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High Hydrostatic Pressure Effects on the Texture of Meat and Meat Products

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ABSTRACT: High hydrostatic pressure (HHP) treatment can influence meat protein conformation and induce protein denaturation, aggregation, or gelation. The means whereby HHP treatment exerts effects on meat protein structure change are due to the rupture of noncovalent interactions within protein molecules, and to the subsequent re-formation of intra- and inter-molecular bonds within or among protein molecules. Depending upon the meat protein system, the pressure, the temperature, and the duration of the pressure treatment, meat can be either tenderized or toughened. Muscle texture variation induced by heat treatment is due to breakage of hydrogen bonds, whereas changes from high pressure treatment are due to the rupture of hydrophobic and electrostatic interactions. Pressure treatment has little effect on the toughness of connective tissue. Juiciness, springiness, and chewiness are increased upon HHP treatment. Prerigor HHP treatment tenderizes meat, whereas tenderizing effects of postrigor HHP treatment are only measureable if pressure and heat treatment are combined. The limitations and future applications of high pressure technology are also discussed.

Keywords: hydrostatic pressure, meat protein, temperature, texture

Introduction

The effect of high pressure processing on food systems was first reported by Hite (1899). However, few studies were published until the 1970s because of technical difficulties and costs associated with HHP processing units and packaging materials (Galazka and others 2000). Currently these limitations are seen to be surmountable (Rastogi and others 2007). There is significant interest in understanding the effects of high pressure on food and food ingredients (Galazka and Ledward 1998), and further applications of the technology can be anticipated.

HHP treatment is now gradually being adopted by the food industry for the processing and preservation of meat and meat products. Some HHP-treated meat products are already sold in the marketplace: sliced ham and turkey, pork and poultry cuts, thick sliced ham, chicken and turkey products, whole and sliced Serrano ham, salami, and chorizo are available in Spain; natural, minimally processed cooked sliced meat, roasted chicken (whole birds, breasts, and drumsticks), sliced chicken and turkey, chicken sausages, sliced turkey and strips of chicken in modified atmosphere packaging (MAP) and prosciutto (whole and sliced) are sold in the United States. Nitrate-free HHP treated cooked pork products are sold in Japan; HHP treated Parma ham (prosciutto), salami, and pancetta are sold in Italy (PFV 2009).

High pressure, up to 1000 MPa, can affect protein conformation and can lead to its denaturation, aggregation or gelation. The outcome is dependent upon protein susceptibility, the applied pressure and temperature, and the duration of the pressure treatment. HHP treatment of foods can be used to create new texturized products without thermal degradation, or to obtain analogue products with minimal effects on flavor, color, or nutritional value (Vardag and Korner 1995). HHP treatment can also be used as an alternative method to heat treatment for preservation (Gould 1995). High pressure processing of meat has been a fascinating research subject for years due to its potential to inactivate microorganisms and extend shelf-life (Ledward 1998). A large body of research has shown that pressure treatment can also induce changes in meat structure (Carlez and others 1995) and texture (Bouton and Harris 1972; Macfarlane 1973; Bouton and others 1977a, 1977b, 1980; Beilken and others 1990; Suzuki and others 1993; Angsupanich and Ledward 1998; Angsupanich and others 1999; Ueno and others 1999; Jung and others 2000a).

It is notable that the effect of HHP treatment on isolated myofibrils and on whole meat is different. Suzuki and others (1991) investigated the use of high pressure (100 to 300 MPa, 2 to 4 °C, 5 min) on isolated myofibrils and whole meat and found that the changes were completely different in each. At a lower pressure (100 MPa), changes in the sarcomere structure occurred in both myofibrils and the whole meat system; however, at 200 MPa, Z-disks disappeared and dense material aggregated on both sides of the lost M-lines, but only in whole meats.

Of all foods and food constituents, muscle and muscle proteins are probably the most responsive to pressure. This is due to the relatively high sensitivities to pressure of muscle glycolytic processes and of the associations between myofibrillar proteins (Macfarlane 1985).

HHP treatment is currently being used to eliminate pathogenic microorganisms, extend shelf-life, maintain higher sensory quality, and improve the safety of commercial processed (cooked or cured) meat products (PFV 2009). However, HHP treatment can increase lipid oxidation and induce color changes in red meat, which make it have a cooked appearance (Yagiz and others 2009).

In this review, the influence of hydrostatic pressure on meat texture and sensory characteristics as well as the limitations of the technology are discussed.

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Effects of Hydrostatic Pressure Processing on Meat Texture

Fundamental mechanism of texture change induced by high pressure

Under HHP treatment, meat from different animal species undergoes the same mechanism of texture change due to a decrease in volume of proteins (Okamoto and others 1990). The volume of a protein in solution is determined by the constitutive volume of the atoms, by the volume of the internal cavities, and by the solvation of peptide bonds and amino acid side chains (Masson 1992). Upon application of high pressure, the volume of a treated protein decreases because of the compression of the internal cavities (Messens and others 1997). High-pressure effects on proteins are primarily related to the rupture of noncovalent interactions (electrostatic and hydrophobic) within protein molecules (Galazka and Ledward 1995), and to the subsequent re-formation of intra- and inter-molecular bonds within or between protein molecules (Messens and others 1997).

Since the weak linkages stabilizing the secondary, tertiary, and quaternary structures of proteins respond differently to heat and pressure treatments, high-pressure treatment at different temperatures will produce different effects on meat texture (Galazka and Ledward 1998). Pressure denaturation of proteins seems to occur because of the destabilization of noncovalent interactions in tertiary structure (Pittia and others 1996a, 1996b; Tedford and others 1999; Chapleau and others 2004). Treated proteins retain much of their secondary structure; nevertheless, a small degree of unfolding occurs, which exposes hydrophobic regions of the protein. This is assumed to be the cause of protein aggregation (Mozhaev and others 1996; Tedford and others 1999).

Generally, the unfolding of the protein during high-pressure treatment is followed by formation of hydrophobic and disulfide bonded aggregates after pressure release (Funtenberger and others 1997).

During prerigor HHP treatment, intense contraction (a length reduction of 35% to 50%) and severe disruption of muscle structure occurred (Macfarlane 1973; Bouton and others 1977c; Kennick and others 1980). Under the light microscope, HHP treatment was observed to cause Z-disk collapse in adjacent sarcomeres of the myofibril yielding what were termed "contraction bands" (Bouton and others 1977c). Macfarlane and Morton (1978) observed stretching of nearby sarcomeres not involved in the contraction bands using a transmission electron microscope (TEM) to follow changes in ovine *semimembranosus* muscle under HHP treatment (100 MPa, 25 °C, 1 min). Extensive and regular convolution of the sarcolemma of contracted fibres of bovine *supraspinatus* muscle under HHP treatment (103.5 MPa, 35 °C, 2 min) was also observed by Kennick and others (1980). For bovine *longissimus dorsi* muscle under HHP treatment (103.5 MPa, 37 °C, 2 min), Elgasim and Kennick (1982) observed the disappearance of H-zones and M-lines and a degradation of Z-disks. Thus sarcomere structures were dramatically modified by the formation of contraction bands observed under scanning as well as transmission electron microscopy.

During postrigor HHP treatment, extensive modifications in sarcomere structure, but no contraction bands were observed. Macfarlane and Morton (1978) found that the most noticeable effects induced by pressure treatment (100 MPa, 60 min, 25 °C) were the absence of M-lines in the central region of the A-bands and a loss of I-band filament integrity (adjacent to the Z-disks) in ovine *semimembranosus* muscle. On the contrary, although they exhibited a more granular structure, the Z-disks were not extensively altered. In addition, the initial bridges present between thick filaments were no longer observed in transverse sections made in the M-line region. Therefore, it was proposed that a translocation of thin filaments onto the thick filaments occurred, which was used to explain why the H-zone in the centre of the sarcomeres was no longer visible.

Iwasaki and others (2006) investigated the effect of hydrostatic pressure pretreatment on thermal gelation of chicken myofibrils and pork meat patties using penetration tests and observed that pressure treatment of pork and chicken myofibrils improved texture and apparent elasticity of myofibril gels. Based on the results from different electron microscopic observations, they proposed a mechanism of texture change induced by HHP treatment, which included the dissociation of thick and thin filaments of the myofibrils. They indicated that pressure-induced destruction of the Z-line was possibly due to the release of α-actinin. After HHP treatment at 200 MPa, the following 3 phenomena were observed: (i) disappearance of the M-line, (ii) dissociation of each of the thin and thick filaments, and (iii) destruction of the Z-line (chicken myofibril) in the presence of 0.2 M NaCl. Iwasaki and others (2006) suggested that depolymerization of thin filaments was the cause of the high apparent elasticity of a heat-induced myofibrillar gel after 200 MPa HHP treatment. When pressurized at 300 MPa, the decreased apparent elasticity of a myofibrillar gel observed was thought to have been caused by pressure-induced shortening of myosin filaments. This may have been due to depolymerization of thin filaments, which in turn inhibited head-to-head interaction among myosin filaments. Nonetheless, the rheological properties of heat-induced gels were improved by HHP treatment.

Differences of muscle texture variation induced by heat and pressure treatment

High pressure can modify the structure and function of many proteins. For example, isolated myosin from both meat and fish will be denatured by pressure and develop a gel-like structure (Cheftel and Culioli 1997). These structural changes will affect the texture of the muscle, and since the effect of pressure on protein structure is different from the effect caused by thermal denaturation (Mozhaev and others 1996), the textures of heat- and pressure-treated proteins may also be very different. In a heat-treated system, of the weak bonds maintaining the secondary, tertiary, and quaternary structures, it is the hydrogen bonds that are most labile. In a pressure-treated system, it is the hydrophobic and electrostatic interactions which are most vulnerable, hydrogen bonds being mostly unaffected by pressure (Galazka and Ledward 1998).

Okamoto and others (1990) compared textural changes of pressure- and heat-induced gels of egg white and yolk, carp actomyosin, rabbit meat paste as well as soy protein and found that the gels induced by HHP were glossy and soft in comparison with heat-induced gels. They found that both HHP and heat induced denaturation of proteins, whereas the mechanisms involved were different: pressure-induced denaturation of protein was caused by a decrease in protein volume, while heat-induced denaturation of protein was caused by the violent movement of molecules that result in destruction of hydrogen and covalent bonds. Therefore, the mode of denaturation induced by HHP and heat will differ and this may noticeably influence the subsequent textural quality.

During the process of denaturation induced by HHP, the muscle proteins may dissolve or precipitate depending on the pressure used. In the range 100 to 300 MPa, the changes are normally reversible but when the pressures are higher than 300 MPa, the changes are usually irreversible (Rastogi and others 2007). Due to HHP treatment, the rupture of some noncovalent interactions within protein molecules, and the consequent reformation of

intra- and inter-molecular bonds within or between the molecules may occur. Hydrophobic interactions, which are the major forces stabilizing quaternary structure, are very sensitive to pressure (Rastogi and others 2007). No significant changes in tertiary structure were observed when pressure under 200 MPa was used (Rastogi and others 2007). When pressure was greater than 700 MPa, secondary structure changes occurred which led to irreversible denaturation (Balny and Masson 1993).

In the study by Iwasaki and others (2006), scanning electron micrographs were used to clearly illustrate the microstructure of pressure and heat-induced chicken myofibrillar gels. Heat-induced gels were formed from strands, which consisted of bundles of myofibrils, whereas gels from pressurized treatment at 200 MPa before heating consisted of a fine, 3-dimensional network of strands. The latter treatment also increased the apparent elasticities of both chicken myofibrillar gels and pork patties.

Macfarlane (1973) first observed tenderization effects by HHP treatment of prerigor meat. He used HHP (103 MPa, 30 to 35 °C, l to 4 min) to treat various ovine and bovine muscles soon after slaughter, and when cooked, the meat was more tender with both higher moisture content and lower Warner–Bratzler shear values than nonpressurized cooked meat. The tenderizing effect of prerigor HHP treatment has also been confirmed by other researchers (Kennick and others 1980; Riffero and Holmes 1983). However, whether tenderizing of postrigor meat occurs after HHP treatment is complex and basically depends upon HHP conditions used (temperature, pressure, and duration), and this will be discussed in detail later.

Lee and others (2007) showed that both HHP and heat caused denaturation of bovine *semitendinosus* myofibrillar (Mf) proteins; however, the mechanism of HHP-induced denaturation was different from the denaturation induced by heat. They investigated the level of soluble proteins in Mf suspensions caused by HHP in 0.1 M and 0.6 M KCl. It was found that at 0.1 M KCl, with increasing pressure up to 400 MPa, Mf solubility increased and then decreased slightly at 500 MPa. However, the solubility of the Mf protein suspension induced by heat treatment increased with increasing temperature up to 55 °C. At 0.6 M KCl, when pressures were higher than 300 MPa, the solubility of the myosin heavy chain (MHC) and actin in the Mf suspension simultaneously decreased. With increasing temperature, the solubility of MHC gradually decreased; however, the solubility of actin was unchanged until the temperature reached 50 °C. It is believed that meat tenderization is correlated with protein solubility; therefore HHP treatment induces meat tenderization by increasing protein solubility.

From this study it is evident that HHP-treated fresh meat becomes more tender after cooking whereas similar meat without HHP treatment becomes tougher after cooking.

Gelation properties of meat proteins influenced by high pressure

The effects of HHP treatment on isolated protein components of muscle (myosin, actomyosin and actin) have been well reviewed by Macfarlane (1985) and the major effect observed was depolymerisation.

Since high pressure processing has been shown to depolymerize isolated actin and actomyosin and to promote solubilization of other myofibrillar proteins (titin or connectin), changes in gelation properties of these proteins after pressurization may be anticipated (Cheftel and Culioli 1997). Ivanov and others (1960) found that actin was very sensitive to high pressure and underwent depolymerization at 100 MPa. They suggested that under pressure actomyosin could split into actin and myosin. It is well established that HHP treatment results in increased solubility of myofibrillar proteins as a consequence of depolymerisation. Macfarlane (1974) observed that a significant increase (up to threefold) in solubility of sheep myofibrillar proteins occurred when HHP treatment (150 MPa, 0 °C, 5 min) was applied to meat homogenates of ovine *longissimus dorsi* muscle in saline solutions (pH 6.5, 0.5 M NaCl). Further study by Macfarlane and McKenzie (1976) showed that the solubility of myofibrils by HHP was dependent upon temperature (generally being greater at 0 °C than at 30 °C), the nature and the concentration of the salt, as well as the pH. Their results indicated that HHP could enhance solubilization of all the major component proteins of the myofibril. This was further confirmed by Lee and others (2007) as previously discussed regarding tenderization, and solubilization of protein also led to improved gelation at < 400 MPa.

Pressure-induced gelation of meat proteins depends upon the protein system and upon the high pressure processing conditions (HPP) (for example, pressure level, time, and pressurizing temperature) (Jiménez Colmenero 2002). When Yamamoto and others (1992) tested a myosin solution with increased pressure and treatment time, changes began to occur at 140 MPa. It was found that pressure treatment of myosin leads to head-to-head molecular interaction to form oligomers (clumps). Therefore the latter researchers concluded that the pressure treatment most likely did not affect the original helical structure of the tail area of the myosin monomers. They also found loss of protein structure at 300 to 400 MPa, where myosin and actin were both denatured along with many of the sarcoplasmic proteins. At pressures over 400 MPa, myoglobin was irreversibly denatured.

The nature and the concentration of any salts present also affect the gelation of meat. Yamamoto and others (2002) observed that HHP (200 to 300 MPa) induced gelation of chicken myofibrils (40 mg/mL) in 0.1 to 0.2 M NaCl and that the gel strength increased with these pressures at 0.1 M NaCl, whereas it remained almost the same at 0.2 M NaCl.

Temperature influences the outcome of pressure treatment, but the effects of each are interconnected. Pressurization can either accentuate or reduce the effect of temperature on protein structure and the mechanism of protein denaturation differs with the pressure/temperature combinations used (Messens and others 1997). Cooking meat generally increases toughness. HHP treatment can either enhance or reduce this effect based on the HHP conditions applied. Usually at relatively lower pressures (100 to 300 MPa), tenderizing of fresh meat occurred after cooking (Macfarlane 1973; Kennick and others 1980; Riffero and Holmes 1983). However, at higher pressure (>300 MPa), fresh meat became significantly tougher than unpressurized samples (Jung and others 2000a; Ma and Ledward 2004). Studies conducted on muscle protein gelation processes have been classified according to meat system conditions (pre- or postrigor, raw, or preheated), as well as pressure/temperature combinations applied, and have been thoroughly reviewed by Jiménez Colmenero (2002).

In spite of the potential for fat to play a role in the gelation of meat protein, little research has been done to study how fat may influence the texture of meat products following HHP. Carballo and others (1996) investigated the effect of HHP (100 and 300 MPa, 5 and 20 min, at about 8 °C) on the texture of uncooked and cooked low fat (6%) and high fat (23%) meat batters by penetration testing. Their results indicated that pressure caused a significant increase in the mechanical resistance of uncooked batters. However, HHP treatment (prior to heating) did not enhance thermal gelation ability of meat batters. Elasticity of the meat matrix was decreased in high fat cooked batters pressurized at 300 MPa. Carballo and others (1997) also studied the effect of HHP (100 and 300 MPa, 5 or

20 min) on characteristics such as texture, microstructure of low-fat (9.2%) and high-fat (20.3%) beef patties. With HHP treatment, compared to high-fat patties, the low-fat product exhibited significantly higher Kramer shear force and Kramer energy. Thus high fat level has the effects of reducing elasticity and increasing penetration force (toughness) of meat batters after HHP treatment.

In general, HHP treatment increases the solubility of myofibrillar protein and causes depolymerization, therefore improving gelation of fresh meat and results in desirable tenderization; however, pressure should be applied at relatively low levels to avoid toughness. Meanwhile, effects of temperature, salt concentration and fat level can also influence HHP effects on meat gelation properties.

Toughness and tenderness of meat is influenced by high pressure

Meat tenderness has been resolved into at least 2 different components, "actomyosin toughness" and "background toughness" (Locker 1960; Marsh and Leet 1966; Marsh 1972). Actomyosin toughness is the toughness attributed to the myofibrillar proteins, whereas background toughness is due to the presence of connective tissue and other stromal proteins (Ueno and others 1999).

There is agreement among consumers that tenderness is the most important factor of all the attributes that characterize the eating quality of meat (Jung and others 2000a; Denoyellea and Lebihan 2003). High pressure treatment is a relative new technique used to tenderize meat. To date there have been a large number of papers published on accelerated tenderization of meat during conditioning (ageing) which results from structural changes of the myofibrils caused by high pressure (Macfarlane 1973; 1985; Bouton and others 1977b; Kennick and others 1980; Riffero and Holmes 1983; Locker and Wild 1984; Suzuki and others 1990, 1992; Cheftel and Culioli 1997).

Of significance from studies investigating muscle texture changes in meat, poultry and fish during HHP treatment (Macfarlane 1973; Bouton and others 1980; Beilken and others 1990; Angsupanich and Ledward 1998; Angsupanich and others 1999; Jung and others 2000b; Chevalier and others 2001; Iwasaki and others 2006) are observations that high pressure can tenderize meat when applied prerigor, but that at low temperature it had no measurable beneficial effect on postrigor meat. Indeed, some results indicated that HHP treatment alone caused meat hardening or toughening (Macfarlane and others 1980-81; Yuste and others 1998; Jung and others 2000a, 2000b).

Additional understanding of HHP effects on tenderization of meat have come from studies on its effects upon connective tissue in conjunction with thermal treatments. Ratcliff and others (1977) showed that although pressure-heat treatment effectively eliminated myofibrillar toughness, the tenderness of treated samples was limited by the connective tissue or background toughness. Macfarlane and others (1980-81) also found that although a transition peak in the thermogram of pressurized muscle normally attributed to F-actin was absent, the connective tissue transition peak was unchanged. Beilken and others (1990) found that pressure treatment at temperatures ranging from 40 to 80 °C had little or no effect on the background toughness other than to raise the temperature at which heat treatment alone produced a decrease in this toughness. In another report, Suzuki and others (1993) found no significant differences in the ultrastructure, thermal solubility, or thermograms of isolated intramuscular collagen of control (untreated) and pressurized muscles. These observations were significant in explaining the limited effectiveness of HHP on postrigor meat because Nishimura and others (1995, 1996) found that the weakening of the intra-muscular connective tissue caused during normal extended ageing correlated with meat tenderization.

Gekko and Koga (1983) established that myofibrillar proteins as well as connective tissue protein, collagen, are the components of muscle controlling toughness. As collagen is primarily stabilized by hydrogen bonds, it is little affected by pressure, whereas changes in the structure of contractile myofibrillar proteins are thought to be primarily responsible for the changes in desirable textural properties observed when meat is subjected to high pressure. Differential scanning calorimetry (DSC) has been widely used to relate the denaturation of individual muscle proteins to the textural changes in meat caused by cooking (Martens and others 1982; Findlay and others 1986) and pressurization (Angsupanich and Ledward 1998; Angsupanich and others 1999). The 3 major endothermic transitions seen in beef muscle, attributed to myosin, collagen, and actin, have been associated with specific changes in beef texture (Angsupanich and Ledward 1998). High pressure treatment at different temperatures will induce different effects on meat texture since the weak linkages stabilizing the secondary, tertiary, and quaternary structures of a protein respond differently to heat and pressure (Galazka and Ledward 1998).

Bouton and others (1977b) found that a pressure of about 100 MPa applied for 2.5 min or longer to postrigor muscle at 40 to 60 °C improved the tenderness of the meat and Beilken and others (1990) reported that pressure treatment at 150 MPa and 40 to 80 °C prevented the development of myofibrillar toughness, but had little or no effect on the connective tissue component of toughness.

Most work published indicates increased meat tenderness as a consequence of high pressure- induced modifications of the myofibrillar structure when heat treatment was used. However, Jung and others (2000a) reported the opposite effect at 10 °C when HHP treatments were done at 130 and 520 MPa for 260s. At this lower temperature, the greater integrity of myofibrils appeared to be responsible for reduced effects rather than the connective tissue component. Thus, myofibrillar proteins appeared capable of increasing toughness and/or neutralizing the effect of HHP on tenderness in the absence of heat treatment. Other parameters such as sarcomere contraction or cooking loss, of course, can be factors influencing the final texture and tenderness of meat.

Ma and Ledward (2004) found that the toughness of beef muscle increased with increasing pressure (200 to 800 MP) at constant temperatures of 20 to 40 °C, with further increases at increased temperature and ambient pressure. However, consistent with other reports, tenderness was increased significantly with application of 200 MPa pressure at 60 and 70 °C. Under the conditions of these tests, they speculated that accelerated proteolysis rather than structural changes in protein was likely to be the major factor contributing to the loss in toughness observed.

In summary, HHP effects on meat toughness or tenderness are dependent upon rigor stage, pressure, temperature and their combination. Usually low pressure (< 200 MPa) treatment can tenderize prerigor meat, whereas tenderization postrigor with HHP can only be achieved at higher temperature (40 to 80 °C).

Springiness, chewiness, and juiciness properties of meat as influenced by high pressure

Except for juiciness which is evaluated by sensory panels, both springiness and chewiness are tested by texture profile analysis (TPA, a typical penetration test), to determine muscle protein gel textural characteristics. Fernandez and others (1998) reported that pressurization of chicken batters at 200 MPa resulted in products with increased hardness and chewiness compared to nonpressure treated samples, although at 400 MPa chicken batters had

structures which were coarser, more irregular, less compact, and aggregated. These samples had decreased springiness, cohesiveness, and chewiness. This phenomenon could be explained by high pressure protection of meat proteins from heat denaturation to some extent (Fernández-Martín and others 1997). Macfarlane (1985) and Suzuki and others (2006) reported that pressure treatment improves the cohesion between meat particles in reformed meat or fish-type products.

Angsupanich and Ledward (1998) found that pressure treatment of cold water fish yields effects that are similar to these seen with red meats. Beneficial effects on texture were mainly observed at pressures lower than 400 MPa. They found that the characteristic pressure-treated texture was different from that seen in either raw or cooked fish, being harder, chewier, and gummier than the cooked product.

Pérez-Mateos and Montero (2000) showed that high-pressure (200 to 420 MPa, 10 to 30 min) treatment of fish (blue whiting) yielded gels having lower cohesiveness and higher elasticity than heat-induced gels. A combination of pressure, temperature and time (200 MPa/<10 °C/10 min and 375 MPa/37 °C/20 min) produced more elastic gels, whereas gels made under high pressure at chilled temperature (<10 °C) were much harder, more deformable, and more cohesive. High pressure appears to have application in the formation of protein gels at low (0.2 M) KCl levels with fish and a number of other animal species (Suzuki and others 2006).

Crehan and others (2000) investigated the application of 150 and 300 MPa pressure on frankfurter quality at 1.5% and 2.5% NaCl. Among the combination of different pressure/salt levels used, the 300 MPa/1.5% (pressure/NaCl) treatment appeared to be the best because juiciness, hardness, springiness, cohesiveness, gumminess, and chewiness scores were the highest. Their results suggested that high pressure (300 MPa) treatment can be used to improve the sensory properties of frankfurters processed with lower salt levels (1.5%).

In a study by Mor-Mur and Yuste (2003), it was shown that high pressure-processed (500 MPa, 5 or 15 min, 65 °C) sausages were less firm, more cohesive, had lower weight loss, and higher sensory panel preference scores compared to those treated at higher temperatures (40 min at 80 to 85 °C) without pressure.

High pressure treatment of prerigor meat

HHP treatment can result in reversible or irreversible structure changes in meat depending on the pressure level used. It also seems that the extent of structural damage of meat proteins is greater when the period between animal slaughter and HHP processing is shorter than that needed for development of muscle stiffness through rigor onset. While HHP treatment of prerigor fresh meat resulted in shortened muscle (which usually means tougher meat), its structure was severely damaged (Pandrangi and Balasubramaniam 2005). Macfarlane (1973) was the first to propose the use of high pressure for tenderization of prerigor meat. In this study, when different ovine and bovine muscles were treated with high pressure (103 MPa), very firm and compact raw meat resulted; however, meat was more tender and had higher moisture content than nonpressurized meat cut from carcasses prerigor (hot-boned), postrigor, or after cooking. The more tender high pressure-treated meat was obviously less juicy than untreated hot-boned meat even though the former contained more moisture.

Macfarlane and others (1982) used a pressure vessel with a window to observe a strip of muscle maintained under tension (90 to 160g/cm^2) and found there was an instantaneous severe contraction upon pressure application, but the muscle strip then became extended. The authors assumed that extension was due to pressure disruption of myofibrillar proteins and they concluded that tenderization improvements noted resulted from these effects.

Bouton and others (1977b) also restrained muscle tissue during pressure treatment and consequently decreased contraction from 39% to 15%. They also obtained a reduction in shear values of 62% versus 80% compared to that in muscle left free to contract. When correlated with changes in tenderness, results indicated that the tenderizing effect of pressure upon prerigor meat was directly related to the degree of contraction induced.

The initial observations by Macfarlane (1973) on tenderizing effects by pressurization of prerigor meat have been confirmed by other researchers (Kennick and others 1980; Schumann and others 1982; Riffero and Holmes 1983). Therefore, it appears that meat tenderization by HHP is facilitated when applied to muscle tissue still able to contract before glycogen exhaustion at rigor.

High pressure treatment of postrigor meat

Bouton and others (1977a) found that pressure application (100 MPa for 1 min or more) at low temperature (<30 °C) did not show any beneficial effects on postrigor beef tenderness. Ma and Ledward (2004) found that when heated at ambient pressure meat expectedly became tougher at higher temperature, and that pressure alone did not improve the tenderness, gumminess or chewiness of postrigor beef *longissimus dorsi*. Although some structural damage to myofibrillar proteins occurred in heated (40 to 70 °C) and pressure (200 to 800 MPa) treatment, these effects and improved tenderness noted were attributed to accelerated proteolytic activity during warming come-up times in the pressure cell. Nevertheless, high pressure treatment at 30 °C was shown to have positive effects on the texture of cold-shortened meat, partly counteracting myofibrillar toughness. This improvement was limited and required 4 h or more pressure treatment (Macfarlane and McKenzie 1986).

Some proteins are quite sensitive to low pressure (<100 to 200 MPa). At these pressures a reversible dissociation of subunits or a partial unfolding occurs (pressure dissociated native proteins of the myofibril can re-associate upon pressure release). When pressure was higher than 200 MPa, changes were usually irreversible and protein denaturation occurred with unfolding of monomeric proteins or aggregate formation (Chapleau and de Lamballerie-Anton 2003). Since low pressure-induced modifications appeared reversible, Bouton and others (1977b) recommended combining pressure with heat so that texture modifications became irreversible. Some researchers (Bouton and others 1977a, 1978, 1982; Ratcliff and others 1977) concluded that the tenderization effect was due to modifications of only myofibrillar structure and not that of connective tissue as observed earlier; this reflects the fact that pressure does not cause the disruption of hydrogen bonds that are responsible for maintaining the helical structure of collagen.

When postrigor meat was briefly treated by pressure of 300 to ≥500 MPa (5 min), meat tenderization could be achieved without any additional heating. According to Suzuki and others (1990, 1992) cold pressurization over 150 MPa had a clear effect on beef tenderness. Myofibril fragmentation was obviously increased, gap filament integrity was reduced, and ultrastructure was significantly modified. As reported by others, the influence of high pressure (100 to 300 MPa) on the physicochemical properties and ultrastructure of beef intramuscular collagen fibrils, examined immediately after pressurization, was very limited (Suzuki and others 1993). Notwithstanding the effects on tenderness reported previously for postrigor meat, overwhelming evidence suggests that HHP improvements in tenderness are only obtained following treatment of prerigor meat (Rastogi and others 2007).

The results of Jung and others (2000a) showed that HHP treatment (130 and 520 MPa, 10 °C, 260 s) significantly increased toughness of both raw and cooked (1 h, 65 °C) postrigor bovine meat at the higher pressure. This result conflicts with those of earlier researchers and may be explained by the difference in HHP treatment conditions used. It is still unclear whether the effect of HHP on tenderness is influenced by interactions between the myofibrillar and connective tissue proteins of postrigor meat (Jung and others 2000b).

Sikes and others (2009) investigated effects of HHP on texture of low-salt beef sausage batters and found that at all salt concentrations (0% to 2%), the hardness and gumminess of pressure-treated (up to 400 MPa, 10 °C, 2 min) samples were higher compared to those untreated. There was greater acceptability in terms of both appearance and texture of HHP-treated low salt sausages in comparison with unpressurized samples. It was shown by SDS-PAGE that pressure caused protein solubilization and partial unfolding, which increased binding and gelation.

HHP Limitations and Future Applications

The greatest limitations for the application of HHP technology for meat texture enhancement have been the substantial capital equipment cost and need for its application to hot-boned (prerigor) meat for consistent benefit. An additional problem is that current batch sizes are a restriction for operations with high line speeds.

It would be valuable if conditions could be established that would optimize both antimicrobial effects of HHP treatment (Cheftel and Culioli 1997; Torres and Velazquez 2005) and its beneficial texture effects simultaneously. In addition, it appears that undesirable meat color changes (browning) when used in conjunction with thermal treatment are a limitation. Although pressurization of frozen meat is being studied to avoid color changes, the use of HHP treatment of frozen hot-boned meat to prevent shortening caused by thaw-rigor and to enhance overall tenderness would appear to be an area worthy of study.

Since HHP has value in the formation of gels from myofibrillar protein at low (0.2 M) salt concentration, HHP may have potential for the development of low sodium-containing processed meat products. Current technological limitations of 1.7% to 2.1% (w/v) NaCl may be overcome by the use of HHP and enable manufacture of these products with more healthful levels of NaCl.

It is of interest that HHP has seen recent commercial success as an antimicrobial treatment for vacuum packaged processed meats to reduce the risk of *Listeria* contamination.

Conclusions

HHP treatment causes protein denaturation, aggregation, or gelation which can result in meat becoming either tenderized or toughened, depending on the meat protein system, the temperature, the pressure, and its duration. Juiciness, springiness, and chewiness are increased upon HHP treatment. However, HHP has little effect on the toughness of connective tissue. Process conditions must be carefully controlled to enhance tenderizing effects in meat and gelation of fish muscle. Prerigor HHP treatment tenderizes meat, whereas tenderizing effects of postrigor HHP treatment are only measureable if pressure and heat treatment are combined. HHP treatment is not only a promising technology because of its tenderizing effects on meat, but also because of its potential to inactivate microorganisms and extend the shelf life of meat and meat products.

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