

## Changes in the texture, histological structure and degradation of myofibrillar protein of beef round meat during ordinary or vacuum cooking.

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### Introduction:

Recently, vacuum cooking has been used as one of the meat cooking methods at hotels and restaurants. Vacuum cooking, whereby meat is heated in a vacuum-sealed plastic bag, is useful in promoting the tenderness of meat and longer storage. The texture, especially tenderness, of cooked meat is one of its most important quality attributes. Heat-affected changes in meat components are connected with meat tenderness (Seidman, 1987). The SDS-PAGE and Western blotting analysis established that the myofibrillar protein of beef longissimus thoracis was coagulated at 73°C for 120 minutes. Then, when heated at 90°C, protein was degraded to lower molecular polypeptides (Fritz, 1992). The objective of this study was to investigate the change of muscle fiber structure, degradation of myofibrillar protein and tenderness during heating in vacuum-sealed plastic bags (in the range 65~100°C) in an attempt to find possible relationships.

### Methods

**Cooking procedures.** Beef round meat was obtained from a commercial source. After the fat and tendon (perimycium) were removed, the meat was cut into pieces weighting about 150g (2cm thick). For vacuum cooking, each piece of meat was put into a plastic bag (Asahi Kasei HN Type) and then sealed by a vacuum sealer (Sharp SQ 202). These samples were heated at 65, 75, 85 and 100°C for 60 minutes in the water bath.

**Histological observation of meat structure.** After cooking, samples were frozen in the freezer and cut into thin vertical sections. Observations were performed using an optical microscope (Nikon optiphot).

**Measurement of meat tenderness.** Cooked meat tenderness was measured by Warner-bratzler Meat Shear Model 2000.

**Sodium dodesyl sulfate polyacrylamide gel electrophoresis (SDS - PAGE).** Cooked meat samples were cut into small pieces and solubilized in a phosphate buffer (pH 7.0) containing 20% SDS and 4% mercaptoethanol. These solubilized samples were mixed with Wang's tracking dye and heated at 50°C for 20 minutes. SDS - PAGE was performed according to the methods of Leammli (1970). 60 µg of each protein sample were applied onto the gel. PAGE was carried out on the slab gel using SDS - Tris buffer system.

**Transfer condition and western blotting.** The proteins separated by SDS - PAGE were electrophoretically (5 hr at 90V) transferred from the gel to a nitrocellulose membrane (0.45 µm, Bio - Rad) in a Trans - Blot cell unit (Model 250/2.5, Bio - Rad) using 25mM Tris, 192mM glycine, and 10% (v/v) methanol buffer (pH 8.39). This transfer time was not enough for bands less than 170 kDa. After the blocking, the membrane was incubated in the first antibody, diluted 1:40, in the blocking solution at 4°C for 16 hours. The following treatment was made according to the usual procedure (Nakaya, 1994).

### Result & Discussions.

**Shear force value of cooked meat.** As shown in Fig.1, the shear value of cooked meat decreased as the temperature was raised after a certain point (75°C). The meat heated at 75°C revealed the highest value. Heated at 85°C, the toughness of meat when comparing ordinary to vacuum cooking showed no significant differences in shear value. However, heated at 100°C, vacuum-cooked meat was more tender than ordinary cooked meat.

**Structure of cooked meat.** Fig.2-a,b,c,d,e shows the structure of cooked meat heated at 75°C and 100°C for 60 minutes. At 75°C, muscle fiber shrank remarkably in ordinary cooking, whereas no shrinkage occurred in vacuum-cooked muscle fiber. At the highest temperature (100°C), cracks occurred in the vacuum-cooked sample. The first shrinkage at 75°C may have been due to the thermal denaturation of the muscle fiber. The observed minimum shear value in vacuum cooking at 100°C may be explained by the appearance of cracks in the muscle fiber structure.

**SDS -PAGE and Western blot analysis.** Fig.3 shows SDS-PAGE gel patterns of meat protein cooked in the range of 65 and 100°C. SDS-PAGE gel patterns comparing ordinary-cooked meat to vacuum-cooked meat showed that the myosin heavy chain was degraded at 65 and 75°C in both cooking methods, whereas at 85 and 100°C, the myosin band of vacuum-cooked meat was lower. Fig.4 shows the pattern of the degradation of myosin to lower molecular components using myosin antibodies after the protein was separated by tricin-SDS-PAGE. Fragments of 85, 58, and 48KDa were observed at 65°C cooking in both methods. This observation suggested that myofibrillar protein hydrolyzed by protease at this temperature. However, heated at 75°C and 85°C, these components were not observed, and 39 and 30KDa fragments increased. Heated at 100°C, 30KDa fragments disappeared. This result suggested that myofibrillar protein was degraded to lower molecular polypeptides during heating at 100°C for 60 minutes.

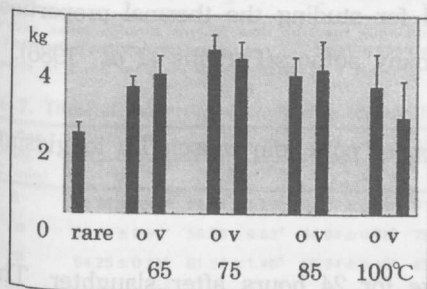


Fig.1 Changes of shear force value of cooked meat.

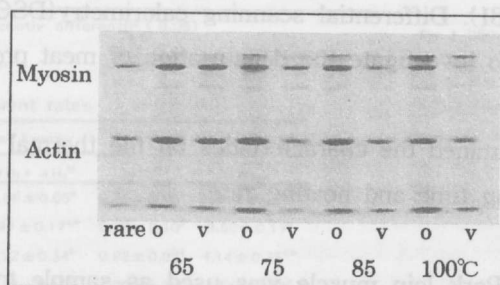


Fig.3 SDS-PAGE gel patterns of cooked meat.

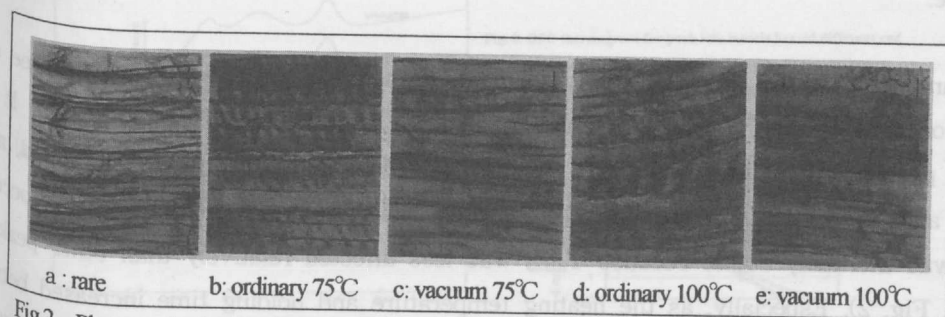


Fig.2 Photograph of opti-microscope (The scale bar indicates 100 μ m.)

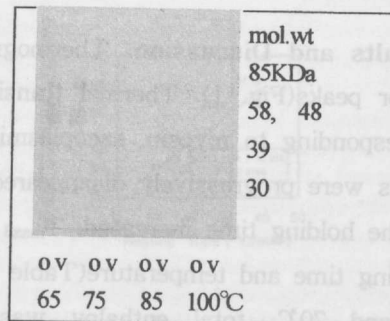


Fig.4 photograph of myosin antibody reaction

## Conclusion

The shear force value of meat heated at 75°C showed the highest toughness, compared with other temperatures, and some shrink and frizz were visible in the muscle fiber when observed by optical microscope (×100). Heated at 100°C, vacuum-cooked meat was obviously more tender than ordinary-cooked meat, and cracks were observed in the muscle fiber. The SDS - PAGE gel patterns of vacuum-cooked meat at 100°C for 60 minutes revealed that myosin was degraded. We analyzed the degraded components of myosin using the western blotting method with antibodies of myosin, and the low molecular components (30kDa) was increased by raising the temperature. Moreover, when vacuum-cooked at 100°C for 60 minutes, the band of 30kDa almost disappeared. This observation suggested that these components were degraded to lower molecular polypeptides.

## Literature

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