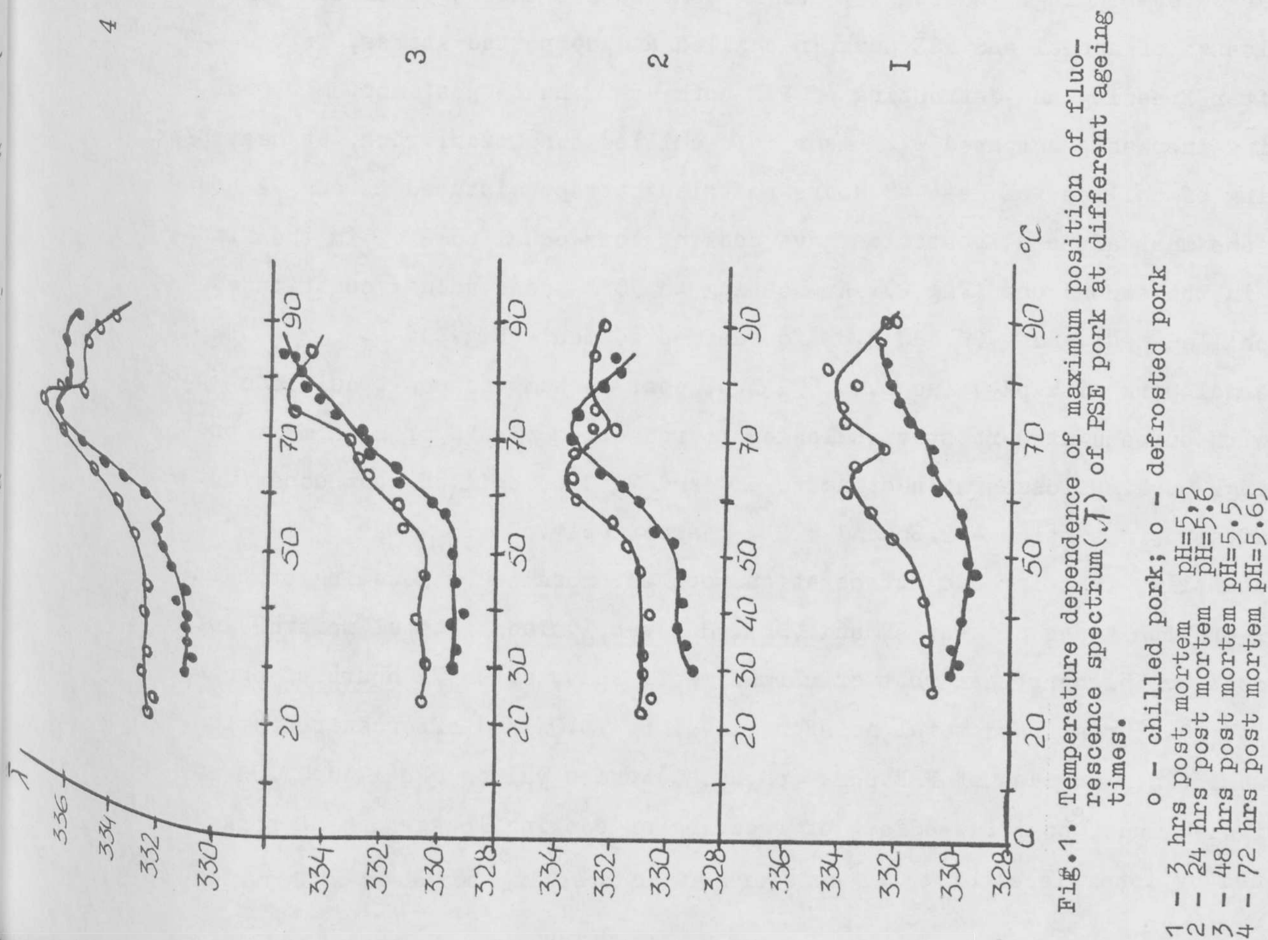

Fig.1. Temperature dependence of maximum position of fluorescence spectrum(λ) of PSE pork at different ageing times.

o – chilled pork; • – defrosted pork

1 – 3 hrs post mortem pH=5.5
2 – 24 hrs post mortem pH=5.6
3 – 48 hrs post mortem pH=5.5
4 – 72 hrs post mortem pH=5.65

of loosening of myofibrillar proteins structure ($\Delta\bar{\lambda}$) in the whole temperature range(on 2-3 min.). Along with that, shifts, bearing character of denaturation and coagulation, are poorly accomplished and indistinctly differentiated which allows to suppose simultaneous running of both processes in different parts of sarcomeres at the same temperature.

By all mentioned traits defrosted PSE meat differed from the earlier studied normal meat, which, during heat denaturation after defrosting, showed increase in loosening of structure as compared to chilled meat, and besides, it showed distinct coagulation shift in 70-80°C temperature range (Oreshkin E.F. et al.,1986).

However, the noted aggregated state of structure of PSE pork myofibrillar proteins, stipulated by freezing and subsequent defrosting, did not inhibit development of denaturation loosening in process of heating, which is illustrated by change of maximum position of fluorescence spectrum (Fig.1), i.e. the process of structural aggregation is on the stage, far from the stage of heat denaturation. Thus, it is evident, that freezing and defrostation cause partial destruction of proteins conformation in PSE pork, compared with proteins of chilled meat. This can probably explain greater degree of aggregation of their structure. However, difference with chilled meat is significantly lower, than in case of normal meat, exhibiting weakining of system of structural bonds in defrosted meat at the first stage of heating, and along with that widening of structure, resulting in abrupt coagulation at further rise of temperature. Further stage of research included determining of WBC of PSE and normal meat. We supposed that more aggregated and probably more orderly state of myofibrillar structure of defrosted PSE meat must create conditions for better water-binding. And really, during comparative thermogravimetric analysis of cooking losses of normal and PSE pork in chilled and defrosted states, it was shown that:

1. After freezing and defrosting of PSE pork 48-72 hours post mortem WBC of meat during heating increases compared with meat in a chilled condition. Thus, at heating to 60°C cooking of chilled PSE meat 48 hours postslaughter consistuted 3% and 72 hours - 2.2%. The same meat after defrostation gave cooking loss equal to 2.1% in the first case and 1.2% in the second one (Fig.2). At heating to 70°C these values constituted 6.0% and 4.8% for chilled meat and 4.0% and 2.8% for defrosted meat (Fig.3).
2. Normal pork with pH-value 6.15 (72 hrs. post mortem) showed traditional picture. Freezing with subsequent defrostation leaded to reduction of WBC of such meat proteins during heating. Cooking losses at heating to 60° and 70°C of chilled meat constituted 1% and 2.[illegible] and after defrostation - 2.6% and 4.2%, respectively.

Naturally, freezing and defrostation were accompanied by lowering of total moisture level of both types of meat (N and PSE). However, value of total moisture of defrosted PSE meat was higher, than that of normal meat. Thus, after 72 hours of ageing chilled pork with pH_1=6.15 possessed total moisture level of 75.0% and after defrostation - 73.8%. At the same time, in case of PSE pork (pH_1=5.40) these values equalled 77.2% and 75.1%, respectively. Thus, positive effect of freezing on cooking losses of PSE pork cannot be explained by lower level of total moisture after freezing compared with normal meat. Pro-

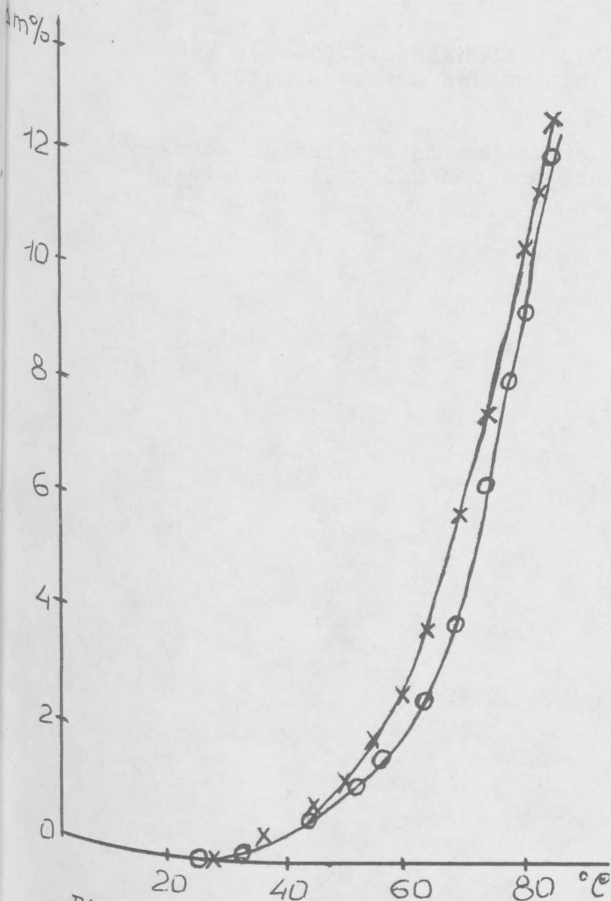

Fig.2. Graph of temperature dependence of PSE pork (pH=5.4) cooking losses after 48 hours of ageing.

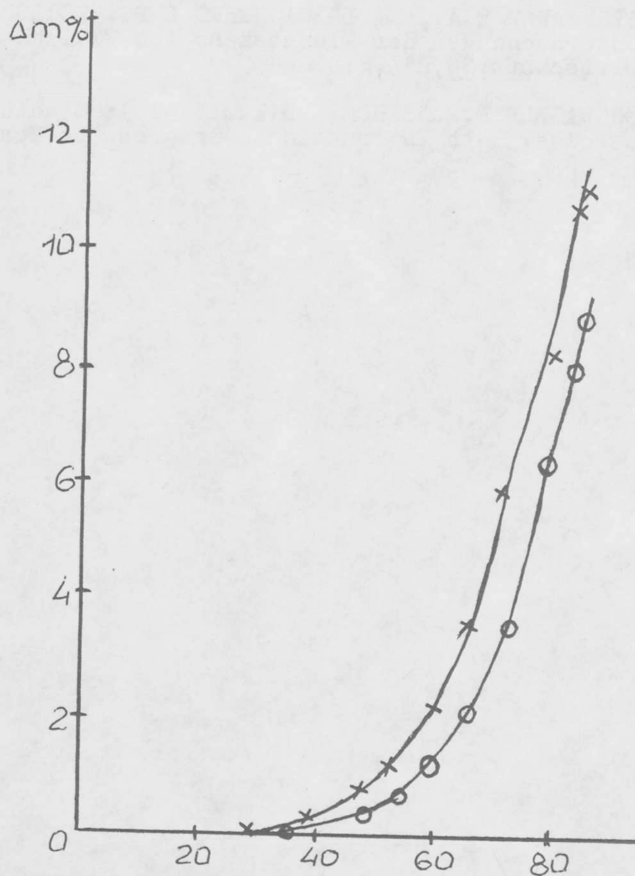

Fig.3. Graph of temperature dependence of PSE pork(pH=5.4) cooking losses after 72 hours of ageing.

bably, this effect is stipulated by specific state of myofibrillar proteins structure in PSE meat after defrostation, which as it was shown above, now possesses higher degree of aggregation than before freezing. In this case good conditions are created for stabilization of hydrophobically-bound netweork of immobilized water.

CONCLUSIONS: Thus as a result of conducted research, it became evident that contrary to traditional meat with normal pH-value, freezing and further defrostation positively influence PSE meat, stabilizing structure of its myofibrillar proteins and increasing WBC at ageing time exceeding 24 hours.

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