MORPHOLOGICAL ASPECTS OF LYZOSOME STUDY AT ELECTRO-MECHANICAL TREATMENT OF BEEF MUSCLE

L.S.KUDRYASHOV, L.V.GORSHKOVA, T.P.BUSLAJEVA* AND A.S.BOLSHAKOV**
*The Kemerovo Technological Institute of Food Industries, Kemerovo, USSR
**The Moscow Institute of Applied Biotechnology, Moscow, USSR

SUMMARY: Determination of lyzosomal enzymes activity with the help of electrone microscope on the level of single cell organelles allowed to show the dynamics of beef lyzosomes change as influenced by technological factors. Results of ultracytochemical tests support earlier obtained data on biochemical and hystological analysis testifying to an increase of lyzosomal proteinases-cathepsins activity under the influence of electrical and mechanical treatments of muscle tissue. Experiments showed that hot meat electrostimulation is accompanied by disturbance of lyzosomal cover integrity and vacuole contents output into sarcoplasma. Electromassaging of cured muscle tissue leads to more noticeable destructive changes of lyzosomes. Electromassaging, followed by mechanical treatment is accompanied by, as it is seen from obtained data, significant destruction of lyzosomal membranes and enzymes output into sarcoplasma.

INTRODUCTION: In previously described investigations (Kudryashov L.S. et al.,1988) activity of lyzosomal proteinases (Cathepsin D and acid phosphatase) of pork muscle were studied using the methods of biochemistry and hystochemistry. Muscle was cured under electrical and mechanical treatment. Such treatment activates tissue enzymes and thus allows to intensify meat ageing and curing. In order to determine an important role of lyzosomes in post-mortem proteolytic changes of animal tissues it is necessary to identify various components of lyzosomal system on the level of cell organelles that is possible at electron microscope use.

MATERIALS AND METHODS: Tests were made on beef L.dorsi removed in 45-60 min. post mortem. Control samples were selected from hot muscle from which pieces of 10 x 10 x 10mm were extracted for testing under electron microscope. Test samples for right side were treated as the following: electrostimulation, brine injection and mechanical treatment under vacuum; samples for left side muscle were brine injected, treated with electric current (electromassaging) and then in a vacuum-mixer. Experiment procedure is detaily described in the work of Kudryashov et al.(1988). After each type of treatment samples for ultracytochemical analysis were selected. Test material was fixed consequently in 3% solution of glutaraldehyde prepared on phosphate buffer at pH 7.2-7.3, 2% solution of osmium according to Palade G.E. (1952) on phosphate buffer by Millonig G. (1962) at pH 7.4. Dehydrated samples were included into araldyte epoxyde resins.

Ultrathin slices (0.4-0.5nm) were prepared using ultramicrotone IKB-3 and contrasted by lead citrate according to Verable et al. (1965). Obtained slices were analysed and photographed using transmission electron microscope IEM-100B at magnification of 24.000 and 32.000. In total there were 240 electronogrammes.

RESULTS AND DISCUSSION: With the help of the current work on morphological data, taking into account biochemical and hystochemical studies, we tried to give more information on the mechanism of lyzosomal enzymes activity at post-mortem changes and during technological treatment. Test data (Fig.1) show that in hot non-treated meat lyzosomes are presented as small electron-dense bubbles of round form located along muscle fiber by single clusters. Membrane fully surrounds endocytose vacuole. Hot meat electrostimulation is accompanied by disturbance of lyzosomal cover integrity (Fig.2). At the same time it is seen how vacuole contents transfer to sarcoplasma and a liberated space is being filled by cell material. Brine injection and electromassaging of meat results in lyzosomal membranes destruction and substance output into surroundings (Fig.3). In the centre one can see a secondary lyzosome where endocytose digestion of cell components takes place; it is being grasped by a primary lyzosome as the result of it integrity destruction. Lyzosomes changes at electromassaging and further mechanical treatment are given in Fig.4. Data of electron-microscopic studies testify to significant destructions of lyzosomal membranes and enzymes output into sarcoplasma. A great number of small primary and secondary lyzosome is found in sarcoplasma. There is noticeable destruction of muscle fibers myofibrillar structures.

CONCLUSIONS: Based on the studies it is possible to stay t that use of intensifying factors at meat ageing and curing leads to a partial destruction of lyzosomal membranes and liberation of cathepsins. Comparing data obtained by electron microscopy with the results of biochemical (Gorshkova et al., 1988) and hystochemical investigation (Kudryashov et al., 1988) allowed to demonstrate the consequence of activity display of beef lyzosomal proteolytic enzymes.

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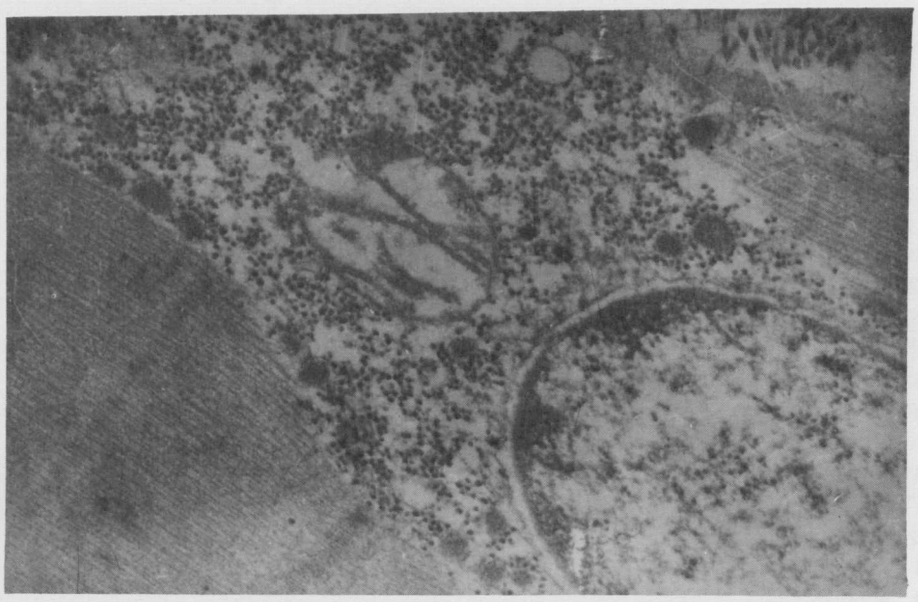

Fig. 1. Electronogram of hot beef muscle section (x40.000)

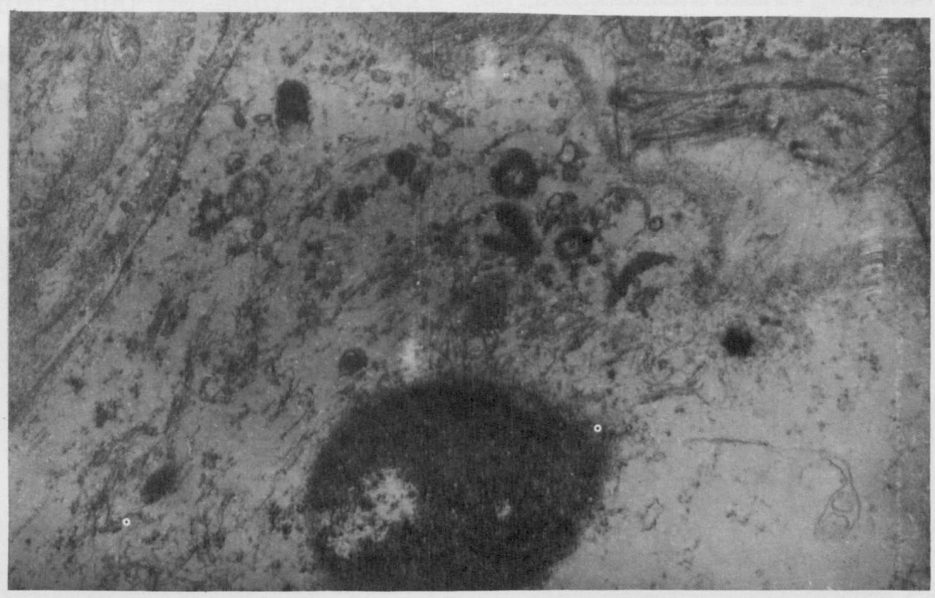

Fig. 2. Electronogram of electrostimulated beef muscle section (x40.000)

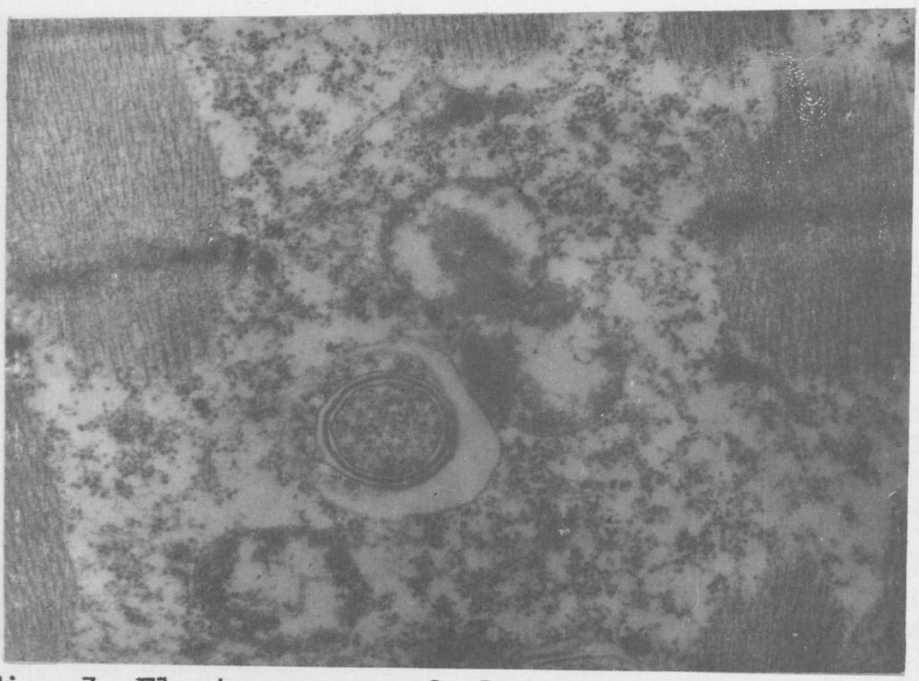

Fig. 3. Electronogram of electromassaged beef muscle section (x40.000)

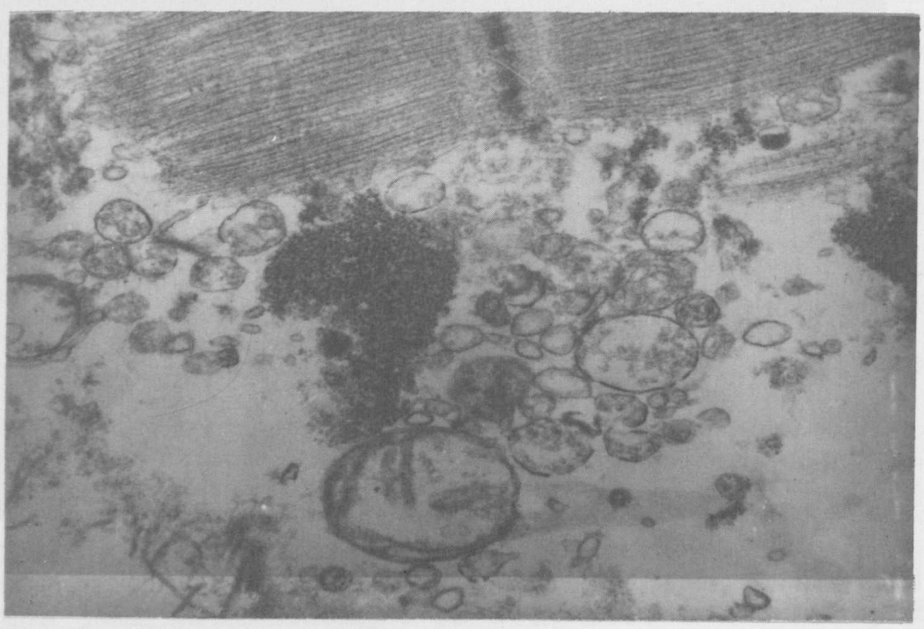

Fig. 4. Electronogram of electromassaged and mechanically treated beef muscle section(x40.000)