SURE EFFECTS ON THE TEXTURE, ULTRASTRUCTURE AND MYOFIBRILLAR PROTEIN OF BEEF SKELETAL MUSCLE

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Summary

hydrostatic pressures of 100 MPa to 300 MPa were applied to beef post-rigor muscle to investigate the
ciency of pressurization as a meat tenderizer.

he hardness measured by Rheometer decreased and the fragmentation of myofibrils increased with increasing
sure applied to the muscle. The degree of fragmentation reached to its maximal level after briefly exposing
in) post-rigor muscle to the highest pressure (300 MPa). Electron microscopic studies of the pressurized
le revealed that marked rupture of I-band and loss of M-line materials had progressed in the myofibrils
increasing applied pressure. However degradation of the Z-line in myofibrils that can be observed natural-
conditioned muscle was not apparent in the pressurized muscle. There was no significant difference in the
trophoretic pattern of myofibrillar protein among the control and pressurized samples in spite of the
ed change of ultrastructure.

rom the results, it is suggested that the application of a high hydrostatic pressure to post-rigor muscle
es tenderization of the meat in a different manner from that of conditioning.

Introduction

ntly, the application of high hydrostatic pressure of food processing has attracted much attention in Japan,
ese changes in the properties of food materials induced by pressurization proceed in a different manner from
of heat processing (HAYASHI,1989). Of all foods and food constituents, muscle and muscle protein are prob-
the most responsive to pressure as has been suggested by MACFARLANE (1985). Since MACFARLANE's observation
3) that a brief exposure of pre-rigor muscle to high pressure (100 MPa) for a few minutes at ambient temper-
e produced a marked drop in shear value, a new tenderization method for meat by high pressure treatment has
reported in a series of papers by MACFARLANE (1974,1985) and others (KENNICK et al.,1980;ELGASIM & KENNICK,
; RIFFERO & HOLMES,1983). However, there are few papers describing tenderizing effect of high pressure
ment on post-rigor muscle. BOUTON et al (1977) suggested that post-rigor muscle proved less amenable to
improvement unless long exposure at high temperature (50-60 ℃). was used. LOCKER & WILD (1984) also
rted that pressure-heat treatment tenderized post-rigor meat effectively only after considerable periods at
levated temperature. From the standpoint of the commercial application of high pressure, tenderization of
rigor meat is more important than that of pre-rigor meat.

his paper describes the effects of a high hydrostatic pressure treatment on the texture profile, fragmen-
on, ultrastructure and myofibrillar protein of post-rigor muscle to investigate the efficiency of pressuri-
on as a meat tenderizer.

Materials and Methods

surization of the muscle.

Meat was excised from the shoulder part of a beef carcass (Holstein) 4 days after slaughter and stored in

a freezer at -25 ℃. As required, it was tempered overnight in a cold room (2 ℃), and then cut into small pieces (50x50x30mm). Each piece was vacuum sealed in a polyethylene bag and transferred to a large polyethylene bag. The space between the bags was filled with cold water. Each bag was transferred to a pressure vessel, which was maintained at about 10 ℃ with water and crushed ice, and pressure applied at 100, 150, 200 and 300 MPa for 5min with a Cold Isostatic Press from Nippon Kokan Co., Ltd. After pressurization, the sample was taken out and immediately cooled in an ice box.

Texture profile.

Texture profile of the pressurized muscle was measured with a NRM-20002J Rheometer (Fudo Kogyo Co., Tokyo) using a conical plunger.

Preparation of myofibrils.

Myofibrils were prepared from the pressurized muscle by the method of BUSCH et al (1972). The preparation was suspended in 100 mM KCl and 1 mM NaN_3.

Fragmentation of myofibrils.

The degree of fragmentation was measured essentially according to the procedure described by TAKAHASHI et al (1967). The protein concentration of the myofibrils was adjusted to 0.25 mg/ml and observed under a phase-contrast microscope at a magnification of 1500. The degree of fragmentation of the myofibrils is expressed as the percentage of the number of fragments composed of 1-4 sarcomeres to the total number of myofibrils observed.

Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE).

Myofibrils sedimented by centrifugation at 5000xg for 10min were solubilized in 0.01 M sodium phosphate buffer (pH 7.0) containing 5% SDS and 1% 2-mercaptoethanol in boiling water for 30min, with subsequent centrifugation at 10,000xg for 15min. The clear supernatant was subjected to an SDS-PAGE analysis according to the procedure described by WEBER & OSBORN (1969). The electrophoresis was performed for 3.5 hr at 8mA per gel on 8cm gels containing 5% polyacrylamide (bisacrylamide-acrylamide, 1:37(w/w)).

Electron microscopy.

An electron microscopic observation of the myofibrils was carried out by the method described in our previous paper (SUZUKI et al, 1978). Specimens were examined with a JEM 100S electron microscope operated at 80 kV.

Results and Discussion

Effect on texture profile

The effects of high-pressure treatment on texture profile of the muscle are shown in Fig. 1, the results being representative of those obtained by repeated measurements for different position of the muscle. The hardness of the pressurized muscle decreased to 60, 20 and 10% of the control (untreated) at 100, 150 and 300 MPa, respectively. Whereas significant difference in the elasticity was not observed between the control and pressurized muscle. This result indicates that brief exposure of post-rigor muscle to high pressure induces the meat tenderization without heat treatment.

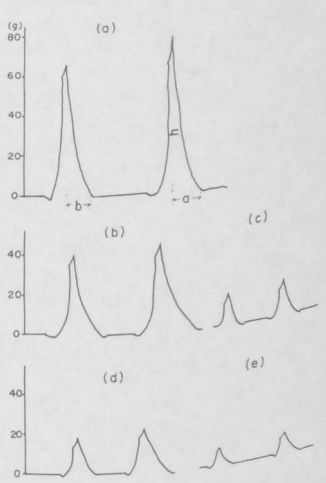

Fig. 1. Texture profile of pressurized bovine skeletal muscle. The muscle was pressurized at 0(a), 100(b), 150(c), 200(d) or 300(e) MPa at 10°C for 5min. h, hardness; b/a, elasticity.

on the fragmentation of myofibrils

ffect of high-pressure treatment on the degree of fragmentation is shown in 2. A sigmoidal relationship was obtained between the degree of fragmenta- and intensity of pressure applied to the muscle. The degree of fragmenta- which was less than 10% in the untreated muscle, was accelerated by pres- ation and reached over 30, 70, 80 and 90% at 100, 150, 200 and 300 MPa, res- ely. It is well-known that the myofibrils prepared by homogenizing condi- d muscle were shorter and composed of fewer sarcomeres than those prepared unconditioned muscle (TAKAHASHI et al,1967), and that breaks in myofibrils line were correlated with the increase in meat tenderness (TAKAHASHI et al, DAVEY & GILBERT,1967; FUKAZAWA & YASUI,1967; HENDERSON et al,1970; MOLLER ,1973). Therefore, myofibrillar fragmentation has been shown to be useful predicting meat tenderness (OLSON et al,1977; CALKINS & DAVIS,1980). As shown in Fig. 2, fragmentation of yofibrils increased with the increase of applied pressure to the muscle, and the degree of fragmentation ed its maximal level with brief pressurization as compared with that of myofibrils naturally occuring in tioned muscle. From the results of this fragmentation, a brief exposure of post-rigor muscle to high pres- seems to be useful for meat tenderization.

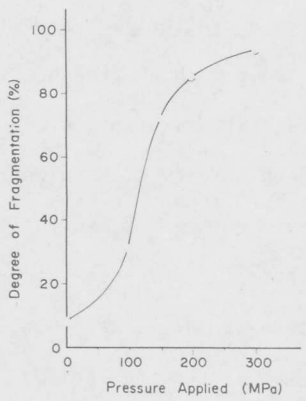

Fig. 2. Effect of pressurization on the degree of fragmentation in myofibrils prepared from pressurized bovine skeletal muscle.

t on the ultrastructure

structure of the myofibrils prepared from the pressurized muscle is shown in Fig. 3. In the myofibrils pre- from the muscle pressurized at 100 MPa, a contraction of the sarcomere was observed, and the difference ensity between the A-band and I-band became indistin- ishable as compared with the control. Marked rupture of filamentous structure of the I-band and a loss of the he materials were observed in myofibrils from the mus- pressurized at 150 MPa. The Z-line structure seemed to ut of register. In the myofibrils from the muscle pres- ed at 200 MPa, the structural continuity of the sarco- was almost completely lost, with broken A- and I-fil- t spread over the sarcomere. Complete loss of the M-line thickening of the Z-line, probably due to a collapse of I-filament, were also observed. Cleavage of the A-band ng to the many changes already mentioned was observed yofibrils from the muscle pressurized at 300 MPa. The th of the sarcomere initially contracted by pressuri- on at 100 MPa seemed to have gradually recovered with increase of pressure applied to the muscle, because of the increasing loss of structural continuity. As dy mentioned, fragmentation of the myofibrils during conditioning is derived from breakage of the myofibrils the Z-line, whereas the Z-line in the fragmented myofibrils prepared from pressurized muscle apparently re- ed intact. In spite of the short time (5min) and low temperature (about 10 ℃) of the pressure treatment ap- d to the post-rigor muscle, changes in the ultrastructure of the muscle shown in this paper were principally

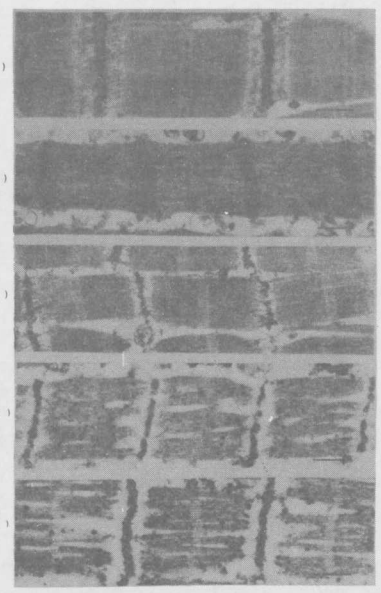

Fig. 3. Ultrastructure of myofibrils prepared from bovine skeletal muscle. Condition of pressurization was the same as those specified for Fig. 1. Each scale mark indicates 0.5 μm.

in accordance with those reported by MACFARLANE (1978,1985) and by LOCKER & WILD (1984).

Effect on myofibrillar protein

The effect of high-pressure treatment on the protein constituents of the myofibrils was investigated by SDS-PAGE, which is shown in Fig. 4. No significant difference in the electrophoretic pattern of myofibrillar protein was observed among the control (untreated) and pressurized muscle preparations in spite of the marked changes in the ultrastructure that was observed in the pressurized muscle. The appearance of 30,000 dalton protein, which was commonly observed in the myofibrils prepared from the conditioned muscle (PENNY,1980), was not observed in the myofibrils prepared from the pressurized muscle.

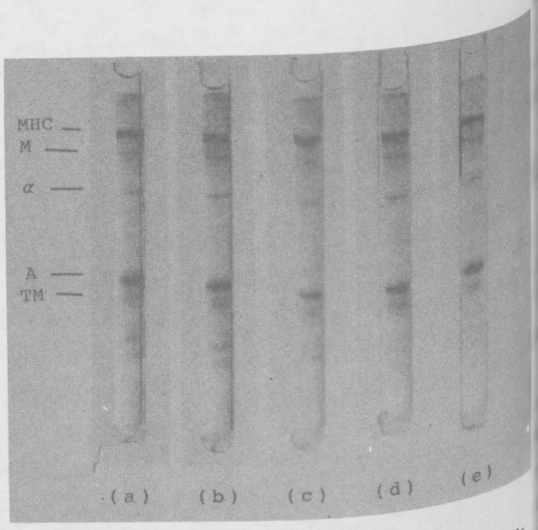

Fig. 4. SDS-PAGE patterns of myofibrillar protein prepared from pressurized bovine skeletal muscle. Myofibrillar prpteins were analyzed on 5% gels containing 0.1% SDS. Condition of pressurization was the same as those specified for Fig. 1. MHC, myosin heavy chain; M, M-protein; α, α-actinin; A, actin; TM, tropomyosin.

Conclusion

From the results obtained in this report, it is suggested that a high hydrostatic pressure-treatment of post-rigor muscle causes tenderization of the meat in a different manner from that of conditioning.

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