

## Electron Microscopic Studies of Mutton Treated with Microbial Protease Mesenterine 11-11

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### Introduction

The microstructural changes occurring in the muscular tissue as affected by the enzymes of microbial origin protease 15, rosim P-11, rosim A-4, NT proteolytic ferment, hydrolase D, hydrolase TR, suptilopeptidase E-30 and P-90, telrsin and rimo Solovet, V. I., 1966; Velinov, P. D., 1972; Skalinsky, E.I. and Belousov A.A. 1978. The enzymes of microbial origin degrade to a great extent the protein structures of muscular fibres and exert a slight influence on the fibrous elements of the connective tissue. They cause no apparent changes in the structure of collagen but they affect the mucopolysaccharides of the main substance which results in relaxation of the connective tissue layers (Skalinsky E.I. and Belousov A.A., 1978). The microstructural changes occurring in mutton as affected by the microbial protease Mesenterine 11-11 have been described by S. Singha et al., 1981). The objective of the present work was to investigate the effect of the microbial protease Mesenterine 11-11 on the ultra-structure of mutton.

### Material and Methods

The studies were carried out using mutton muscles longissimus dorsi. The test samples were injected with Mesenterine 11-11 0.35% in 2% salt solution while the controls were treated with 2% salt solution in the ratio of 10% to their weight. The experiments were conducted with muscles taken immediately after the slaughtering of animals (1st group) and with muscles subjected to cooling for 48 hours (IInd group). The enzyme-treated samples and the controls of the first group were sterilized for 2 hours on the 48th and on the 120th hour after injecting while these of the second group were processed on the 120th hour. The materials for the electron microscopic studies were taken after sterilization. After fixation in 5% glutar aldehyde for 1-2 hours, postfixation with osmium tetroxide for 1-2 hours, dehydration by an ascending series of alcohols and passing over propylene oxide the samples were introduced in durcupan. Ultra-thin sections made by the ultra-microtome LKB-III and contrasted with uranyl acetate and lead citrate were examined using the electron microscope TESLA BS-613 at 80 kV.

### Results and Discussion

Ultra-structural changes in mutton treated with enzyme solution immediately after slaughtering of animals.  
The ultra-structural changes in the muscular fibres of controls sterilized 48 hours after treatment with salt solution (electron microscopic photograph 1) showed that the myofibrils had lost their fibrillar character and the myosin protofibrils converted in a homogeneous mass as a result of the thermal denaturation. The Z-lines were strongly fragmented while irrespective of this the separate sarcomers were outlined. Traces of the H-zones were slightly observed. The electron microscopic picture of the corresponding test samples (electron microscopic photograph 2) revealed that

destructive changes occurred in the isotropic and the anisotropic discs under the influence of the microbial protease Mesenterine 11-11. The modified sections were characterized by the formation of a granular mass and by the complete lysis of separate fragments of myofibrils.

The ultra-structural changes in the muscular fibres of the controls sterilized 120 hours after the treatment with salt solution (electron microscopic photograph 3) resembled to some extent those described in the electron microscopic photograph 1. However, the strongly fragmented Z-lines being located in the middle of the considerably larger bright bands appeared on the places of the isotropic disks. Few sarcomers revealed strongly expressed thermal destructive changes in the protofibrils. The H- zones and the M-lines were better marked. The electron microscopic picture of the muscular fibres of test samples (electron microscopic photographs 4 and 5) indicated that the longer storage time of meat treated with enzymes was characterized by more significant changes in the myofibrils. The enzyme-affected fragments were either converted in granular mass or completely lysed. The sarcoplasmatic proteins were entirely lysed.

Ultra-structural changes in cooled mutton treated with enzyme solution 48 hours after slaughtering of animals.

The muscular fibres of controls sterilized 120 hours after application of the salt solution (electron microscopic photograph 6) revealed the disappearance of the fibrillar structure of myofibrils, the conversion of myosin protofibrils in homogeneous mass and the changes in the I-disks. It is worth noting that some sarcomers obtained a tubby shape due to the changes occurred. The Z-lines were broken. The electron microscopic picture of the muscular fibres of test samples (electron microscopic photographs 7 and 8) showed that the myofibrils were torn and granularly degraded.

under the influence of the microbial protease Mesenterine 11-11. It is seen on some spots that the enzymic action has started in the isotropic disks and spread to the anisotropic disks. In the studies on the action of the enzyme terizin on the muscular fibres E.I. Skalinsky and A.A. Belousov (1978) established that the proteolysis caused the destruction of the Z-lines and passed through the actin protofibrils to the interior of the A-disks of the sarcomers.

### Conclusions

The microbial protease Mesenterine 11-11 showed proteolytic activity on the myofibrillar and the sarcoplasmatic proteins. Mesenterine 11-11 produced destructive modifications and granular degradation of actin and myosin protofibrils to their complete lysis.

The destructive changes and the granular degradation of the myofibrils were strongly marked in the treatment of mutton with Mesenterine 11-11 carried out immediately after slaughtering of animals as compared with mutton treated for 48 hours after cooling.

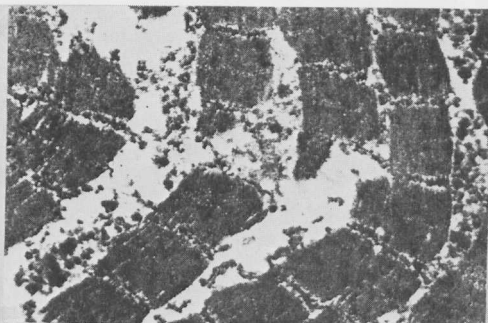
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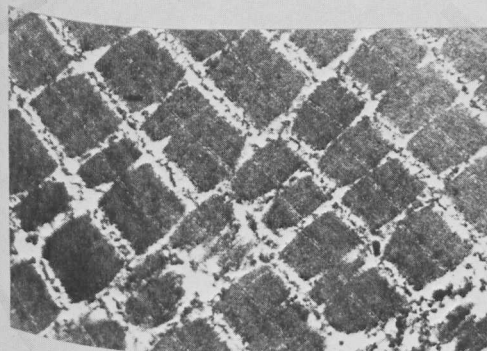
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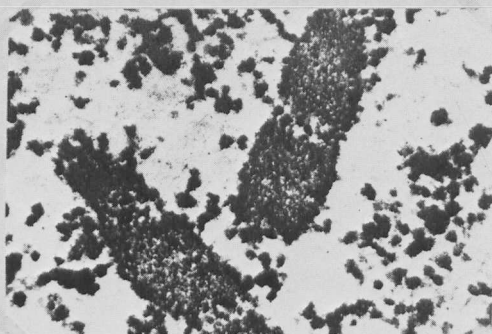
Electron microscopic photograph 1  
enlarged 14000 times



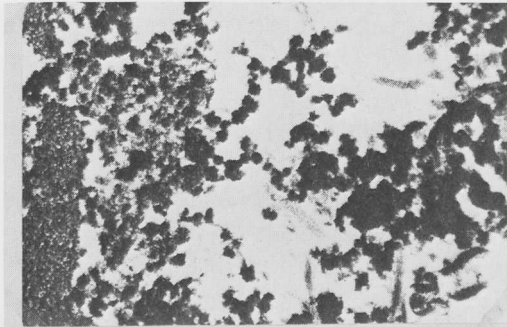
Electron microscopic photograph 2  
enlarged 10000 times



Electron microscopic photograph 3  
enlarged 10000 times



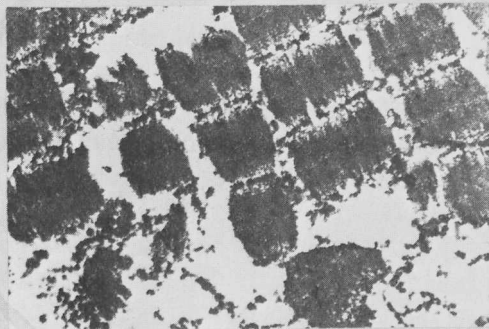
Electron microscopic photograph 4  
enlarged 14000 times



Electron microscopic photograph 5  
enlarged 14000 times



Electron microscopic photograph 6  
enlarged 10000 times



Electron microscopic photograph 7  
enlarged 10000 times



Electron microscopic photograph 8  
enlarged 14000 times