

Structure formation of cured products prepared from fresh warm beef in case of using multi-component protein-containing brines

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The extension of the range of raw materials used for processing and further intensification of technological processes in cured meats manufacture continue to be one of the urgent tasks of the meat industry. In the USSR purposeful studies into cured meats preparation from pre-rigor beef have been recently carried out /1/. It has been found that their quality can be improved by means of electromassaging (EM) during curing, i.e. by means of treating precured pre-rigor muscle with impulse electric current /2-5/. To develop cured meat products with pre-set functional and biological properties, of high quality, with improved yields, it is suggested that various components - protein-containing ones included - be incorporated into curing brines /6/. Of special importance is the necessity of their uniform distribution throughout muscle. The purpose of this investigation was to study the pattern of the structure development of cured products prepared from pre-rigor beef by using multi-component protein-containing brines (MPB) under EM and cyclic mechanical treatment (CMT). To provide more rational utilization of meat and dairy by-products, blood plasma and skim milk were incorporated into the curing brine in addition to salt, sugar, sodium nitrite and spices. To increase the protein level and to improve the processing properties of MPB, an isolated soy protein was added to it. When formulating qualitatively and quantitatively MPB, we took into account medico-biological requirements to the amino acid balance of cured meat products. Our calculations provided for solving a system of linear equations of the material balance for amino acids /6/. Experiments were made with the l. dorsi muscle of pre-rigor ($38 \pm 1^\circ\text{C}$) carcasses of 1-2-year-old beef animals (Finish Grade II). For this the muscle was dissected within 45-50 minutes after electrostunning. MPB was injected at the points $2 \cdot 10^{-2}\text{m}$ apart from each other; the injection level was 20% of the raw meat weight. The injected muscle was electromassaged for $4.8 \cdot 10^{-2}\text{s}$ with alternating current (220 V, 50 Hz, impulse duration 0.6s, impulse intervals 0.4s). CMT was made for $28.8 \cdot 10^{-2}\text{s}$ (massaging for $1.8 \cdot 10^{-2}\text{s}$, pause for $1.8 \cdot 10^{-2}\text{s}$) at the angular rotation speed of 4.2 rad/s and the load factor of 0.5. During massaging 5% of MPB of the meat weight were added. The temperature of $33 \pm 2^\circ\text{C}$ needed for maintaining a high level of the diffusive-osmotic redistribution of curing components was ensured in the drum using a heat-transfer agent in its steel jacket. Tests were repeated five times. Samples for microstructural analyses were taken directly in the zone of brine injection and at the distance of 1 cm from each other (successively). Samples were analysed immediately after injection, after electromassaging and mechanical treatment. Samples to be tested, sized $2 \times 2 \times 1\text{ cm}$, were fixed in a 20% solution of neutral formalin for 48 hr. From every fixed sample a $1 \times 1 \times 0.3\text{ cm}$ pieces were cut out, their muscle fibers being arranged transversally. They were washed in tap water for 24 hr; dehydrated in alcohols of increasing concentrations and in alcohol-ester; impregnated with celloidine; stuck onto wooden blocks; placed into a desiccator for compacting in chloroform vapours and held in a 70° alcohol. 7-10 μm sections were prepared with a sliding microtome. They were stained with the Erlich hematoxiline followed with eosine staining. Stained sections were treated with alcohols, carbolxylene and xylene and placed under polystyrene onto slides. The sections were microscoped and photographed in the microscope MBI-15 with a 108-fold magnification. The muscle fibers of pre-rigor meat on the transversal sections are closely attached to each other, muscle bundles are interleaved with connective tissue of varying thickness. Where MPB is injected, muscle bundles are greatly moved apart. The spaces between the bundles are most often filled with an aggregated coarsely-dispersed protein phase of the brine, which consists of particles of a different configuration (Fig.1). Identification of the discovered particles as the ones contained in the brine, demonstrated that it was a microscopically suspended fraction of the soy protein dissolved in the brine. By their configuration, they are ring- or sickle-like multilayered formations, similar to a muscle fiber in diameter. Around these aggregates large spaces are observed filled with the dissolved phase of the injected brine. At the spots immediately adjacent to the injection point, pulled-apart fiber bundles are discovered together with single soy protein particles introduced with the curing brine. In more removed muscle layers no coarsely-dispersed protein particles are registered. At the same time, there are spaces between groups of muscle fibers, or single muscle fibers, it indicating the penetration of the soluble MPB phase into these areas (Fig.2). The examination of pre-rigor muscles after EM indicated that there were considerable changes around the injection points, viz., the dimensions of coarsely-dispersed soy protein particles were greatly diminished, and a fine-grained protein mass, filling the adjacent connective tissue spaces, was formed (Fig.3). From here the latter mass penetrated deeper parts of the muscle.

The CMT of the muscle just after EM caused a further more uniform distribution of the injected brine, and at the same time a release of sarcoplasmic and a part of myofibrillar proteins from muscle fibers. After the treatment, the muscle fibers (in most cases, they were swollen, without clear boundaries, of a rounded shape) are interconnected by means of the fine-grained protein mass.

In many areas, usually located around the MPB injection points, single soy protein particles are observed (Fig.4).

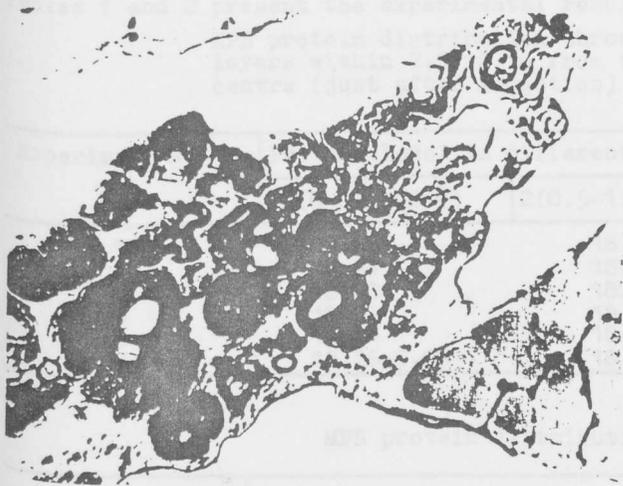


Fig.1. Microstructure of a part of the muscle around the MPB injection point. Accumulation of the coarsely-dispersed protein phase of the brine

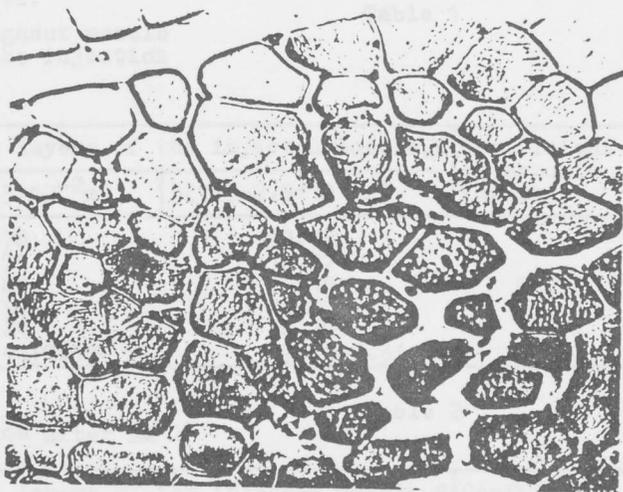


Fig 2. Microstructure of the muscle areas remoted from the injection point. Brine accumulation in the connective tissue interlayers and between single fibers.

The analysis of the experimental results obtained makes it obvious that during MPB injection its coarsely-grained phase is accumulated in the injection zone. This can be attributed to the fact that the dimensions of the non-dissolved particles are about equal to the diameter of a muscle fiber and that they cannot penetrate muscle bundles. However, taking into account that interlayers dimensions do not usually exceed those of muscle fibers, it is clear that their penetration at the moment of brine injection is impeded. Muscle functions as a filter allowing only fine-dispersed and soluble fractions of the brine (salt, sugar, nitrite) from the injection zone to deeper layers. EM causes breakage



Fig.3. Microstructure of the muscle in the MPB injection zone after EM. Protein particles dispersion with the formation of a fine-grained protein mass.

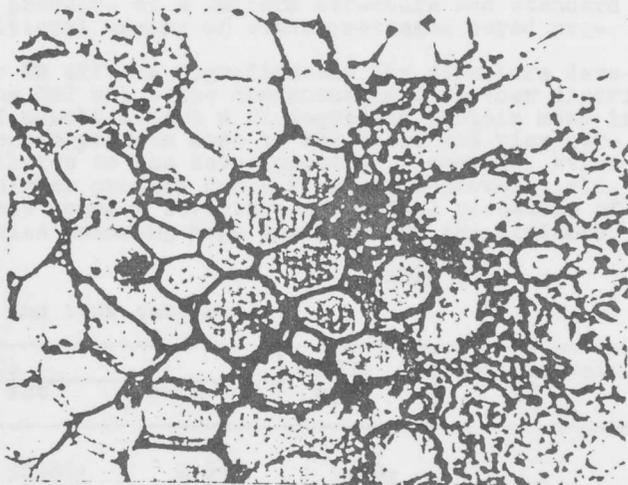


Fig.4. Microstructure of a muscle area after EM and CMT. Swollen muscle fibers with the in-between distributed MPB and the muscle proteins released due to the above treatment.

of the coarsely-dispersed fraction followed with the formation of a fine-grained mass which is forced into deeper muscle layers due to muscle fibers contraction. The data obtained agree well with the estimated rate and extent of the re-distribution of protein components in pre-rigor meat during EM. These parameters were calculated using the coefficient of the uneven distribution of brine proteins throughout muscle layers as follows:

$$K = \frac{A_2 - A_1}{2}$$

where K is coefficient of nonuniformity, A3 is protein level at the centre of brine injection (%), A1 is protein level in the layer adjacent to the injection centre (%). Tables 1 and 2 present the experimental results.

Table 1

MPB protein distribution throughout muscle layers within $2.5 \cdot 10^{-2}$ m from the injection centre (just after injection)

Experiment N°	Protein level in different layers of the injected muscle, %			Coefficient of nonuniformity
	1 ($0-0.5 \cdot 10^{-2}$)	2 ($0.5-1.5 \cdot 10^{-2}$ m)	3 ($1.5-2.5 \cdot 10^{-2}$ m)	
1	18.38	18.78	19.98	0.75
2	18.24	18.70	19.97	
3	18.40	18.89	19.15	
4	18.05	18.50	18.80	
5	18.24	18.24	18.90	
M±	18.26±0.06	18.42±0.15	19.76±0.33	

Table 2

MPB protein distribution after EM

Experiment N°	Protein level in different layers of the injected muscle, %			Coefficient of nonuniformity
	1 ($0-0.5 \cdot 10^{-2}$ m)	2 ($0.5-1.5 \cdot 10^{-2}$ m)	3 ($1.5-2.5 \cdot 10^{-2}$ m)	
1	19.53	20.20	20.02	0.25
2	19.47	20.00	19.89	
3	19.65	20.30	20.10	
4	19.35	20.10	19.95	
5	19.50	20.17	20.05	
M±	19.50±0.05	20.15±0.04	20.00±0.04	

Note. Protein contents in muscle tissue before injection on the average constitutes $20.86 \pm 0.12\%$

The above data indicate that pre-rigor beef electromassaged for $4.8 \cdot 10^2$ s accelerates brine protein re-distribution by 3 times. Our previous experiments and the present micro-structural examination of pre-rigor muscle after EM, however, show that only electric treatment is clearly insufficient to ensure products of a uniform structure and standard quality. The latter is provided with an additional ageing of electromassaged cured pre-rigor meat for 24-48 hr /5/.

Our present work demonstrated that CMT after EM effects significantly the structure development process of cured meats. First, during CMT all brine components are further distributed. Second, due to partial destruction of muscle fibers a fine-grained protein mass is formed, which - together with the fine-dispersed protein mass of the brine and tiny particles of non-dissolved soy protein - contributes to the development of a compact, structurally bound product characterized with a higher protein content and an improved yield. Therefore, CMT following EM allows to complete complex structuro-mechanical processes of the development of typical raw cured properties ensuring high qualities of the finished product (Tables 3-5).

Table 3

Physico-chemical indices of control and test samples

Curing method	Content, %, of:				NaNO ₃ mg %	Caloric value, kJ
	water	protein	fat	salt		
Traditional holding in cure for $259.2 \cdot 10^2$ s (control)	64.21± 0.03	21.16± 0.04	10.81± 0.04	2.79± 0.02	1.38± 0.02	820.613
MPB injection, EM and CMT (test)	62.00± 0.02	23.76± 0.02	10.58± 0.03	2.69± 0.03	1.40± 0.02	904.349

As a result of the studies carried out it was established that EM application in the manufacture of cured products from pre-rigor meat ensured a re-distribution of the protein component of MPB in the product due to the destruction of the non-dissolved coarsely-dispersed phase and its movement together with fine-dispersed particles from injection zones to deeper layers.

CMT of cured, pre-rigor, electromassaged meat contributes to the development of a uniform tender texture which ensures finished products of a high quality without extra holding in cure. The developed technology of cured products from pre-rigor beef made it possible to reduce stored meat shrinkage losses by 2.5%, to improve finished product yields, on the average, by 5-6% as compared to the operating standards, to increase the protein level by

2.5%, on the average, to improve the amino acid profile and to considerably reduce energy and labour consumption.

Table 4

Structuro-mechanical properties of control and test samples

Curing method	Plasticity, $\cdot 10^{-4} \text{m}^2$	Shear stress, $\cdot 10^{-5} \text{Pa}$	Indentor insertion depth, $\cdot 10^{-2} \text{m}$
Traditional holding in cure for $259.2 \cdot 10^3 \text{s}$ (control)	2.29 ± 0.02	4.83 ± 0.04	14.9 ± 0.02
MPB injection, EM and CMT (test)	2.83 ± 0.02	3.81 ± 0.04	17.5 ± 0.03

Table 5

Organoleptical scores for control and test samples

Curing method	Appearance	Colour	Aroma	Taste	Consistency	Juiciness	Total score
Traditional holding in cure for $259.2 \cdot 10^3 \text{s}$ (control)	7.39 ± 0.03	7.41 ± 0.04	7.18 ± 0.04	7.08 ± 0.03	6.83 ± 0.03	6.73 ± 0.03	7.01 ± 0.01
MPB injection, EM and CMT (test)	7.51 ± 0.03	7.59 ± 0.03	7.81 ± 0.02	7.71 ± 0.02	7.83 ± 0.02	7.59 ± 0.02	7.67 ± 0.02

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