ULTRASTRUCTURAL AND BIOCHEMICAL CHANGES IN MUSCULAR TISSUE OF CHILLED BEEF AS RELATED TO STORAGE PROCEDURES

In recent years great attention has been given to the investigations on meat storage in the atmosphere of an inert gas /1-3/. Several authors /4-6/ pointed out that during storage of meat in a gaseous nitrogen at 0°C the surface colour of the meat musculat tissue is better retained, the growth of aerobic psychrotrophic bacteria is inhibited and thus the storage life of beef is increased.

Of interest were the investigations into the effect of the nitrogen atmosphere on changes in ultrastructure, water-holding capacity, solubility and fractional composition of meat proteins during atorage at 0°C.

In our investigations chilled beef (Longissimus dorsi) was stored for 20 days at 0°C in the atmosphere containing 99.8% (±0.2%) gaseous nitrogen. The control meat samples were stored in the air at the same temperature.

The quality of chilled beef was evaluated by ultrastructural and biochemical changes in the meat prior to its storage and after 2, 6, 9, 12, 15 and 20 days of storage.

For electron-microscopic examination samples were fixed with 1% solution of OSO_4 according to Colfield and poured into methacrylate. The sections were made with ultramicrotome «КБ-4801А, contrasted with uranyl acetate and phosphato-tungstic acid, and then on grids - by uranyl acetate. They were examined under an electron microscope of УЭМБ-100 Б type at 5,000-40,000 instrumental magnification.

The fractional composition of proteins was determined by means of electrophoresis in starch gel /7/ in tris-borate buffer /8/, modified by the authors. Myofibrillar proteins were extracted from the muscular tissue with distilled water and 8M urea /9, 10/.

For the determination of the solubility of sarcoplasmic proteins 0.03 M K-phosphate buffer was used, and of that of myofibrillar proteins - 0.01 M K-phosphate buffer + 1.1 M KI/11/. Nitro-

gen content in the protein extracts was determined by means of the Kjeldahl semimicromethod.

Meat water-holding capacity was determined by a press-method /12-13/.

Electron-microscopic examinations of meat prior to its storage demonstrated that muscular fibres in chilled muscles were moved apart, myofibrils were shortened, protofibrils were located in parallel rows and adhered closely to each other. There were no I-discs in myofibrils, and the cross-striation represented the alternation of contrastly outlined narrow Z-strips and packed A-discs. On individual parts of muscular fibres Z-plates were poorly distinguished, with the result of indistinct differentiation of myofibrils into sarcomeres. There were sarcosomes in the spaces between myofibrils, their cristas being preserved and the matrix clarified.

After two-day storage of meat in the air the myofibrils were moved apart and had rectilinear outlines with narrow Z-plates distinguishable in them. As for the sarcosomes, the cristas were destroyed and the lipids were split off the structures (Fig. 1).

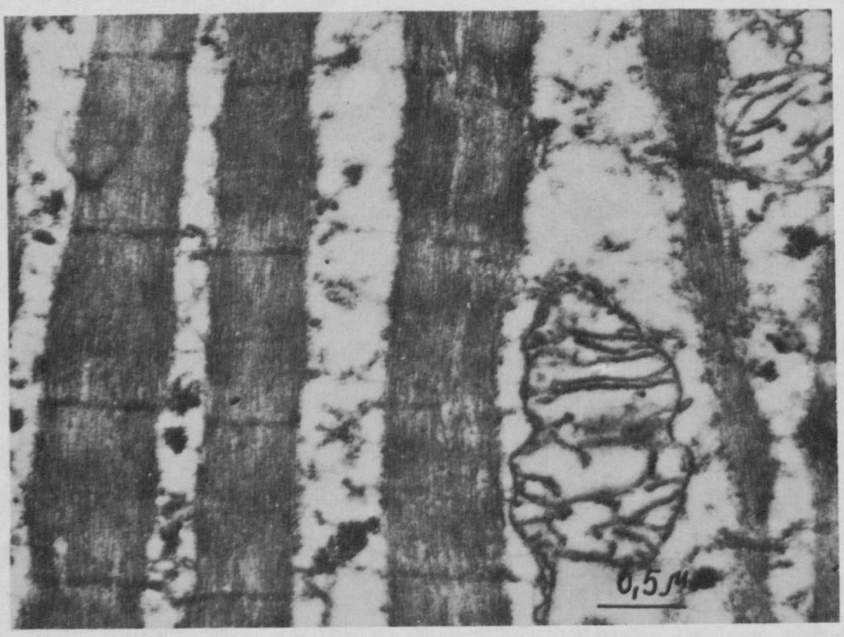

Fig. 1. Electronogram of a part of a muscular fibre of Longissimus dorsi after 2 days of storage in the air at 0°C

During subsequent storage in the air the myofibrils renained essentially rectilinear or became slightly garland-like. I-discs as a rule were not reconstituted. Starting from the 2nd-6th days the myofibrils fragmentation in Z-plates was observed which increased during subsequent storage of the meat. Thus, by the 12th day of storage, along with the local destruction of individual myofibrils, one could reveal transversal fragmentation of the whole groups of them (Fig. 2).

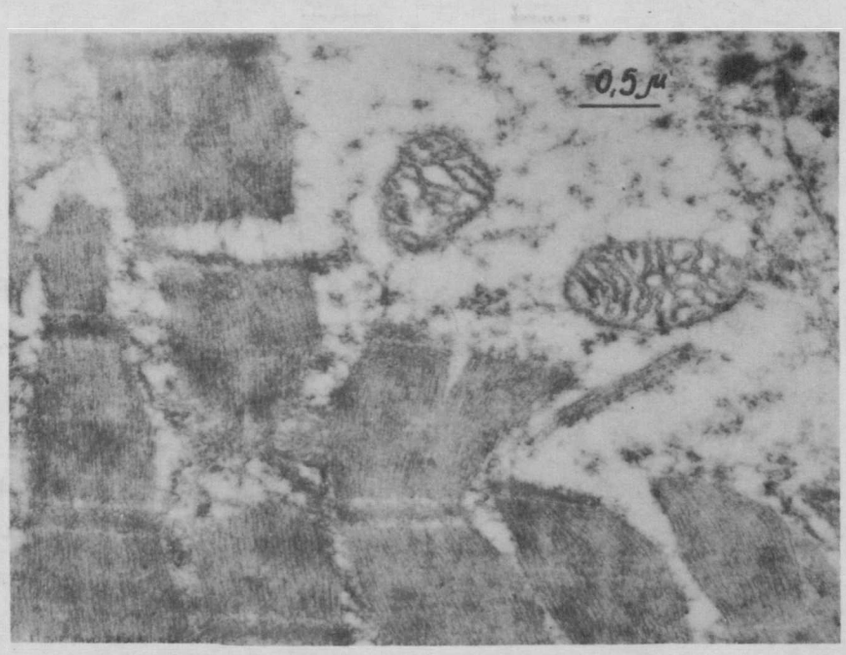

Fig. 2. A part of a muscular fibre of Longissimus dorsi after 12 days of storage in the air at 0°C. Fragmentation of myofibrils

By the 20th day of meat storage in the air, in addition to the increasing fragmentation of myofibrils, their local decomposition was noted, as well as swelling and decomposition of myosin threads. The destruction processes in the sarcoplasm were increasing with storage. These were expressed in the decomposition of sarcosomes, of the sarcoplasmic reticulum and in the acceleration of lipophanerosis.

During meat ageing in a gaseous nitrogen even by the 2nd day of storage myofibrils were considerably thickened. They became garland-like and adhered closely to each other (Fig. 3).

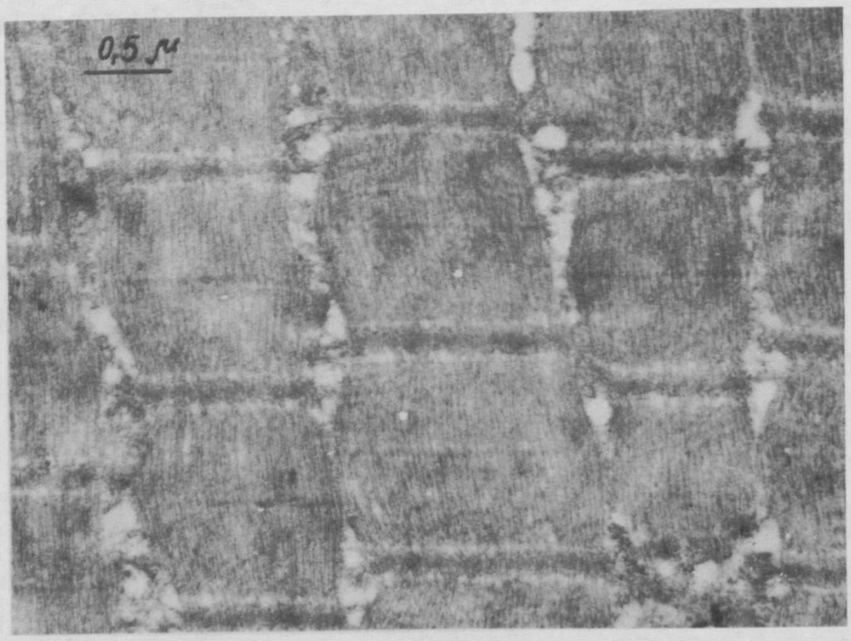

Fig. 3. Electrongram of a part of a muscular fibre of Longissimus dorsi after 2 days of storage in a gaseous nitrogen. Swelling of myofibrils.

Thickened Z-plates were distinctly seen everywhere and somewhere reconstituted I-discs were found. Swollen sarcosomes and enlarged canals of sarcoplasmic reticulum were observed between myofibrils.

The degree of myofibrillar structure swelling increased during subsequent storage in a gaseous nitrogen. By the 12th day of storage in muscular fibres here and there a completely blended mass of myofibrils was observed, where strongly swollen sarcosomes with broken cristas were revealed. In these parts protofibrils were curved, sometimes swollen and loose, there was no good order characteristic of the initial meat sample (fig. 4).

By 12-15th days of meat storage in a gaseous nitrogen, along with strong swelling of myofibrilar structures, destruction of sarcosomes and sarcoplasmic reticulum, fragmentation and local break-down of myofibrils were revealed.

The data given in the Table show that during storage in the air the water holding capacity of muscular tissue and the solubility of sarcoplasmic and myofibrillar proteins reach their minimum and maximum values on 2nd and 3d days, respectively. These values decreased during the subsequent storage.

Fig. 4. A part of a muscular fibre of Longissimus dorsi after 12 days of storage in a gaseous nitrogen at 0°C. Blending of a number of strongly swollen myofibrils.

During meat storage in a gaseous nitrogen, beginning from its second day, water-holding capacity and protein solubility increased and reached the maximum by the 12th day. By the end of the storage (15–20 days) one could observe greater water-holding capacity and protein solubility of the meat samples stored in a gaseous nitrogen.

Thus, the dymamics of water-holding capacity and protein solubility during meat storage in the air and in a gaseous nitrogen were different.

As for nitrogen-containing nonprotein substances, a slighily greater accumulation was observed during meat storage in the air (see the Table). It is, apparently connected with a quicker increase in the microbial load under these conditions.

Electrophoretic investigations showed that the sarcoplasmic proteins of chilled meat were a mixture of 27 fractions, and the myofibrillar proteins – of 10 fractions (Fig. 5, 6).

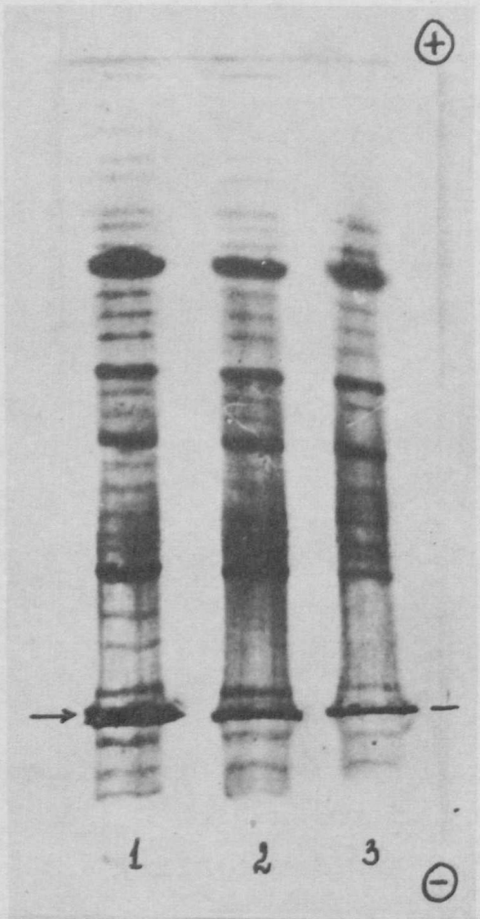

Fig. 5. Electrophoresis of meat sarcoplasmic proteins in starch gel:
1 - initial meat (after chilling); 2 - meat stored in nitrogen for 20 days at 0°C; 3 - meat stored in the air during 20 days at 0°C

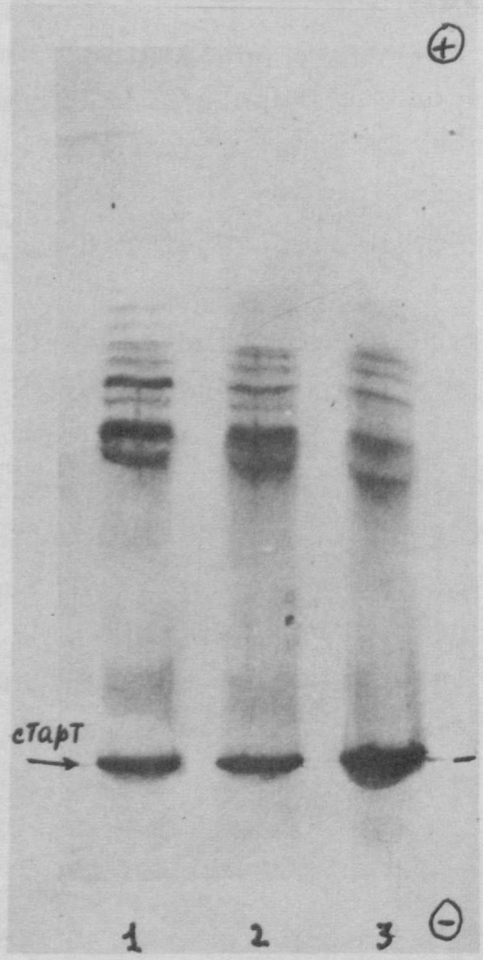

Fig. 6. Electrophoresis of meat myofibrillar proteins in starch gel:
1 - initial meat (after chilling); 2 - meat stored in nitrogen during 20 days at 0°C; 3 - meat stored in the air during 20 days at 0°C

After meat storage in a gaseous nitrogen we could find 23 fractions of sarcoplasmic proteins and 7 of myofibrillar ones. 18 sarcoplasmic and 6 myofibrillar fractions were contained in the meat stored in the air. It should be mentioned that the protein fractions in stored meat were pronounced more poorly than in the initial meat irrespective of the storage procedure. The samples stored in the air contained a smaller number and amount of protein fractions.

Ultrastructural and biochemical changes during beef storage in a gaseous nitrogen and in the air at 0°C were different. In the first case myofibrillar structures, were strongly swollen, this being followed by a considerable increase in meat water-holding capacity and a less intensive development of destructive processes.

A difference in the indices during meat storage in nitrogen is explained by oxygen absence /14, 15/. Therefore, beef storage in a gaseous nitrogen as compared to storage in the air makes it possible to keep better the native properties of meat.

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Table

Water-holding capacity and meat protein solubility during storage in a gaseous nitrogen and in the air (the data of 4 experiments)

Storage time, days	Water-holding capacity, % to muscular tissue		Sarcoplasmic proteins[x]		Myofibrillar proteins[x]		Nonprotein substances[x]	
	Air	Nitrogen	Air	Nitrogen	Air	Nitrogen	Air	Nitrogen
Prior to storage	58.7(0.2)[xx]		23.9(0.7)		51.6(0.6)		9.7(0.4)	
2	57.0(0.3)	62.0(0.1)	20.8(0.6)	24.9(0.3)	45.0(0.9)	52.0(0.4)	8.2(0.4)	9.7(0.6)
6	60.0(0.2)	63.2(0.3)	24.7(0.3)	25.7(0.4)	47.7(0.6)	52.0(0.5)	9.7(0.3)	9.7(0.3)
9	62.3(0.4)	63.5(0.4)	26.6(0.2)	26.0(0.4)	49.9(0.6)	52.4(0.9)	10.1(0.3)	9.9(0.4)
12	61.0(0.5)	66.0(0.4)	25.2(0.7)	27.5(0.9)	49.1(0.5)	52.6(0.7)	11.1(0.5)	9.9(0.6)
15	59.0(0.2)	66.0(0.1)	24.0(0.1)	26.0(0.2)	47.3(0.4)	52.5(0.6)	12.4(0.4)	10.3(0.5)
20	57.6(0.1)	65.7(0.1)	23.5(0.9)	25.1(0.3)	46.8(0.7)	50.2(0.6)	12.0(0.4)	11.3(0.2)

x) % to the total nitrogen

xx) the mean value and a standard deviation